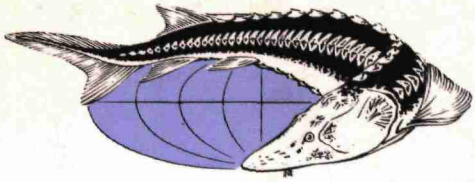
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Acipenser



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BORDEAUX 3-6 OCTOBRE 1989

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PREFACE

Until the late 1970s, there had been few scientific studies in the West on sturgeon. The available literature was almost all in Russian. There was practically no up-to-date information on sturgeon in the Far East.
The collected data at that time revealed a number of disquieting facts: almost all sturgeon populations were in decline; this was attributable to human activity; our knowledge of sturgeon biology was generally inadequate.
In the field of fish farming the only data were those arising from the colossal Soviet restocking programme carried out in the Caspian Sea and the Sea of Azov.
In several countries the early 1980s saw increased concern for endangered species, sports fishermen became more and more interested in sturgeon, and there was a drive in the aquaculture sector towards diversification.
So it came about that a symposium was held at Milwaukee, USA in 1983 to establish the state of our knowledge of North-American sturgeon.
It was in the early 1980s also that CEMAGREF Bordeaux, based on the edge of the last remaining Western European basin with stocks of *Acipenser sturio*, started to develop its own research programme.
After making study trips to the USSR, Hongary, the USA and Canada, scientists from CEMAGREF team decided to stage an international symposium. The ideal opportunity to do so arose when the European Aquaculture Society chose Bordeaux as the venue for its Aquaculture Europe 89 conference. The speed and depth of the response to the call for papers confirmed their belief that the time ws ripe for a symposium of this kind.
Thus it was that ACIPENSER, the 1st International Sturgeon Symposium, was held at the Bordeaux Conference Centre from 3-6 October 1989. The publication now of the proceedings has been undertaken in recognition of the contribution made by all those who took part in the symposium. Almost all the papers delivered at the symposium have been included here, along with five that could not given at the time and the texts of five posters, three of them recast as papers.
Two further articles have been added: one describing a number of biological and ecological characteristics of wild sturgeon, and the other setting out the available data on sturgeon and caviar production.
The member of the team who has undertaken the onerous task of ordering and editing the papers published here is P.Williot. He has organized them into four chapters, dealing respectively with biology, populations in the wild, farming and other related topics. Two indexes has been included to make the volume easier to consult: one thematic and the other organized by species.
It is our hope that both the symposium itself and the publication of the proceedings will stimulate further research and that the international literature on sturgeon will grow accordingly.

The Organizing Committee
December 1990

PREFACE

Jusque vers la fin des années 70, les esturgeons avaient fait l'objet de peu d'écrits scientifiques dans les pays occidentaux. La littérature disponible était essentiellement en langue russe. Pratiquement aucune donnée récente n'existait sur les esturgeons d'extrême-Orient.
Les informations recueillies à l'époque amenaient à quelques constatations inquiétantes: presque toutes les populations d'esturgeons étaient en régression; les causes en étaient les activités humaines; les connaissances biologiques étaient le plus souvent insuffisantes. Dans le domaine de l'exploitation par pêche, la seule référence était le colossal programme de repeuplement entrepris en Mer Caspienne et Mer d'Azov par les soviétiques.
Au début des années 80 l'intérêt pour les espèces en voie de disparition commence à se manifester dans certains pays, les pêcheurs sportifs s'interessaient de plus en plus aux esturgeons et la production aquacole cherchait à se diversifier.
Ainsi en 1983 eut lieu à Milwaukee aux USA un symposium qui fit le point sur les connaissances des esturgeons nord-américains.
C'est également dès le début des années 80 que le CEMAGREF de Bordeaux, situé à proximité du dernier bassin d'Europe occidentale fréquenté par Acipenser sturio, a commencé à développer un programme d'étude sur l'esturgeon.
A la suite de missions en URSS, Hongrie, aux USA et au Canada, les scientifiques de cet Institut envisagèrent d'organiser un colloque international. L'occasion se présenta quand la European Aquaculture Society choisit Bordeaux pour tenir son congrès Aquaculture Europe 89. Très vite le succès de l'appel à communication confirmait l'opportunité de la manifestation.
C'est ainsi qu' ACIPENSER, 1er colloque international sur les esturgeons, s'est tenu du 3 au 6 octobre 1989 au palais des congrès de Bordeaux. Aujourd'hui la publication des actes est la juste récompense de tous ceux qui ont participé à ce colloque. Ce document regroupe la quasi-totalité des textes.
Cinq communications qui n'avaient pas pu être présentées oralement ainsi que les textes de cinq posters dont trois transformés en communication sont également incluses.
Enfin ont été ajoutés deux articles, l'un présente quelques caractéristiques biologiques et écologiques des esturgeons sauvages, l'autre donne les éléments disponibles en matière de production d'esturgeon et de caviar.
Parmi nous c'est P.Williot qui a assumé la lourde charge de mise en forme de ce recueil.
La présentation est faite selon quatre chapitres: biologie/ populations sauvages/ élevage/ autres thèmes. Deux index, l'un thématique, l'autre par espèce, facilitent la lecture.
Souhaitons que ce colloque et ces actes aient une suite, et qu'ainsi, la littérature internationale sur les esturgeons ne cesse de s'enrichir.

Le comité d'organisation
Décembre 1990

A la mémoire
du Professeur Etienne MAGNIN
qui, par ses nombreux travaux, a fait mieux
connaître les populations d'esturgeon
de France puis du Canada.

BIOLOGIE

P.Williot, Ed. ACIPENSER, CEMAGREF Publ., 1991,

ENERGY UTILIZATION BY FARMED SIBERIAN STURGEON (*Acipenser baeri*) FROM 3 AGE CLASSES

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RESUME

Trois expériences ont été réalisées afin d'étudier les différentes étapes de l'utilisation de l'énergie chez des esturgeons sibériens élevés en pisciculture à 18°C et agés de 3 mois (PM : 40g), 10 mois (PM : 230g) et 24 mois (PM : 1500g). L'efficacité d'utilisation de l'énergie d'origine alimentaire a été appréciée au cours d'expériences de croissance durant lesquelles les poissons ont été nourris avec de l'aliment commercial pour truite (Protéines brutes : 49,5 à 50,4% MS ; Energie brute : 21,3 à 21,5 kJ/g MS ; Lipides : 11% MS). L'ingestion volontaire de nourriture a fortement diminué avec l'âge des poissons, passant de 1,9% PM/jour chez les esturgeons de 3 mois à 0,5% PM/jour chez ceux de 24 mois. L'efficacité de rétention énergétique des esturgeons les plus âgés a été la meilleure ; chez ces animaux, l'énergie a été retenue à 55% sous forme de lipides alors que le gain en énergie sous la forme protéique prédominait chez les esturgeons âgés de 3 et 10 mois. Les résultats de l'étude de la digestibilité des composants d'un aliment commercial pour truite ont montré que l'esturgeon sibérien digérait bien les lipides mais la valeur de l'ADC des protéines était faible. Deux méthodes ont été utilisées pour évaluer les pertes endogènes d'énergie : l'analyse de composition corporelle avant et après une période de jeûne ou des mesures de calorimétrie indirecte pendant le jeûne. En raison des différences importantes entre les valeurs issues de l'une ou l'autre des méthodes, le taux métabolique d'entretien de l'esturgeon sibérien n'a pu être précisement établi. Par contre, quelque soit la méthode d'évaluation, les pertes d'azote endogène (60 mg/kg/j) de l'esturgeon sibérien ont été plus faibles que celles de la truite à la même température (18°C). En outre, les réserves corporelles lipidiques ont été la principale source d'énergie chez l'esturgeon à jeun.

Mots clés : Nutrition énergétique, utilisation de l'énergie, digestibilité, dépense énergétique d'entretien, *Acipenser baeri.*

ABSTRACT

Three experiments were conducted to study energy utilization by farmed Siberian sturgeons from 3 age classes (3, 10 and 24 month old with an average body weight of 40 g, 230 g and 1500 g respectively). Efficiency of dietary energy utilization was studied through growth trials performed with commercial trout diets (Crude Protein : 49.5 to 50.4% DM ; Gross Energy : 21.3 to 21.5 kJ/g DM ; Crude Fat : 11% DM). Voluntary feed intake decreased with age from 1.9% MBW/day in the 3 month old sturgeon to 0.5% MBW/day in the 24 month old one. Energy retention efficiency (ERE) was better in the oldest fish in which 55% of RE was in the form of fat, while energy gain in the form of protein predominated in the 3 and 10 month old sturgeons. Data on digestibility of the components of a commercial trout diet showed that siberian sturgeon digested fat very well but ADC value of protein was low. Two methods were used to evaluate endogenous energy losses : body composition analyses on the first and the last day of a fasting period, or indirect calorimetric measurements during the fast. Owing to the discrepancy between the values obtained from the two methods, the value of maintenance energy expenditure could not be exactly known. Irrespective of the method, concordance was however found in the estimation of endogenous nitrogen loss of Siberian sturgeon, which was found to be 60 mg/kgBW/d, a value lower than in trout at the same temperature (18°C). Body fats were the main energy source in fasted Siberian sturgeon.

Key-words : Energy utilization, voluntary feed intake, digestibility, maintenance energy expenditure, *Acipenser baeri.*

INTRODUCTION

Siberian sturgeon, introduced into France as a model to study the biology of chondrostean fish, is now a candidate for commercial production. Successful fish culture depends upon the supply of diets containing an appropriate balance of nutrients and adequate energy to provide the needs and to permit the most efficient growth of the organism. Before the feed components can be used by the organism, they must be digested and absorbed from the digestive tract, the non-digested part being lost as faeces. Not all the digestible energy is available for fish growth. A portion is spent for maintenance requirements, an other portion is used for motor activity. Digestion and absorption processes, transformation and interconversion of nutrients, formation and excretion of metabolic wastes and anabolic processes leading to new tissues synthesis also consume energy. In fish, as in any other animals, energy for maintenance, activity and production is supplied by the oxidation of dietary or endogenous energy-yielding substrates : protein, carbohydrate and fat, which can be used both as energy fuels and as new components of tissues. So, growth and composition of body gain are closely related to energy expenditure and utilization of energy-yielding nutrients.

During the last years, nutritional bioenergetics of commonly farmed fish such as Salmonids has received great attention (Cho et al., 1982 ; Cho and Kaushik, 1985 ; Kaushik and Oliva-Teles, 1985) but data on such a novel species as sturgeon are scarse. Studies of nutrient utilization by Siberian sturgeons were made during ontogenesis (Dabrowski and Kaushik, 1986) and larval stages (Dabrowski et al., 1987 ; Fauconneau et al., 1986). Rearing of larval Siberian sturgeon is now successfully practiced. Early feeding trials (Barrucand et al., 1979) showed that juveniles and adults of sturgeons could be reared with artificial diet. However, production would be undoubtly improved by specific practical diets, which require precise knowledge on the nutrition of this species. Most of the informations on farmed juvenile sturgeon refer to white sturgeon *(Acipenser transmontanus)* which is farmed in North America. In the present study, experiments were conducted to obtain basic data on nutritional energetics of juvenile farmed Siberian sturgeons. Four main points were considered : necessary energy expenditure to sustain life processes, energy intake, digestibility, energy deposition. As the animal's energy needs are known to be affected by the stage of life, the effect of age of sturgeon on utilization of energy was also examined.

MATERIAL AND METHODS

The experiments were performed with Siberian sturgeons (*Acipenser baeri*) produced in the Cemagref hatchery of INRA fish farm at Donzacq (Landes, France). All sturgeons were reared in sand bottomed concrete tanks with flowing spring water at a constant temperature of 18°C and fed commercial trout pellets.

The trials were conducted with Siberian sturgeons from 3 age classes (3, 10 and 24 month old). The ability of sturgeons to utilize dietary energy was studied through growth trials with commercial trout diet (Crude Protein : 49.5 to 50.4% of Dry Matter ; Gross Energy : 21.3 to 21.5 kJ/g DM ; Fats : 11% of Dry Matter). Fish were always fasted 36 hours before the weighings. Maintenance expenditure of fish was evaluated using two different methods : the first one was based on endogenous losses during a 4 or 5 week fast estimated by body composition analysis, the second method was based on endogenous losses estimated by indirect calorimetric measurements. All experiments were conducted in sand bottomed concrete tanks except the indirect calorimetry trial.

In *Experiment 1*, four batches of 100 three month old fish each were alloted to different tanks. Two groups (IBW : 31g) were fasted for 4 weeks. Two others groups (IBW : 48g) were fed *ad libitum* twice a day a commercial trout diet (CP : 50.4% DM ; GE : 21.5 kJ/g DM) for 8 weeks. Feed intakes were recorded. At the beginning and at the end of the trials, all sturgeons were weighed and ten fish from each batch were sampled and frozen until analyses. After the growth trial, a digestibility trial was performed with the same groups. 1% of chromic oxide was added to the commercial

trout diet powder before pelleting. The two batches of sturgeons were fed *ad libitum* twice a day for 10 days. Then, the faeces were collected by manual stripping from twenty fish in each batch, frozen and freeze dried.

Experiment 2 was conducted with ten month old fish. Two groups of 35 sturgeons (IBW : 235g) were fed *ad libitum* twice a day a commercial trout diet (CP : 49.5% DM ; GE : 21.3 kJ/g DM) for 6 weeks. Feed intakes and growth characteristics were recorded and fish sampled as described above. Two other batches of 12 ten month old sturgeons each (IBW : 230g) were put in round 200 l tanks with flowing water (Temperature : 18°C). Fish were accustomed to these experimental facilities for 2 weeks and then fasted. From the eleventh fasting day onward, inflowing and outflowing water from each tank were sampled every hour for 72 hours in succession. Oxygen, carbon dioxide and ammonia in water samples were analysed immediatly after sampling. Water flow rate was also measured each hour. Fish were individually weighed at the end of the measurements.

Another trial (*Experiment 3*) was performed with two groups of 25 individually identified twenty-four month old sturgeons, over 35 days. One group (IBW : 1300g) was fasted, the other one (IBW : 1700g) was fed *ad libitum* twice a day a commercial trout diet (CP : 49.8% DM ; GE : 21.4 kJ/g DM). Growth performance was followed on every individual sturgeon. Four fish were removed for analyses from the initial batch and from each group at the end of the trials.

Analyses : Frozen whole body samples and faeces were freeze dried before analyses. Proximate compositions of the diets, faeces and whole body samples were analysed following usual laboratory procedures : dry matter (110°C, 24h.), crude protein (Kjeldalh nitrogen x 6.25) after acid digestion, crude fat (Chloroform : methanol extraction), gross energy (Gallenkamp adiabatic bomb calorimeter). Chromic oxide in the diet and faeces was measured using a semi-automated procedure (Mathieson, 1970) after perchloric acid digestion (Bolin et al., 1952). Oxygen and carbon dioxide analyses were made in water samples using respectively an oxygen electrode (97-08 Orion, USA) and an ion selective electrode (95-02 Orion, USA). Ammonia nitrogen contents were measured with an auto-analyser (Technicon) following the method described by Kaushik (1980).

Energy and nutrient retention efficiencies were calculated as :

$$R\ (\%) = 100 \times (FBW \times FBC) - (IBW \times IBC) / (TFI \times DC)$$

where IBW and FBW represented initial and final body weights, IBC and FBC initial and final body composition (dry matter, protein, lipid or energy), TFI the total feed intake and DC the dietary components.

Based on diet and faeces composition, apparent digestibility coefficients of dietary components were also calculated :

$$ADC = 100 - \left(100 \times \frac{\%\ Cr_2O_3\ Diet}{\%\ Cr_2O_3\ Faeces} \times \frac{\%\ C\ Faeces}{\%\ C\ Diet}\right)$$

where C represented protein, fat or energy.

Daily endogenous energy losses based on body composition analyses (Experiments 1 and 3), were estimated as :

$$EL\ (kJ) = (FBW \times FBE) - (IBW \times IBE) / (MBW \times Nbr\ days)$$

where IBE and FBE were initial and final body energy content and MBW = (IBW+FBW)/2.

Daily dry matter losses were calculated in the same way.

The O_2 uptake, CO_2 output and nitrogen excretion rates were calculated taking into account the total weight of fish group, the volume of water in the tank and the water flow rate. Indirect calorimetry calculations were based on a standard composition of the three substrates for oxidation : protein (P), carbohydrates (C) and fat (F) (Gnaiger, 1983). The respiratory quotient (moles CO_2/moles O_2) was determined from the data on respiratory gas exchanges. According to Kaushik (1980), the assumption was made that only 85% of the protein nitrogen was excreted as ammonia. The nitrogen quotient (moles N/moles O_2) and the amount of oxidized protein were calculated from the total nitrogen excretion. The daily energy expenditure (H) of fasted sturgeon and the amounts of oxidized body stores for energy purposes were evaluated using the formulae given by Van Waversveld et al. (1988) :

H (kJ) = 0.356 O2 + 0.116 CO2 - 1.863 P
C (g) = -0.067 O2 + 0.095 C02 - 1.086 P
F (g) = 0.038 O2 - 0.038 CO2 - 0.073 P

where O_2 and CO_2 were expressed in mmoles and P in g.

The statistical analyses (ANOVA followed by Duncan's test) were performed using the SAS Package (SAS Inst Inc. North Carolina, USA).

RESULTS

Growth performances and dietary energy utilization

As can be seen in table I, voluntary daily feed intake of Siberian sturgeons decreased with age. When 3 month old fish ingested nearly 2 percent of their body weight by day, feed intake of 24 month old sturgeons was only 0.5 percent of body weight. Specific growth rate of sturgeons also decreased with age. Feed/gain ratio (FGR) was similar in 3 and 10 month old fish but it was higher in the oldest sturgeons. However, energy retention efficiency was the best in the 24 month old sturgeons.

Table 1 - Growth performances and dietary energy utilization of Siberian sturgeon fed commercial diets at a water temperature of 18°C

Age (months)	3	10	24
Characteristics of the diets			
Gross energy (kJ/g DM)	21.50	21.33	21.45
Protein/Energy ratio[1]	23.45	23.20	23.25
Growth performances			
Initial Body Weight (g)	48^c	235^b	1700^a
Daily feed intake[2]	1.90^a	1.20^b	0.50^c
Specific growth rate (%/day)	1.54^a	1.03^b	0.31^c
Feed/Gain ratio	1.20^b	1.18^b	1.60^a
Energy retention efficiency (% of gross intake)	20.17^b	20.83^b	26.5^a

[1] mg crude protein/kJ gross energy
[2] calculated as percent of mean body weight
On each row, means with the same letter are not significantly different ($p<0.05$)

Daily energy gain (Fig.1) of young sturgeons was much higher than that of 24 month old sturgeons, due to the amount of ingested energy. Total energy gain was

respectively 76.7, 48.0 and 25.5 kJ/kg/day for the 3, 10 and 24 old month fish. On the other hand, though the gross energy and the P/E ratio of the 3 commercial diets were similar, partition of retained energy between protein and fat changed with age of fish. As it is represented on figure 1, energy gain in the form of protein predominated in the 3 and 10 month old sturgeons while in 24 month old sturgeon 55 percent of retained energy was in the form of lipid.

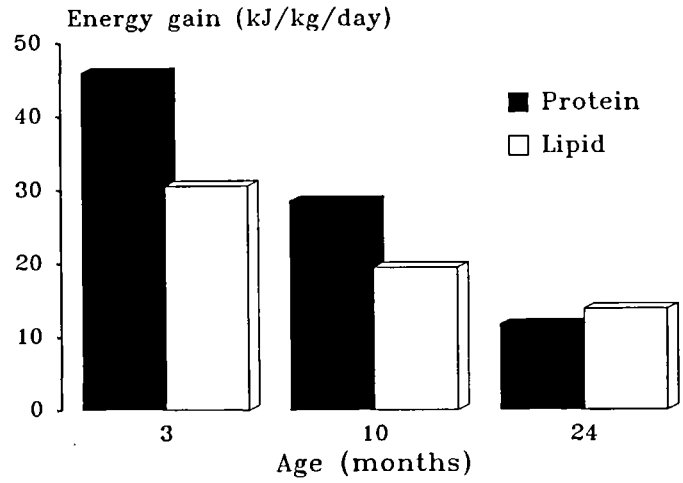


Figure 1 - Composition of relative energy body gain

Whole body proximate composition

Wet body compositions of 3, 10 or 24 month old Siberian sturgeons are shown on figure 2.

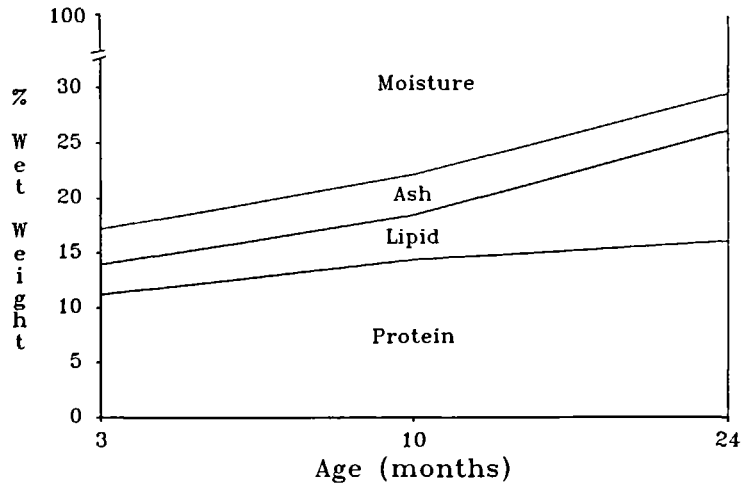


Figure 2 - Wet whole body composition of farmed Siberian sturgeons

Percent moisture significantly decreased with age, while protein and lipid in the wet samples increased. The amounts of lipids were two fold higher in 10 month old sturgeons than in 3 month old ones and five fold higher in the 24 month old sturgeons. Percent ash in wet body did not vary with age.

The data on dry matter composition of farmed sturgeons are reported in table 2. Protein level was the same (65%) in the 3 month and 10 month old fish but significantly lower (54.6%) in the 24 month old fish. On the other hand, the lipid content of dry matter significantly increased with age and was more than two fold higher in oldest sturgeons than in the youngest ones. Consequently, the energy content was also the highest in the 24 month old sturgeons.

Table 2 - Whole body composition of farmed Siberian sturgeon from 3 age classes

Age (months)	3	10	24
Body Weight (g)	30	230	1300
Dry matter(% BW)	17.18^{c}	22.15^{b}	29.42^{a}
Protein (% DM)	65.08^{a}	65.02^{a}	54.64^{b}
Lipid (% DM)	15.96^{c}	18.30^{b}	34.00^{a}
Energy (kJ/g DM)	21.60^{b}	22.52^{b}	26.27^{a}

On each row, means with the same letter are not significantly different ($p<0.05$)

Digestibility of the components of commercial diet

The ability of Siberian sturgeons to digest dietary components was evaluated by indirect method from composition analyses of fish faecal wastes. The results obtained from juvenile sturgeons fed commercial trout diet are given in table 3. The apparent digestibility coefficient (ADC) of protein was low, but the ADC of lipid was high (around 90%). Digestible energy (DE) provided to sturgeons by commercial trout diets was 15.7 kJ/g DM with a P/E ratio of 22.5 mg digestible protein/kJ DE.

Table 3 - Composition of the diet (% DM or kJ/g DM) and apparent digestibility coefficients (%)

	Composition	ADC
Dry matter	93.2	63.9
Protein	51.4	70.9
Lipid	12.5	89.7
Energy	21.5	73.4

Endogenous energy losses and utilization of body stores

Results on daily whole body energy losses due to fasting of Siberian sturgeon are reported in table 4. Among the data obtained from body composition analyses no significant ($p<0.05$) size effect was noticed. Average endogenous energy loss was 50 ± 12 kJ/kgBW/day. But, the value of fasting energy expenditure was significantly

lower (21 kJ/kgBW/day) when the evaluation was based on heat production from indirect calorimetric measurements. Data on some parameters of energy metabolism of fasted 10 month old sturgeons are given in table V. The daily oxygen uptake was closed to 1.5 g/kgBW. Assuming that N-ammonia excretion represented 85% of non-faecal nitrogen wastes, the amount of catabolized protein was 320 mg/kgBW/day. From these data, the utilization of body stores for energy purposes was evaluated : 34% of endogenous energy expenditure was supplied by proteins, 17% by carbohydrates and 49% by fats. Such oxidations of body reserves represented a loss of matter around 0.85 g/kg/day. Total dry matter losses estimated from whole body composition analyses were 1.1 ± 0.2 g/kg/day in fasted 3 month old sturgeons and 1.5 ± 0.8 g/kg/day in fasted 24 month old sturgeons.

Table 4 - Endogenous energy losses in Siberian sturgeon at a water temperature of 18°C

Age (months)	3	10	24
Body Weight (g)	30	230	1300
kJ/kg BW/day	42[1]	21[2]	59[1]
mg Protein/kg BW/day	280[1]	316[2]	370[1]
mg N/kgJ	1.20[1]	2.40[2]	1.10[1]

[1] Data from whole body composition analyses
[2] Data from indirect calorimetric measurements.

Table 5 - Fasting energy metabolism of Siberian sturgeon at a water temperature of 18°C.

O2 uptake (mg/kg/day)	1,486
CO2 output "	1,715
N NH4 excretion "	44
Respiratory quotient	0.839
Nitrogen quotient	0.078
Oxidized body reserves (mg/kg/day)	
Proteins	316
Fats	265
Carbohydrates	261

DISCUSSION

Growth performance and dietary energy utilization

Feeding rates are known to have a great impact on feed efficiency and to affect nutrient requirements (Tacon and Cowey, 1985). According to Hung and Lutes (1987), the optimum feeding rate for hatchery produced juvenile white sturgeon (30g - 100g) was 2.0% BW/day. For 2 year old white sturgeon weighing 2kg, Ruer et al. (1987) gave a daily ration of 0.65% BW. Voluntary feed intakes of 50 g and 1700g Siberian sturgeon were very close to these values. The decrease of voluntary feed intake with age of fish has often been reported (Brett, 1971 ; Marais and Kissil, 1979). However, owing to their long life span and late sexual maturity, 24 old month sturgeon could be yet considered

as juveniles. The feeding rate being the prime factor which can limit growth rate, the low voluntary feed intake of growing Siberian sturgeon could partly explain their low growth rate after their first year of life under farming conditions.

The energy retention efficiency (ERE) values were low (20% to 26% of gross intake), indicating that commercial trout diet was not suitable for sturgeon. An improvement of ERE could be expected with well-adapted diets. Moreover, recent works (Kaushik et al., 1989) showed that ERE raised to 37% of gross intake when diets contained extruded starch (DE 15.4 kJ/gDM) and a lower DP/DE ratio than in commercial trout diet (17.6 instead of 22.5 mg dig. protein/kJDE). The ERE was better in the 24 old month sturgeon than in the younger ones. It might be related to the cost of energy storage. In 3 and 10 month old sturgeon, energy gain was predominantly in the form of protein. In terms of energetics, protein growth is more expensive than the accumulation of fat (Smith et al., 1978 ; Jobling, 1985).

Whole body proximate composition

The proximate composition of the 3, 10 and 24 month old sturgeons randomly taken from the cohorts reared in our fish farm corroborated the results obtained trough growth trials. The decrease of moisture associated with age, observed in farmed Siberian sturgeon, has been commonly described in most other fish. A striking relationship between lipid and water has been shown in a large number of species, an increase in the proportion of one leading to a decrease in the other (Love, 1980). The changes in wet body components are often a mere reflection of the decrease in moisture. But, in farmed Siberian sturgeons, lipid content of dry whole body was also affected by age as could be expected according to the results on nutrient retention.

The values of protein and ash in wet body samples were comparable to those reported by Hung et al. (1987) for cultured white sturgeon. However, in white sturgeon, the percentage of ash was significantly correlated with age but fats in dry samples were not. Besides, percent lipid in wet carcasses of 22 month old white sturgeons was greatly lower than in 24 month old farmed Siberian sturgeons. These differences in body composition and its change with age are not necessarily due to differences between species. They could be the result of the feeding conditions, as it was suggested by Hung et al. (1987), body lipids being markedly influenced by the nutritional status of fish. The variations of whole body proximate composition associated with the increase in energy content in the oldest fish reflect changes with age in feeding rate and in storage of energy-yielding nutrients.

Digestibility of commercial diet

The digestibility of the most commonly used ingredients in fish diet have been studied in Salmonids, especially in rainbow trout (Cho et al., 1982). Salmonids digest protein and fat very well.

Compared to the value found in rainbow trout, the apparent digestibility coefficient of protein was low in sturgeon. The method of collecting faeces might be suspected. Indeed, Cho and Slinger (1979) showed that stripping the fish to remove the rectal contents led to the lowest values of digestibility coefficients compared to other methods of collecting faeces (anal suction, intestinal dissection, settling column). We previously attempted to study sturgeons digestibility in an automatic faeces collector system (Choubert et al., 1982). Unfortunatly, the conical and deep tanks, designed for avoiding losses by leaching, were not suitable for these fish. Owing to their ventral mouth, sturgeons need flat bottomed tanks. However, according to Cho and Slinger (1979), stripping method also affected digestibility coefficients of lipid and of energy in rainbow trout which was not the case in sturgeon. Besides, even with this method, the values of protein digestibility coefficients obtained in trout (around 80% of intake) exceed by far the present evaluation in sturgeon.

To our knowledge, no other data on ADC in sturgeon are to date available. However, enzymatic secretions have been studied in the digestive tract of an other species of sturgeon *Acipenser transmontanus* (Buddington and Doroshov, 1986a, b).

The level of digestive proteolytic enzymes was described to be high in juvenile white sturgeons, reflecting their carnivorous feeding habits. Enzymatic profiles suggested that these fish were well adapted to high level of dietary protein. Results of the digestibility trial based on analysis of faecal losses did not corroborate these data. Further investigations are needed for the knowledge of the ability of sturgeon to digest protein.

Endogenous losses and maintenance needs

Fish, as all other organisms, need a continuous supply of energy to sustain life processes regardless of whether or not they are consuming food. In fed fish, this requirement must be provided by the food. A fasted fish obtains the energy for maintenance by catabolizing body reserves. The energy expenditure of fasted fish including the expenditure for random swimming activity is generally considered as a close approximation of basal energy requirement (Braaten, 1979).

The value of daily oxygen uptake (1.5 g/kg) of fasted Siberian sturgeon was comparable to that of white sturgeon at the same temperature (Ruer et al., 1987) representing a daily energy expenditure of 20 kJ/kg. Energy needs for maintenance of sturgeons would be lower than those of trout (55 to 60 kJ/kg/day) at the same temperature (Cho et al., 1982) which agrees with the quiet behaviour of sturgeon. But the value of maintenance energy expenditure of Siberian sturgeon was more than two fold higher when the evaluation was based on body composition analysis. Such a discrepancy is difficult to explain. Differences in the experimental conditions might be suspected. For respirometry trials, fish were kept in covered tanks smaller than for other trials. So, the voluntary activity could be restricted because of both the size of the tank and the lower light. The influence of the conditions of measurements on the value of standard metabolic rate of fish was recently pointed out by Van Waversveld et al. (1989). These authors suggested that the energy expenditure level for maintenance of fish have been as often as not overestimated. Besides, energy losses of fasted 24 month old sturgeon (calculated from body composition) seemed very high as per the data obtained through growth trials : indeed, energy losses of 59 kJ/kg/day would represent 61.5% of gross energy.

In fasted Siberian sturgeon, the ammonia excretion rate was much lower than in Salmonids. This characteristic was already reported in larval Siberian sturgeon (Dabrowski et al., 1987) and was recently found to be unaffected by age and size of sturgeons (Salin, pers. com.). Unlike endogenous energy losses, protein losses of fasted 10 month old sturgeon estimated from indirect calorimetric measurements were in good concordance with the values obtained from body composition analyses (see Kaushik et al., 1991). From these data, maintenance needs were estimated to be about 60 mg N/kg BW/day in Siberian sturgeon at a water temperature of 18°C while these were 200 mg N/kg BW/day in trout at the same temperature (Luquet and Kaushik, 1981). On the other hand, the contribution of body protein to provide energy requirements in Siberian sturgeons could not be clearly established, due to the doubt on maintenance energy expenditure. According to the values obtained from body composition analysis, body protein utilization for energy purpose would be 1.15 mg N/kJ, a value lower than in Salmonids. But, as per data from indirect calorimetry, the contribution of endogenous protein to energy production would be 2.85 mg N/kJ which is in the same range as in trout (Luquet and Kaushik, 1981). Fat stores appeared to be the major source of energy in fasted Siberian sturgeon since half of the maintenance energy requirement was supplied by body fat.

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REQUIREMENTS FOR PROTEIN AND ESSENTIAL AMINO ACIDS AND THEIR UTILIZATION BY SIBERIAN STURGEON (*Acipenser Baeri*)

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RESUME

Les besoins protéiques du juvénile d'esturgeon sibérien (PMI : 22g) ont été estimés sur la base des réponses zootechniques à des taux de protéines alimentaires variant de 29 à 52%. Cette étude a été réalisée sur une période de 12 semaines. Basé sur les performances de croissance, le besoin optimal en protéines alimentaires brutes chez l'esturgeon sibérien a été estimé à environ 40 ± 2% du régime. Cependant, les performances maximales ont été enregistrées lors de l'utilisation d'un aliment contenant 49% de protéines brutes. Les estimations des besoins protéiques d'entretien basées sur les pertes endogènes au cours d'un jeûne de 4 à 5 semaines chez des juvéniles (Poids Vif : <30g) et des esturgeons plus gros (PV : >1000g) se sont révélées être de l'ordre de 420 à 500 mg/kg PV/jour. Ches ces poissons, la contribution de l'azote corporel pour les besoins énergétiques était comprise entre 1.1 et 1.2 mgN/kJ. Le rapport ARN/protéines décroît en fonction de l'âge et du jeûne, mais le taux de protéines alimentaires n'a aucun effet sur ce rapport. La composition corporelle totale en acides aminés de l'esturgeon a été déterminée au début et à la fin des essais. Nous avons pu ainsi calculer le dépôt quotidien des acides aminés essentiels (AAE) chez les poissons nourris, et la perte quotidienne en AAE chez les poissons privés de nourriture. Le profil du besoin en AAE a été estimé en fonction de l'accroissement quotidien en AAE chez des esturgeons nourris à l'aide de régimes différents. Les estimations sur les besoins en AAE de l'esturgeon sibérien ainsi recueillies, diffèrent peu des données disponibles pour les téléostéens. Il subsiste des doutes quant à la validité de cette technique pour l'estimation des besoins en AAE du poisson. Il en est de même des variations dues à l'âge ainsi que des effets du jeûne sur la composition corporelle totale en acides aminés.

Mots clés : *Acipenser*, protéines, acides aminés essentiels, énergie, besoins, utilisation, rétention.

ABSTRACT

Protein requirements of juvenile Siberian sturgeon (IBW : 22g) were estimated based on growth response over 12 wks to graded levels of dietary protein ranging from 29 to 52%. Based on growth performance, the optimum dietary crude protein requirement of Siberian Sturgeon was estimated to be about 40 ± 2 % of the diet, although maximum performance was recorded when these fish were fed a practical diet containing 49% crude protein. Estimates of maintenance protein requirements based on endogenous losses during a 4 to 5 wk fast of juvenile (BW : <30g) and bigger (BW : >1000g) sturgeon were in the range of 420 to 500 mg/kg BW/day. The contribution of body nitrogen to energy needs in these fish was calculated to be between 1.1 to 1.2 mgN/kJ. Whole body RNA, DNA contents were determined. RNA/protein ratios decreased with increasing age and due to fasting. Dietary protein levels had no effect on such ratios. Amino acid compositions of whole bodies of sturgeon were determined at the beginning and at the end of the trials. Daily essential amino acid (EAA) increments in fed fish and daily EAA losses in fasted fish were thus calculated. The EAA requirement profiles were estimated based on daily whole body EAA increments in sturgeon fed different diets. The estimated EAA requirements of Siberian sturgeon do not differ much from data available for teleosts. Discussion is made on the merits and validity of this technique for the estimation of EAA requirements of fish and on age-related variations and starvation effects on whole body amino acid composition.

Key-words : *Acipenser*, protein, essential amino acids, energy, requirements, utilization, retention

INTRODUCTION

Of the several species of sturgeon, the culture of Siberian sturgeon (*Acipenser baeri*) has of late gained interest in some of the European countries as is evidenced by the theme and venue of the present symposium itself. Once such interest is established, it becomes evident that reliable data on the nutrition of such a novel species be obtained. Earlier studies conducted by ourselves were concerned with nutrient utilization during ontogenesis (Dabrowski and Kaushik, 1986) and larval development (Dabrowski et al., 1985, 1987 ; Fauconneau et al., 1986) and with the utilization of dietary non-protein energy substrates by these fish under farming conditions (Kaushik et al., 1989 ; Médale et al., 1988 ; Médale et Kaushik 1990). Among other sturgeons, currently available information on nutritional requirements is restricted to the protein requirements for maximum growth of white sturgeon (*A. transmontanus*), estimated to be around 40% of the diet (Moore et al., 1988). As the protein requirements of fish are considered to be relatively high and as no information is currently available on the nutritional requirements of Siberian sturgeon for any essential nutrient, trials were conducted to determine the protein needs for growth and maintenance and also to define an ideal dietary essential amino acid (EAA) profile.

The essential amino acid requirement of fish, or of any other animal for that matter, is in general estimated based on growth and/or biochemical or metabolic responses to graded dietary levels of the concerned amino acid (Cowey and Luquet, 1983 ; Kaushik, 1985 ; Kaushik et al., 1988). Some authors have suggested that the whole body tissue EAA patterns or A/E ratios (ratio between individual EAA/sum of EAA x 1000) can well serve to formulate test diets useful for in defining an ideal dietary essential amino acid pattern before conducting studies on quantitative EAA requirements of fish (Arai, 1981 ; Ogata et al., 1983 ; Gatlin, 1987). Few others (Cowey and Luquet, 1983 ; Wilson and Poe, 1985) have shown that the quantitative EAA requirement patterns of a given fish species correlate well with the EAA pattern of the whole body tissue of the same animal. Much earlier, Ogino (1980) employed a method based on daily EAA increment in the whole body tissue of carp and rainbow trout for estimating their EAA requirements. This technique was also successfully applied by Jauncey et al. (1983) to estimate the EAA needs of tilapia (*Oreochromis mossambicus*). A similar method was employed here for the estimation of EAA requirements of Siberian sturgeon fed graded levels of protein through the assessment of daily EAA increment, loss and retention.

A critical analysis of the protein requirements of fish in comparison to those of other vertebrates (Luquet and Kaushik, 1980 ; Cowey and Luquet, 1983 ; Bowen, 1987) show i) that the protein requirements of teleosts are only apparently high, ii) that despite considerable homogeneity in the quantitative EAA needs, some problems do seem to exist as to the regulation of amino acid catabolism and iii) that the provision of proteins in great quantities invariably leads to the diversion of dietary proteins to meet the energy requirements of fish. An attempt is made here also to analyze the protein and amino acid needs of Siberian sturgeon in the light of the preceding observations.

MATERIALS AND METHODS

Three trials were conducted with Siberian sturgeon (*Acipenser baeri*) produced in the sturgeon hatchery (CEMAGREF) of our institute's (INRA) fish farm at Donzacq (Landes, France). All experiments were conducted in sand bottomed concrete tanks with flowing spring water at a constant temperature of 18°C. They were grown with commercial trout pellets until the beginning of the trials.

Trials : The protein requirements of juvenile Siberian sturgeon having an initial body weight (IBW) of 22 ± 2g, were estimated through a growth trial conducted over 45 days with graded levels of dietary protein (Trial 1). Five isoenergetic (Gross Energy : 20 MJ/kg DM) experimental diets were formulated in order to provide digestible protein (DP) levels ranging from 25 to 45% (Table 1). The amino acid compositions of the different diets are given in Table 2. Digestibility values of the different ingredients were based on known data for rainbow trout and also for Siberian sturgeon from earlier trials (Médale et al., 1988). All dietary ingredients were blended and pressed into dry pellets (Simon-Heesen Press) of 2.5 mm diameter. Ten groups of 100 sturgeon each were randomly alloted to different tanks and each diet was distributed *ad libitum* twice a day to duplicate groups of fish. All fish were weighed in group at the beginning, every 2 wks and at the end of the trial (45 days) and feed intakes recorded. Fish were fasted for 24 h and ten fish were sampled from the initial batch and from each batch at the end of the growth trial, weighed individually and frozen for subsequent analyses.

Table 1 - Composition of the experimental diets

Ingredients	Diets				
(%)	D25	D30	D35	D40	D45
Fish Meal	27.5	34.5	41.5	48.5	55.5
Soyabean meal	6.1	6.7	7.5	8.5	9.5
Extruded wheat	15.0	14.0	12.5	10.5	8.5
Crude corn starch	6.0	8.8	11.8	14.9	18.0
Gelatinized starch	33.3	24.8	16.4	8.2	
Fish oil	6.1	5.2	4.3	3.4	2.5
Vitamin mix [1]	2.0	2.0	2.0	2.0	2.0
Mineral mix [2]	2.0	2.0	2.0	2.0	2.0
Binder	1.0	1.0	1.0	1.0	1.0
Chromic oxyde	1.0	1.0	1.0	1.0	1.0
Proximate Composition (g or MJ/kg dry matter)					
Crude Protein	294.2	336.8	400.6	466.1	526.1
Crude Fat	75.3	78.1	87.3	96.7	98.9
Gross Energy	19.7	19.8	20.0	20.1	20.5
P/E ratio [3]	14.9	17.0	19.9	23.2	25.6

[1] Vitamin mix contained the following diluted in cellulose (g/kg mix) : vit A (500 000 IU/g), 1.5 ; vit D3 (100 000 IU/g), 1.5 ; vit E (500 IU/g), 6 ; vit K, 0.25 ; thiamin, 0.75 ; riboflavin, 1.5 ; pyridoxine, 0.75 ; nicotinic acid, 8.75 ; vit C, 25 ; folic acid, 0.25 ; vit B12 (1000mg/kg), 2.5 ; inositol, 50 ; biotin (2%), 6.25 ; calcium pantothenate, 2.5 ; choline (50%), 200.

[2] Mineral mix contained the following ingredients (g/kg mix) : calcium carbonate, 215 ; magnesium hydroxyde, 124 ; KCl, 90 ; ferric citrate, 20 ; KI, 0.4 ; NaCl, 40 ; calcium hydrogen phosphate ($CaHPO_4$), 500 ; copper sulfate, 3 ; zinc sulfate, 4 ; cobalt sulfate, 0.2 ; manganese sulfate, 3 ;

[3] mg crude protein/kJ gross energy

Two other trials were conducted to estimate the maintenance requirements based on endogenous losses during a 4 to 5 wk fast. In trial 2, four groups of 100 fish each (IBW : 30 ± 3g) were either fed *ad libitum* twice a day, a commercial diet (Crude Protein : 54.5% of dry matter ; Gross Energy : 21.9 kJ/g DM) or fasted for 4 wks. Growth characteristics were followed and fish samples were drawn as described above.

Table 2 - Amino acid composition of the different experimental diets (g/16gN)

DIET	D25	D30	D35	D40	D45
ARG	5.67	6.66	7.56	6.39	7.61
LYS	7.25	11.74	15.50	13.54	12.97
HIS	1.65	2.37	2.63	2.20	2.42
ILE	4.88	6.23	7.16	6.10	6.04
LEU	8.06	11.74	10.71	8.80	8.75
VAL	5.25	6.67	7.06	6.42	6.75
PHE	4.51	4.68	4.73	4.60	4.61
THR	4.62	5.51	6.21	5.37	5.80
TYR	3.45	4.12	5.36	4.80	4.19
ASP	9.72	13.20	10.41	12.07	11.75
GLU	16.40	21.04	24.30	22.99	20.40
SER	4.58	5.38	5.55	4.71	5.44
GLY	5.79	6.77	6.46	5.57	6.93
ALA	5.85	7.23	8.14	7.89	5.98
Sum of EAA	45.33	59.73	66.92	58.66	59.14

Another experiment (Trial 3) was conducted with two batches of 25 individually identified sturgeon (IBW : 1100 ± 100 g) each over 35 days. One group was fed a commercial trout diet (CP : 49.9% DM ; GE : 21.5 kJ/g DM) and the other was fasted. Growth performance was followed on individual fish. At the begininning and at the end of the trial, 4 fish from each group was sampled for analyses.

Analyses : Analyses of dry matter (110°C, 24h), crude protein (Kjeldahl nitrogen x 6.25), gross energy (Gallenkamp adiabatic bomb calorimeter, crude fats (methanol : chloroform extraction) of diets and freeze-dried whole body tissue samples were performed following usual laboratory procedures. Whole body RNA and DNA contents were determined following procedures described by Buckley (1979), using DNA sodium salt from Salmon testes (Sigma D1626) and RNA purified from baker's yeast (Sigma R6750) as standards. Dry or freeze-dried samples were hydrolyzed with 6N HCl at 110°C for 24h and chromatographic separation and analyses of amino acids (HPLC- Varian model 5000, Aminotag reversed phase column) were performed after OPA (orthophthaldehyde, Sigma P1378) derivatization of amino acids following slightly modified procedures of Gardner and Miller (1980) and of Rajendra (1987). Unfortunately however, good separation and quantification of the following amino acids could not be done with this chromatographic procedure : tryptophan, cystine, and proline.

Based on body composition analyses, nutrient retention efficiencies were calculated as

$$NRE\ (\%) = 100x(FBW \times FBc - IBW \times IBc)/(TFI \times Dc)$$

where IBW and FBW represent initial and final body weights, IBc and FBc the initial and final whole body composition (dry matter, protein, energy or EAA), TFI the total feed intake and Dc the nutrient content (dry matter, protein, energy or EAA) in the diet. Daily EAA gain or loss per 100g fish was calculated as

$$EAAI \text{ or } EAAL = (FBW \times FBc - IBW \times IBc)/(MBW \times 100 \times n° \text{ days})$$

where MBW = (IBW+FBW)/2 and IBc and FBc the initial and final EAA composition of the whole body tissues. EAA losses were determined only in fasted sturgeon of > 1 kg BW (Trial 3).

The EAA requirements (as % of dietary protein) were computed as

$$\text{Need} = (EAAI)/DPI \times 100 \text{ or } (EAAI+EAAL)/DPI \times 100$$

where DPI = mean daily digestible protein intake per 100g fish. All statistical analyses (ANOVA followed by Duncan's test, break point analysis and polynomial fitting of the growth data to dietary protein levels) were made using SAS statistical procedures for microcomputers (SAS, 1987).

RESULTS AND DISCUSSION

Growth performance of fish : The results of growth trials are reported in Table 3. Final batch mean body weights ranged between 39 and 45g, although individual FBW ranges of 10 fish from each tank were considerably higher (28 to 66g). All groups were fed ad libitum twice a day. Nevertheless, the voluntary feed intakes did not vary between the different groups (around 2% of IBW /day). This is not surprising since it has been shown that the voluntary feed intake of fish is dependant on energy density and as the energy density of the different diets were the same. The values of the different parameters of growth and of protein utilization were in the upper range of values reported for many teleosts (see data compiled by Bowen, 1987). The feed gain ratios (FGR) decreased with increasing dietary protein levels and were extremely low in all treatments showing the excellent overall performance of all groups (Table 3). The protein efficiency ratios (PER) were also very high (>3) in fish fed diets containing less than 40% protein and decreased at dietary protein levels of 46% and above (2.8). This inverse relationship between PER and dietary protein levels is well in conformity with what is known for most animals. This relation was influenced further by the changes in the protein/energy (P/E) ratios of the diets containing highly digestible carbohydrates. Inclusion of digestible carbohydrates has been shown to improve protein utilization in Siberian sturgeon (Kaushik et al., 1989). Similarly, although no significant differences ($P > 0.05$) were found in nutrient retention efficiencies (Table 3), a tendency of a decrease in protein retention with increasing dietary protein levels was observed.

Protein requirements for Growth : In Fig. 1, the percent daily weight gains are plotted against dietary crude protein levels. The maximum slope occurred at a dietary crude protein level of 38.7 %. Break point analysis of individual final weights showed that the breakpoint occurred at a dietary crude protein level of 40 ± 2%. This data is comparable to what has been observed for white sturgeon (*A. transmontanus*) by Moore et al. (1988), but differs from what we had inferred from an earlier study on the utilization of digestible carbohydrates by Siberian sturgeon (Kaushik et al., 1989). Expressed in terms of absolute requirements (protein intake per unit body weight at maximum growth), juvenile sturgeon appear to require about 7-8 g of protein/kg BW/day ; this is considerably lower than the median values both of fish and even of other vertebrates (Bowen, 1987). As the growth rates of sturgeon in this study were high and since considerable amount of digestible energy in the form of digestible carbohydrates was also available, it is preferable to express the needs in terms of unit body weight gain. This amounts to a protein requirement of about 300g per kg weight gain and is comparable to data for other fish. The optimum P/E ratio is in the range of 20 to 22 mg protein / kJ.

Table 3 - Growth performance of sturgeon fed the different expermintal diets for 45 days at a water temperature of 18°C [1]

	Diets				
	D25	D30	D35	D40	D45
Growth characteristics					
Initial Weight (g)	22.00	21.00	21.00	20.00	18.75^{ns}
Final Weight	38.9^{a}	39.21^{ab}	39.67^{ab}	44.72^{b}	44.71^{b}
Daily Feed Intake [2]	2.00	1.97	1.95	1.80	2.20^{ns}
Daily Wt Gain [2]	1.96^{a}	2.05^{a}	2.17^{ab}	2.91^{b}	2.81^{ab}
FGR [3]	0.87^{a}	0.86^{a}	0.80^{a}	0.76^{ab}	0.66^{b}
PER [4]	3.63^{a}	3.41^{a}	3.12^{ab}	2.80^{b}	2.73^{b}
Nutrient retention efficiencies (% of intake) [5]					
Dry matter	23.44	23.86	23.89	24.12	28.50^{ns}
Protein	39.02	38.25	33.19	33.04	31.70^{ns}
Energy	30.11	28.60	28.20	28.05	33.85^{ns}

[1] Mean values are given. Values in the same line followed by a same superscript letter are not significantly different from each other ;
[2] Calculated as percent of initial body weight (IBW)
[3] Feed : Gain ratio = Dry feed intake/Wet weight gain
[4] Protein efficiency ratio = Unit Wet weight gain/ Unit Protein intake
[5] Nutrient deposited/Nutrient intake x 100

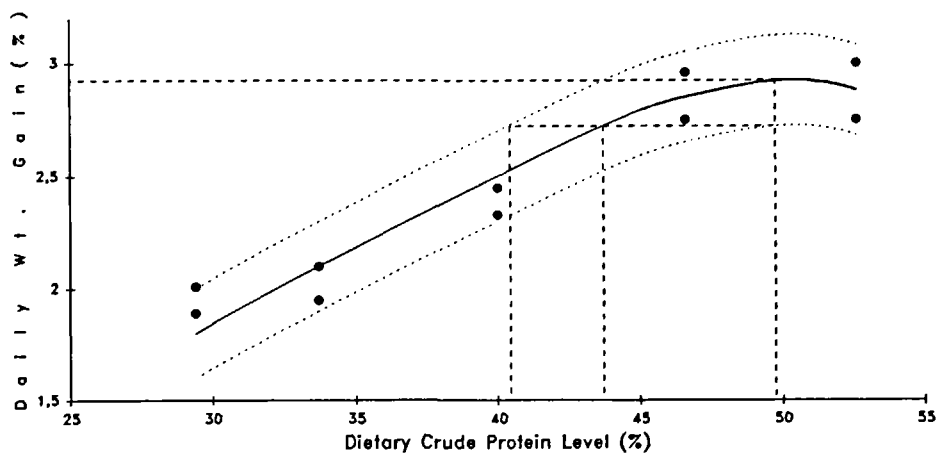


Figure 1 - Daily weight gain of Siberian sturgeon (IBW : 22 g) fed graded levels of dietary protein over 45 days at 18 °C

Endogenous losses and maintenance needs : Data on daily whole body protein and energy losses due to fasting are reported in Table IV for Siberian sturgeon of two different sizes. No significant ($P < 0.05$) size effect was noticed. During fasting, the average body protein and energy losses were respectively 340 ± 50 mg protein and 44 ± 12 kJ energy/kgBW/day. The contribution of body proteins to meet the energy

requirements were found to be in the range of 1.15 ± 0.15 mgN/kJ, irrespective of size, data lower than those for rainbow trout (Luquet and Kaushik, 1980), but considerably higher than those of higher vertebrates (around 0,2 mgN/kJ ; Reeds, 1988). This definitely shows that in sturgeon, like in other teleosts, contribution of body proteins to meet the energy requirements is high.

The endogenous nitrogen loss in sturgeon (60 mgN/kgBW/d) appears to be considerably lower than data available for other species (see Luquet and Kaushik, 1980). Ammonia excretion measured in very small Siberian sturgeon (BW : 100 to 300 mg) were found to be lower than in other teleosts (Dabrowski et al., 1987). Estimates of ENE made by Salin (pers. com.) in sturgeon were quite variable, depending upon the duration of fast with an average ENE value of less than 40 mgN/kg BW/d after a fortnight's fast. This ENE value is also lower than what has been found for trout at 18°C (Kaushik, 1980). Although, contrary to statements made in this regard by Bowen (1987), the endogenous losses (neither ENE, the endogenous nitrogen excretion nor body nitrogen losses) by themselves cannot be recognized as truly representative of maintenance requirements (Luquet and Kaushik, 1980), they can at least serve as an approximate measure of maintenance needs. The biological value of proteins are known to vary with incorporation levels (Ogino and Chen, 1973). Calculations based on an average biological value (50 to 60%) of good quality protein sources normally incorporated in the diets of farmed fish, provide an estimation of the maintenance needs of sturgeon to be about 420 to 500 mg protein/kg BW/day.

Table 4 - Whole body protein (% DM) and Amino acid composition (g/16gN) of sturgeon fed graded levels of dietary protein [1]

group	initial	d25		d30		d35		d40		d45	
		Mean ± s.d.		Mean ± s.d.		Mean ± s.d.		Mean ± s.d.		Mean ± s.d.	
Prot %DM	66.19	57.33	0.62	59.99	1.26	60.73	0.92	63.75	2.11	63.09	0.92
ARG	5.31	6.73	0.02	6.24	0.01	7.17	0.12	6.74	0.29	7.15	0.43
LYS	5.38	12.25	0.83	10.65	0.22	12.49	0.21	11.72	1.75	10.86	0.06
HIS	1.22	2.18	0.03	1.90	0.21	2.15	0.10	2.03	0.10	2.04	0.14
ILE	3.74	5.19	0.00	4.66	0.19	5.34	0.25	5.09	0.26	4.90	0.33
LEU	6.27	10.04	0.22	7.82	1.51	7.28	1.61	6.79	0.78	6.70	1.55
VAL	3.83	5.56	0.02	5.02	0.19	5.50	0.15	5.29	0.24	5.28	0.37
PHE	3.40	4.03	0.00	3.56	0.09	4.19	0.06	4.06	0.09	3.78	0.26
THR	3.54	5.19	0.00	4.62	0.11	5.25	0.07	4.99	0.17	4.98	0.33
TYR	2.99	3.78	0.04	3.45	0.13	4.22	0.00	4.02	0.18	3.65	0.13
ASP	4.71	13.48	0.36	9.66	1.04	8.21	0.15	8.37	3.07	10.44	0.68
GLU	12.12	19.51	0.29	18.04	0.66	19.55	0.23	18.46	0.13	17.34	1.13
SER	4.13	5.17	0.25	4.60	0.09	4.90	0.11	4.76	0.07	5.03	0.42
GLY	7.86	12.24	0.08	11.60	0.12	10.16	0.43	9.19	0.36	10.20	0.90
ALA	5.63	7.99	0.11	7.36	0.10	7.98	0.07	7.58	0.11	5.68	0.49
EAA	35.67	55.02	0.45	48.26	2.95	54.13	1.43	50.93	2.40	49.74	3.45

[1] Mean ± 1.s.d. of two replicate samples of ten fish each are given.

Amino acid composition of fish : The amino acid composition of fish fed the different diets (Trial 1) and of bigger sturgeon either fed or fasted for 35 days (Trial 3) are reported in Tables 5 and 7 respectively. The whole body amino acid composition of sturgeon is comparable to data reported for other teleosts (Wilson and Cowey, 1985). No significant differences were found in the amino acid compositions of sturgeon fed

graded levels of protein over 12 wks (Table 4). On the contrary, in the case of big sturgeon (> 1 kg BW), some differences were noted between fasted and fed fish, especially for the basic essential amino acids (Table 7). Under fasting conditions, a change in the relative proportions of structural and globular proteins can account for such differences in whole body amino acid composition. Besides, a comparison of the amino acid composition of small sturgeon to that of big sturgeon (from Tables 4 and 7) also shows that some minor changes do occur in the amino acid composition of body proteins with body size. Statistical analysis of combined data from big and small sturgeon showed significant differences ($P<0.05$) for at least one essential amino acid (lysine) and for few non-essential ones (aspartic acid, glycine, serine and tyrosine). This then is in some way slightly contradictory to what was observed by Wilson and Poe (1985) in channel catfish (*Ictalurus punctatus*) of two different sizes (30 and 800g BW) and in common carp (*Cyprinus carpio*) by Schwarz and Kirchgessner (1988).

Data on the effect of diets or of fasting on whole body amino acid composition is rather scarce, despite numerous studies on the changes in the free amino acid composition of different tissues during fasting or after a meal. Of the few works, Créach and Serfaty (1965) observed some differences due to fasting in the protein-bound amino acids of common carp fasted for several months, whereas Timoshina (1970) found no such effect of fasting on the amino acid composition of fish muscles, including those of carp and of rainbow trout. Schwarz and Kirchgessner (1988) found no effect of dietary changes in protein/energy on the amino acid composition of carp whole body proteins. Expressed however in terms of body weight (% carcass dry matter), considerable differences were observed.

Table 5 - Mean daily essential amino acid increment (mg/100gBW/day) and estimation of dietary amino acid requirement profile (% of dietary protein) of Siberian Sturgeon
IBW : 22g ; 18°C

	EAA increment		EAA Requirement				
	mean[1]	sd	Sturgeon mean[2]	sd	Trout[3]	Carp[3]	Tilapia[3]
ARG	11	2	2.8	0.6	3.1-4.0	3.3-4.0	2.8
LYS	26	4	6.3	2.0	4.7-6.0	4.7-6.0	3.8
HIS	4	1	1.1	0.4	1.4-1.8	1.2-1.5	1.1
ILE	9	1	2.1	0.6	2.1-2.8	2.0-2.6	2.0
LEU	13	5	3.2	1.8	3.9-5.0	3.7-4.8	3.4
VAL	9	1	2.3	0.6	2.8-3.6	2.6-3.4	2.2
PHE	6	1	1.5	0.4	3.0-4.1	2.9-3.8	2.5
THR	9	1	2.2	0.6	1.9-2.4	1.8-2.3	2.9

1 means of all ten groups of fish fed different levels of dietary protein
2 calculated as (increment/digestible protein intake) x 100
3 Range of values for Rainbow trout and for Carp from Ogino (1980) and values for Tilapia from Jauncey et al. (1983)

Table 6 - Endogenous losses and the contribution of body proteins for energy purposes in Siberian sturgeon

Body Weight (g)	30	1000
mg Protein/kg BW/day	280	370
kJ/kg BW/day	36	54
mgN/kJ	1.23	1.09

It is generally recognized that the protein-bound amino acid composition of related species is extremely constant. This appears to be true also in the case of higher animals. Analysis of available data seems to indicate that interspecific differences in the whole body amino acid composition would be much smaller than differences in the amino acid compositions of different tissues within any given species (Reeds, 1988). In addition, the tissue composition of growth would be the major source of variation in the amino acid composition of the whole body. In this regard, although muscle tissue represent the major tissue (*ca* 50% of body weight) in the case of fish, the protein synthesis and turnover rates in this tissue are considerably lower than in many other active tissues like the digestive tract. Tissue-specific differences in the efficiency of protein deposition (protein increment / protein synthesis) have also been shown to exist in teleosts (Fauconneau and Arnal, 1985). Allometric changes in the relative muscle mass combined with age-related differences in protein synthesis and turnover rates can considerably alter the whole body amino acid composition. Besides, the variation in the whole body amino acid composition (Table 4) might also be due to differences in size of the free amino acid pools. Unfortunately, we do not have any data on the free amino acids of the same fish.

Table 7 - Whole body protein (% DM) and Amino acid composition (g/16gN) of sturgeon (BW :> 1000g) either fed or fasted for 35 days at a water temperature of 18°C [1]

	DIET INITIAL		FED FISH		FASTED FISH	
			mean	sd	mean	sd
Prot %DM	49.88	54.64	57.23	3.68	64.57	6.53
ARG	4.83	5.82	7.66	0.37	5.82	0.58*
LYS	5.42	9.97	11.13	1.11	8.71	0.76*
HIS	1.88	2.55	2.37	0.10	2.08	0.12*
ILE	5.39	6.30	5.38	1.24	5.53	0.39
LEU	16.17	14.73	7.38	1.83	10.91	2.50
VAL	6.21	6.01	6.01	1.78	5.18	0.34
PHE	4.53	5.09	3.76	0.46	4.47	0.37
THR	4.61	5.73	5.39	0.83	4.89	0.39
TYR	6.92	6.34	5.29	0.86	6.49	1.14
ASP	11.24	13.67	10.21	2.13	12.13	0.97
GLU	20.04	21.79	18.29	1.63	20.27	1.69
SER	6.08	5.97	5.63	0.73	6.03	0.62
GLY	7.59	11.24	11.89	1.66	14.79	1.78
ALA	7.07	8.25	7.31	2.66	9.01	0.92
Sum EAA	55.97	62.54	54.76	7.42	54.63	5.85

[1] Mean ± 1 s.d. values obtained for four individual fish are presented. * The asterisk denotes a statistically significant ($P<0.05$) difference between fed and fasted groups

Essential amino acid requirements : Data on daily EAA increments and on EAA needs are reported in Table 6. These data show that the quantitative requirements of Siberian sturgeon for most essential amino acids (except for lysine and phenylalanine) are in the same range of values found with a similar approach in other teleosts. Schwarz and Kirchgessner (1988) found considerable discrepancy between the whole body EAA pattern of carp and the official EAA requirements for the same species (both expressed in relation to lysine), especially for the branched chain amino acids (Ile, Leu, Val) and for basic amino acids like arginine and histidine. Ogino's (1980) estimates on the EAA requirements of carp and rainbow trout were slightly lower than the

established recommended (NRC, 1981) values for some of the ten essential amino acids. Similarly, recent data of Santiago and Lovell (1988) on the individual EAA needs of juveniles (initial body weight : 40 to 90 mg) of nile *Tilapia* based on growth studies (8 wks) do not compare very well with earlier data obtained by Jauncey et al. (1983) which were based on daily whole body EAA increments. However, in the present work, the EAA needs were calculated based on average values of EAA increments from all groups of fish fed different levels of protein in relation to actual digestible protein intakes, whereas in earlier works, a more or less a theoretical DP intake level was used in such calculations. Besides, Wilson and Poe (1985) consider that such underestimations might be due to the non-recognition of the contribution of the EAA for maintenance and other metabolic activities of the fish. In the case of small sturgeon, no correction was made for this possible endogenous EAA loss.

On the other hand, data obtained during trial 3 with big sturgeon, show that the daily EAA increments (Table 8) are much lower than those of fast growing under yearling Siberian sturgeon. In this trial, the daily EAA losses were also measured in fasted fish, and these values may be considered to reflect the EAA requirements for maintenance. Despite the recognition of such endogenous losses, data presented in Table 8 show that the requirements of big sturgeon for many EAA, are not much different from those of small sturgeon (Table 5). Some differences appear however to exist : requirement for histidine appears to be low and the daily increment in leucine and in phenylalanine in fed fish were too low to allow any reasonable estimation of requirements for these EAA. Daily losses were also very high in the case of leucine.

Table 8 - Mean daily EAA increment (mg/100gBW/day), daily EAA loss (mg/100gBW/day) and estimation of dietary amino acid requirement profile (% of dietary protein) of Siberian Sturgeon weighing >1000g ; 18°C

EAA	EAA increment		EAA loss		EAA Need	
	mean	sd	mean[1]	sd	mean	sd[2]
ARG	4.6	0.6	1.0	0.5	2.2	0.7
LYS	4.1	1.9	3.4	2.0	3.7	1.4
HIS	0.5	0.2	1.0	0.6	0.6	0.4
ILE	2.3	0.5	2.0	1.4	0.8	0.5
LEU			8.5	5.7		
VAL	6.0	1.2	3.2	1.4	1.6	0.9
PHE			2.6	1.2		
THR	2.7	0.5	2.0	1.5	1.3	0.4

[1] losses measured in sturgeon fasted for 35 days
[2] calculated as (increment + loss / digestible protein intake) x 100

Data presented in Tables 5 and 8 can however be considered only as indicative of the true EAA requirement profile of the Siberian sturgeon ; to be truly valid, these data should be corroborated with data obtained in specific studies based on growth or other responses (biochemical or metabolic) to graded dietary levels of individual essential amino acids.

RNA/DNA and RNA/protein ratios : Diet quality and fasting have been shown to exert some influence on protein synthesis in sturgeon (Fauconneau et al., 1986). The ratio of RNA to DNA can be related to both long-term and recent growth history of fish (Buckley, 1979). In Table 9 and 10 are reported the values on the whole body RNA, DNA contents, RNA to DNA and RNA to protein ratios of Siberian sturgeon. A comparison

between fasted and fed sturgeon either of 30g initial body weight or of bigger size show that significant differences exist (Table 9). Since RNA to protein ratios can be considered to represent to a certain extent the rates of protein synthesis (McMillan and Houlihan, 1988), then two major observations can be made : that the whole body protein synthesis decreases considerably with increasing age/size and that long term fasting has a negative effect also on protein synthesis. McMillan and Houlihan (1988) did not find any differences in the rates of protein synthesis in the muscle of rainbow trout after a six-days fast. Barrows et al. (1988) found that dietary protein levels had some effect on RNA/DNA ratios in the muscle tissue of Walleyes. Our data on sturgeon fed different dietary crude protein levels do not indicate any significant difference either in the RNA/DNA ratios or in the RNA/protein ratios of the whole body (Table 10).

Table 9 - Effects of fasting on whole body RNA and DNA contents (mg/g DM), RNA/DNA and RNA/protein (µg/mg) ratios in Siberian sturgeon of different sizes [1]

	INITIAL		FED FISH		FASTED FISH	
	mean	sd	mean	sd	mean	sd
30 g Sturgeon						
RNA	14.8	0.2a	15.4	1.1a	9.9	0.3b
DNA	5.6	0.3a	4.9	0.1a	5.4	0.6a
RNA/DNA	2.7	0.1a	3.1	0.2a	1.9	0.2b
RNA/Protein	24.3	2.3a	25.8	3.4a	14.8	3.6b
> 1000g Sturgeon						
RNA	3.0	0.5c	4.2	1.1c	2.9	0.8c
DNA	3.1	1.1bc	4.2	0.7ab	2.4	0.3c
RNA/DNA	1.1	0.3c	1.0	0.2c	1.2	0.3c
RNA/Protein	6.0	1.4c	7.3	1.1c	4.5	0.9d

[1] Values in the same line followed by a same superscript letter are not significantly different from each other ($p<0.05$)

Table 10 - Effects of feeding different dietray crude protein levels on whole body RNA and DNA contents (mg/g DM), RNA/DNA and RNA/protein (µg/mg) ratios in Siberian sturgeon

Dietary Protein level (%)	RNA mg/gDM		DNA mg/gDM		RNA/DNA		RNA/Protein µg/mg	
	mean	sd	mean	sd	mean	sd	mean	sd
25 %	13.6	1.8	4.2	0.7	3.3	0.1	23.7	2.1
30 %	13.7	1.3	4.5	0.5	3.1	0.1	22.8	1.9
35 %	12.6	0.7	4.2	0.1	3.0	0.2	20.7	2.8
40 %	14.6	2.7	5.0	0.0	2.9	0.6	22.4	2.3
45 %	15.5	1.6	5.3	0.6	2.9	0.1	24.6	1.8

Amino acid utilization : Other interesting observations here concern the relative differences in daily increments of few EAA like lysine, arginine and leucine and in th retention efficiencies which appear to vary considerably depending on the DP intake levels (Fig. 2, 3) Indeed, for most essential amino acids, the retention efficiencies are significantly higher in fish fed diet D25 (25% DP) than in fish fed higher protein levels (Fig. 2). This maximal utilization of ingested EAA is already a probable indication of insufficient input of EAA or of protein in relation to what is required by the fish. Once there is sufficient input, the retention efficiencies are more or less the same.

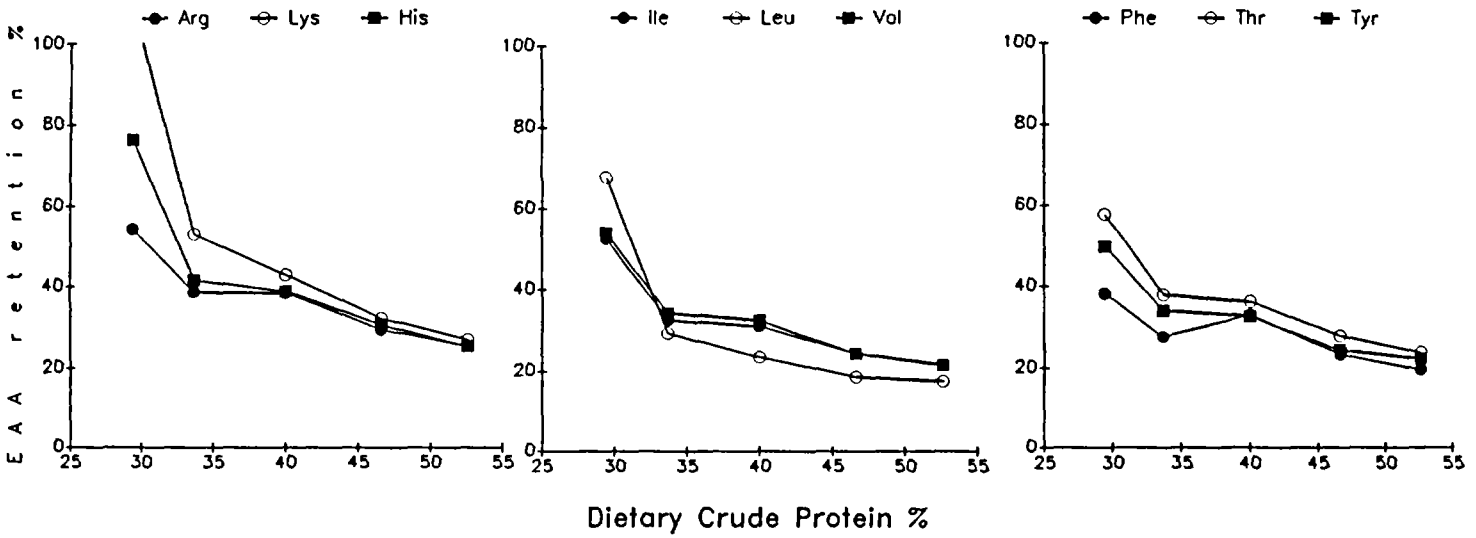


Figure 2 - Effect of dietary crude protein levels on EAA retention efficiencies in Siberian sturgeon.

Apparent availability values of ingested amino acids to Siberian sturgeon are high, much comparable to other teleosts, judging from our previous studies (Dabrowski et al., 1987). However, if we consider the daily mean essential amino acid increments in relation to dietary protein levels, there seem to exist an optimum below and above which, considerable variations occur especially for at least two essential amino acids : lysine and arginine (Fig. 3). Daily increment in another essential amino acid, leucine, shows a decrease with increasing dietary protein levels (Fig. 3). In Fig. 4 are presented the essential amino acid needs in relation to dietary crude protein levels. While the quantitative needs for most EAA, expressed as percent dietary protein, appear to be independant of the dietary protein level, those for arginine and lysine show much variations, in accordance with the differences in the increments.

This observation requires further elucidation in the light of 1) possible antagonism between these two EAA, which has been shown to exist at least in rainbow trout (Kaushik and Fauconneau, 1984), 2) existing variability in the available data on arginine requirements of rainbow trout (Kaushik et al., 1988) and 3) the specific role of such amino acids in protein synthesis and degradation (Fauconneau et al., 1986).

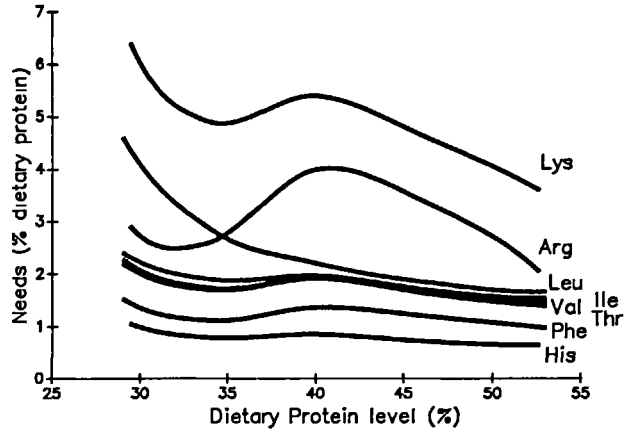


Fig. 3 - Daily EAA increment in Siberian sturgeon fed graded levels of dietary protein

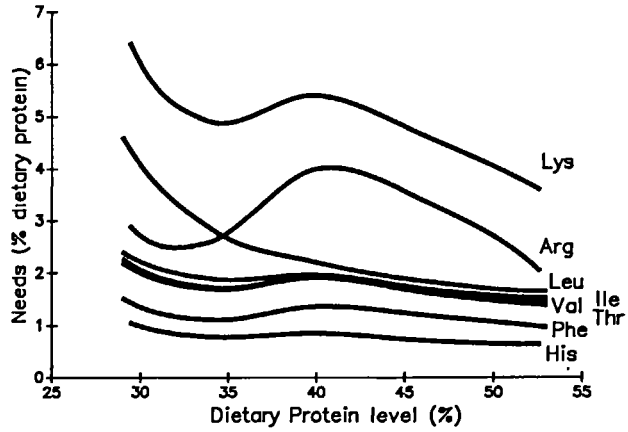


Fig. 4 - Essential amino acid needs of Siberian sturgeon in relation to dietary protein levels

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LIPID MOBILIZATION DURING EARLY DEVELOPMENT OF STURGEONS

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RESUME

Les oeufs d' esturgeons sont plus riches en lipide que ceux de la plupart des autres espèces, à cause de leur forte teneur en triacylglycerol. Les phospholipides des oeufs contiennent une quantité relativement élevée en phosphatidyl-ethanolamine. La plupart des lipides sont catabolisés pendant le développement larvaire. Avant le passage à l'alimentation exogène, jusqu'à 60 % des lipides (principalement des Triacylglycerols) sont utilisés. Les oeufs et larves d'esturgeons contiennent de grandes quantités d'acides saturés et mono insaturés. Celles-ci diminuent de 1,5 fois durant le développement. Au début de l'ontogenèse le rapport n 3/n 6 est très petit. Dès le début de l'organogenèse et jusqu'à l'éclosion l'importance des lipides dans le métabolisme énergétique est plus faible que dans la formation des structures nouvelles. La consommation d'énergie liée au mouvement après l'éclosion provoque une augmentation importante du métabolisme énergétique. Après le passage à l'alimentation active, le taux d'accumulation des lipides de structure est plus important que celui des lipides de réserves. Simultanément on observe un accroissement notable des lipides de type n 3 et une diminution de ceux du type n 6, en particulier pour les lipides polaires ce qui provoque l'augmentation du rapport n 3/n 6.

Mots clés : *Acipenseridae*, embryogenèse, larves, lipides, composition fractionnelle, acides gras, métabolisme énergétique.

ABSTRACT

The sturgeons'eggs are richer in lipids than the eggs of many other species due to the high quantity of triacylgycerol. The phospholipids of the eggs contain a relatively high quantity of phospatidyl-ethanolamine. The majority of lipids are catabolised during larval development. Before the transition to exogenous feeding, up to 60 % of lipids are catabolised. Triacylglycerol is a major component of the catabolised lipids. A high quantity of saturated and monounsaturated fatty acids is characteristic of sturgeon eggs and larvae. During the development, the proportion of these fatty acids declines by 1.5 times. In the early development of sturgeons the ratio n 3/ n 6 is very low. Between the organogenesis and hatching, lipids are utilized rather for the formation of new structures than as a source of metabolic energy. Upon hatching metabolic activity increases substantially because of the utilization of energy for locomotion. After the transition to exogenous feeding the rate of accumulation of structural lipids is higher than the rate of accumulation of lipid reserves. At the same time a marked increase in the levels of the family n 3 and a depletion in the levels of the family n 6 are observed especially in polar lipids, which results in the increase of the ratio n 3/n 6.

Key-words : *Acipenseridae*, embryogenesis, larvae, lipids, lipid class composition, fatty acids, energetic metabolism.

INTRODUCTION

Sturgeons are one of the most ancient representatives of the present ichthyofauna. Their survival up to the present time and relatively high abundance in some geographical areas prompted us to consider the mechanisms, primarily metabolic characteristics which may be responsible for their adaptation to changing habitat. Increasing anthropogenic pressure on the ecosystems is creating their greater dependence upon artificial reproduction. Effective artificial reproduction demands knowledge of metabolic processes, especially in the early development of sturgeons. In this paper lipids metabolism, one of the main aspects of metabolism, in embryogenesis of sturgeons and the first 10 days of exogenous feeding is treated.

MATERIAL AND METIIODS

Several species of sturgeons were used in this study. Sterlet (*Acipenser ruthenus L.*) eggs and larvae were collected from pond hatcheries in the Rostov and Donetsk regions, stellate sturgeon (*A. stellatus Pallas*) eggs and larvae from the sturgeon hatchery on the river Volga, beluga (*Huso huso L.*) and chip (*A. nudiventris Lov.*) from the sturgeon farm on the river Ural, Siberian sturgeon (*A. baeri Brandt*) from the full cycle warm water farm.

Eggs were contained in Yuschenko's and "Osetr" Incubators. Water temperature during the incubation period of beluga, sterlet, chip and Siberian sturgeon fluctuated from 12 to 16°C. Stellate eggs were incubated at 18°C. Live zooplancton, predominantly cladocerans and decapsulated *Artemia* eggs were used as the first food for larvae. The larvae were reared in tanks and throughs at densities 15 000/tank. Stellate larvae were contained in ponds. Water temperature in tanks was up to 20°C, but most of the period 17-18°C. In ponds where the stellate larvae were contained the water temperature was higher up to 22°C.

Stages of development were determined according to Dettlaff et al. (1981). Samples were homogenized in a mixture of chloroform : methanol (2:1 v/v). Lipid was weighed. Lipid classes were separated by thin-layer chromatography (TLC) on Merck and Silufol plates. For separation we used hexan : diethyl ether : acetic acid (86 : 14 : 1,5 by vol.) as the solvent and developed in 10 % (w/v) phosphomolybdic acid. Samples were scanned in reflected light at the wave length of 650 nm. Phospholipids were separated using chloroform : metanol : water (65:25:4 by vol.) and chloroform : methanol : acetic acid : acetone : water (30:9:4:2,5:2 by vol.) as solvents and developed in iodine vapour. Samples were identified by comparison with known standards and scanned in the reflected light at the wave length of 540 nm.

Fatty acid classes were separated by gas chromatography. Fatty acid methyl esters were quantified on a Micromat HRGC 412 with two flame-ionizing detectors. The chromatograph was equipped with fused capillary columns utilizing standard phases of different polarization OV-1 (24 m x 0,32 mm x 0,20 μm) and OV-351 (24 m x 0,32 mm x 0,20 μm). The oven was programmed to raise the temperature from 100°C to 200°C at a rate of 15°C min, 240°C - for the injector, 250°C - for the detector, the rate of hydrogen as the carrier gas being 2 ml/min. Several samples were separated on a GC-9A equipped with a capillary column utilizing a stationary phase Carbowax (30 m x 0,32 mm x 0,20 μm). The oven was programmed to raise the temperature from 150°C to 275°C at a rate of 5°C min, 300°C - for the injector, 325°C - for the detector, the rate of the carrier gas being 1 ml/min.

Lipid class composition was identified by comparison of the obtained retention values with the calculated relative retention and equivalent chain lengths of standard fatty acid methyl esters determined on the above mentioned columns under similar conditions. Doubtful samples were identified using mass-spectrometry. Acid ratios were quantified on an integrator using the method of area normalization.

RESULTS

Throughout the period of sturgeon's eggs development, lipid content declined about 20 times. The decline was uneven. Thus, at stage 16 blastomeres lipid proportion exhibited a sharp decrease and increased a bit by gastrula stage. But on the whole, fatness dropped from 12,8 % to 0,7 % of wet weight by hatching in stellate and from 10,7 % to 0,5 % in sterlet.

Lipid content of embryos was rather high. After the hatching, lipids of stellate and sterlet embryos accounted for 18 % and 12,1 % of wet weight respectively. Through further development lipid content of larvae declined and by stage 45 (the transition to exogenous feeding) it dropped to 3 % of wet weight in stellate and to 2,6 % in sterlet. There were no significant differences in lipid content of other sturgeons larvae at the same stage, i.e. 3 % in beluga and 2 % in chip.

Lipid loss during sturgeon's embryogenesis accounted for 50-60 % of the initial level, making 0,9 mg/ind. in stellate and 0,6 mg/ind. in sterlet.

After the begining of exogenous feeding lipid content continued to decline and after 10 days accounted for 1,5 % in beluga and stellate , 1,1 % in sterlet and to 0,8 % in chip.

The content of triacylglycerol (TAG) in sturgeon eggs and larvae is very high (table 1). Throughout the development the proportion of these lipids declined and the proportion of structural lipids increased. The process continued after the transition to exogenous feeding. It should be noted that the proportion of sterol ester in eggs and larvae is very low. In coldwater sturgeons, beluga and Siberian sturgeon, the proportion of this class is much higher than in other species.

Phosphatidylcholine (PC) is a major class among structural lipids. The proportion of PC was twice as much as that of phosphatidylethanolamine (PE). Through the larval development the quantity of the latter increased more rapidly than that of PC : between hatching and the transition to exogenous feeding their proportions increased 3 times and 1,5-2 times as high respectively. Moreover, calculations showed that by the transition to exogenous feeding the quantity of PC declined by 10 % as compared with eggs, while the quantity of other phospholipids increased. On the whole the proportion of phospholipids increased twice as much during embryogenesis and represented the essential lipid class in feeding larvae (tables 1 and 2).

Table 1 - Changes in composition of lipid classes in early life history of sturgeons (weight % of total lipids)

stage	species	TAG	SE	FFA	S	PL
eggs	SV	80,1	0,5	0,3	7,6	11,5
stage	ST	76,4	tr.	0,1	6,2	17,3
4	SS	67,0	5,0	tr.	8,9	19,0
stage	SV	81,4	0,2	0,5	7,1	10,8
36	ST	78,8	0,5	0,5	5,0	15,2
	SS	63,8	8,7	tr.	11,3	16,2
	SV	57,3	0,5	0,4	13,1	28,6
larvae	ST	56,0	0,4	0,4	13,4	29,8
stage	SS	41,6	19,4	tr.	15,7	23,3
45	BL	50,6	18,2	1,0	16,2	20,7
	SP	48,0	7,0	0,6	9,5	28,2
	SV	30,1	1,7	0,1	23,1	45,0
10 days of	ST	24,0	2,6	0,2	23,2	50,6
exogenous	SS	32,7	22,1	tr.	14,5	29,8
feeding	BL	33,8	13,4	3,5	17,5	31,8
	SP	27,0	4,0	2,2	29,8	36,9

TAG : *triacylglycerol* ; SE : *sterol ester* ; FFA : *fatty free acids* ; S : *sterol* ; PL : *phospholipids*
SV : *stellate sturgeon (Acipenser stellatus).*
ST : *sterlet (Acipenser ruthenus).*
BL : *beluga (Huso huso).*
SS : *siberian sturgeon (Acipenser baeri).*
SP : *chip (Acipenser nudiventris).*

Table 2 - Changes of phospholipid class composition in early development of stellate and sterlet sturgeons (weight % of total lipids)

stage	eggs				larvae			
	stage 4		stage 36		stage 45		10 days exog.feed.	
species	SV	ST	SV	ST	SV	ST	SV	ST
Phosphatidyl-choline	7,8	11,1	7,6	11,3	14,4	16,3	24,2	28,8
Phosphatidylethano-lamine	2,7	4,8	2,8	3,3	9,9	9,5	15,6	17,8
Lyso-phosphati-dylcholine	0,5	0,7	0,2	0,3	0,9	1,3	0,7	0,9
Sphin gomyelin	0,4	0,7	0,2	0,3	2,4	1,3	3,1	1,5
Cardiolipin	tr.	tr.	tr.	tr.	1,1	0,5	1,4	1,1

SV : *Acipenser stellatus*
ST : *Acipenser ruthenus*

Lipid class composition in the progeny from females of one species is highly variable. For example, the variation coefficient for TAG in Siberian sturgeon was 12 %, sterol ester content could exhibit 10-fold increase or decrease.

Sturgeon eggs are characterized by a high content of saturated (about 30%) and monounsaturated fatty acids (about 40%). The low proportion of the linolenic family is also typical of fatty acid composition of sturgeon eggs and larvae total lipids, thus the ratio n 3/n 6 is low in sturgeons (table 3).

Fatty acid composition in eggs of different sturgeon species is similar. The proportion of saturated and monounsaturated fatty acids increased post-hatch as compared to eggs (especially of acids 16:0, 18:1 and 20:1), while the proportion of polyunsaturated acids especially of those with four and more double bounds declined respectively.

Throughout the development of larvae the proportion of saturated acids decreased and the proportion of polyunsaturated acids increased. At the same time a substantial increase in the proportion of the linolenic family and the linoleic family but to a smaller extent was observed. The increase in the proportion of eicosapentaenoic and docosahexaenoic acids was extremely high.

In feeding larvae the proportion of saturated fatty acids increased and that of monounsaturated fatty acids dropped drastically on the background of the continuing increase in the content of polyunsaturated fatty acids. The increase in the proportion of polyunsaturated fatty acids as dependent on the habitat and food composition can be explained by the higher content of dienoic, trienoic or tetraenoic, pentaenoic and hexaenoic acids (table 3).

Table 3 - Fatty acid composition of eggs and larvae of *A. stellatus* and *A. ruthenus* (weight % of total amount)

Fatty acids	*A. stellatus*				*A. ruthenus*			
	eggs	larvae			eggs	larvae		
	stage 4	stage 36	stage 45	10 d. of exog. feed.	stage 4	stage 36	stage 45	10 d. of exog. feed.
saturates								
14:0	1,2	1,5	1,4	0,5	1,2	1,4	1,1	1,4
15:0	0,5	0,6	0,5	0,3	0,6	0,7	0,5	0,5
16:0	21,3	24,2	22,0	21,5	23,4	27,0	24,6	21,0
17:0	0,6	0,8	0,5	0,6	0,8	1,0	0,8	1,3
18:0	3,2	3,7	3,9	11,0	3,1	4,3	5,4	11,9
19:0	0,1	0,1	0,1	0,1	0,1	0,1	0,2	0,2
20:0	0,1	0,1	0,1	0,1	-	0,2	0,2	0,4
iso-acids								
15:0	0,3	0,3	0,3	0,1	0,3	0,3	0,2	0,4
16:0	0,2	0,2	0,2	0,1	0,2	0,3	0,2	0,2
17:0	0,8	0,9	0,6	0,3	1,1	1,3	1,1	1,0
18:0	0,1	0,1	-	-	0,1	0,3	0,3	0,6
antiisoacids								
15:0	0,2	0,1	0,2	tr.	0,1	0,1	0,1	tr.
16:0	tr.	tr.	tr.	0,2	0,1	tr.	0,2	0,2
17:0	0,5	0,2	0,3	0,2	0,7	0,3	0,4	0,1
18:0	0,1	tr.	tr.	0,1	0,1	0,1	0,1	0,4
monoenes								
14:1 n-7	0,2	-	-	-	-	-	tr.	tr.
16:1 n-7	7,7	8,0	6,9	3,8	10,6	10,7	9,0	7,9
16:1 n-5	0,3	0,4	0,4	0,3	0,5	0,7	0,5	1,1
17:1 n-12,9	1,4	1,7	1,5	1,8	1,8	2,3	1,7	2,5
18:1 n-12,9,7	34,4	35,4	35,9	21,8	27,5	28,5	29,1	17,8
18:1 n-5	0,2	0,2	0,2	tr.	0,4	0,4	0,4	0,4
19:1 n-15	0,2	0,2	0,3	tr.	0,4	0,3	0,3	0,2
19:1	0,1	0,5	0,6	tr.	-	0,6	0,1	0,7
20:1 n-9	1,5	1,9	1,5	0,9	0,8	1,0	0,9	0,7
20:1 n-7	-	-	-	-	0,2	0,4	0,3	0,2
22:1 n-9	0,2	0,4	0,1	-	tr.	0,4	0,1	0,4
dienoics								
16:2 n-6	0,1	0,2	0,2	tr.	-	0,5	0,2	0,4
18:2 n-7	1,0	1,0	1,1	0,6	0,5	0,5	0,5	0,6
18:2 n-6	4,0	3,8	4,5	2,6	2,1	1,8	2,0	4,3
18:2 n-5	0,2	0,3	0,3	0,2	0,3	0,4	0,3	0,6
20:2 n-7	0,2	0,2	0,3	0,2	tr.	0,2	0,2	0,4
20:2 n-6	0,4	0,9	0,6	0,4	0,4	1,1	0,4	1,1
22:2 n-6	0,1	0,5	0,3	-	0,1	0,5	0,2	0,4

Table 3 cont.

trienoics								
17:3 n-7	0,1	0,3	0,2	tr.	0,2	0,3	0,1	0,3
18:3 n-6	0,5	0,6	0,7	0,2	0,4	0,3	0,2	0,5
18:3 n-3	1,9	1,6	1,8	1,5	1,2	0,9	1,0	3,2
20:3 n-6	0,3	0,3	0,2	0,4	tr.	0,2	tr.	0,4
20:3 n-3	0,2	0,2	tr.	0,2	0,2	0,2	0,1	0,3
tetraenoics								
16:4 n-3	0,1	tr.	0,1	-	0,2	-	0,1	0,1
18:4 n-3	0,4	0,4	0,8	0,3	0,8	0,4	0,4	0,8
20:4 n-6	2,3	1,2	1,9	5,4	2,7	1,6	2,9	2,7
20,3 n-3	0,2	0,2	0,1	0,3	0,3	0,2	0,2	0,4
22:4 n-6	0,3	0,2	0,4	0,2	0,2	0,1	0,2	0,2
Pentaenoics								
19:5 n-3 + 19:4	tr.	0,1	0,2	0,2	-	0,2	0,5	0,2
20:5 n-3	2,9	2,1	2,5	4,5	5,5	3,6	4,3	5,9
21:5 n-3	0,1	0,1	tr.	-	0,2	0,2	0,1	-
22:5 n-6	0,3	0,1	0,1	0,4	0,5	0,2	0,4	0,1
22:5 n-3	1,1	0,6	0,7	1,7	1,7	1,0	1,2	1,5
22:6 n-3	7,3	2,9	6,2	15,5	7,8	2,7	6,5	1,9
TOTAL								
Saturates	29,1	32,7	30,1	36,0	31,9	37,3	35,4	39,5
Monoenes	46,1	48,8	47,3	27,9	42,5	45,4	42,6	31,1
polyenes	24,0	17,9	22,7	34,3	25,3	16,9	22,0	26,6
n-3	14,3	8,2	12,0	23,7	17,9	9,4	14,4	14,3
n-6	8,2	7,9	8,8	9,2	6,4	6,2	6,6	10,1
n-3/n-6	1,8	1,0	1,4	2,6	2,8	1,5	2,2	1,4

Separate analysis of changes in fatty acid composition of polar and neutral lipids appears to demonstrate the absence of changes in the compostion of polar lipids throughout the egg development. It should be noted that there is some increase in the proportion of monounsaturated fatty acids, a stable reduction of pentaenoic acids and an increase of docosahexaneoic acid. Substantial reduction in the proportion of palmitic, stearic and docosahexaenoic acids in polar lipids of post-hatch larvae as compared to eggs at stage 35, i.e. pre-hatch eggs, suggests that the egg envelopes contained high proportions of these fatty acids.

Polar lipids of post-hatch larvae demonstrated a marked depletion in the levels of palmitic acid as well as in the levels of oleic and linolic families with 18-atom carbon radicals, the proportion of acids of these families with 20-atom and 22-atom radicals increased, except for docosahexaenoic acid (table 4).

The composition of polar lipids of feeding larvae was altogether different. The proportion of the linolic family dropped drastically as the result of the depletion in the level of arachidonic acid and the ratio n 3/n 6 increased.

Table 4 - Changes of fatty acid composition of polar lipids in A. *baeri*, (Weight % of total amount)

Fatty acid	stage age								
	4	5	18	29	35	36	45	10 d. exogen. feed.	20 d. exogen. feed.
saturates									
14:0	0,6	tr.	0,3	0,3	0,1	0,2	tr.	0,1	0,1
15:0	0,1	0,2	0,3	0,3	0,2	0,2	0,1	-	tr.
16:0	9,7	13,5	16,0	14,2	16,0	14,0	6,9	8,0	6,6
17:0	0,3	0,8	1,0	1,7	0,7	0,4	0,2	0,2	0,3
18:0	4,7	6,2	7,7	6,6	7,6	6,8	5,5	5,4	4,0
20:0	1,0	0,8	0,6	0,5	0,4	0,6	1,1	2,0	2,8
monoenes									
16:1 n-7,5	1,4	0,9	1,4	1,7	1,5	1,2	0,4	1,1	1,0
17:1 n-12,9	1,2	1,5	0,9	0,9	0,8	0,7	0,6	0,4	0,3
18:1 n-12,9,7,5	7,7	11,0	13,7	12,4	13,7	12,0	6,2	9,9	8,0
19:1	1,3	0,3	0,3	-	0,3	1,1	0,8	-	-
20:1 n-9,7	5,7	2,3	2,3	2,9	2,7	2,5	5,2	5,2	4,0
22:1 n-9	5,3	2,6	1,5	2,6	1,1	2,4	4,1	3,9	1,9
24:1 n-9	3,6	1,1	0,3	1,6	1,1	1,6	3,5	4,7	1,7
dienoics									
18:2 n-7	1,0	1,1	1,0	1,0	1,0	0,8	0,6	0,6	0,8
18:2 n-6	3,4	4,5	5,2	4,5	5,2	4,5	2,0	1,8	1,8
20:2 n-7	-	0,7	0,5	1,1	0,4	0,6	1,4	1,5	-
20:2 n-6	1,4	1,6	1,6	1,1	1,4	1,4	2,2	2,1	3,7
22:2 n-7	-	0,7	0,2	-	0,3	1,5	-	1,1	1,4
22:2 n-6	2,0	1,1	0,3	-	0,1	1,1	2,8	1,7	1,4
trienoics									
18:3 n-6	-	-	-	-	-	0,3	0,5	0,2	0,4
18:3 n-3	1,5	0,9	0,8	0,7	0,7	0,9	1,1	1,3	1,4
20:3 n-6	1,0	1,0	1,0	1,5	0,9	1,1	3,2	0,9	1,4
20:3 n-3	1,1	-	-	-	-	-	-	3,2	3,6
22:3 n-6	1,7	-	-	-	-	0,3	1,4	-	3,0
22:3 n-3	1,3	1,3	0,4	0,8	0,4	0,7	1,9	3,1	3,2
tetraenoics									
18:4 n-6	2,2	1,7	1,6	1,4	1,5	1,4	0,8	0,9	1,5
18:4 n-3	0,7	0,4	0,4	0,4	0,4	0,4	-	6,4	3,9
20:4 n-6	9,0	13,9	14,3	12,2	14,3	13,5	14,6	2,0	3,2
20:4 n-3	1,5	-	-	-	-	tr.	-	2,8	1,5
22:4 n-6	1,2	1,3	-	1,1	-	0,5	2,6	0,7	1,5
22:4 n-3	1,4	0,6	-	0,4	-	0,3	2,1	1,3	1,2

Table 4 cont.

pentaenoics									
20:5 n-3	3,0	2,2	2,1	2,5	2,1	2,5	2,3	7,1	8,8
21:5 n-3	-	-	-	0,5	-	-	-	1,6	-
22:5 n-6	1,2	0,4	-	-	-	0,3	2,2	1,3	1,5
22:5 n-3	1,0	1,5	1,8	-	-	0,8	1,4	1,3	5,6
22:6 n-3	10,9	13,0	13,3	13,8	15,3	11,8	8,6	6,5	6,5
TOTAL									
Saturates	16,1	21,5	25,9	23,6	25,1	22,2	13,8	15,7	13,8
Monoenes	26,2	19,7	20,4	22,1	21,2	21,5	20,8	25,2	16,9
Polyenes	46,5	47,9	44,5	43,0	44,0	44,7	51,7	39,4	60,0
n-3	22,4	19,9	18,8	19,1	18,9	17,4	18,2	34,6	38,4
n-6	23,1	25,5	24,0	21,8	23,4	24,4	32,3	11,6	19,4
n-3/n-6	1,0	0,8	0,8	0,9	0,8	0,7	0,6	3,0	2,0

Special emphasis should be placed on changes of fatty acid composition of neutral lipids primarily composed of TAG (table 5). Throughout the egg development the proportion of saturated and monounsaturated fatty acids increased to some extent, while the proportion of polyunsaturated acids decreased beginning from stage 18, i.e. from the initiation of organogenesis. During hatching a marked depletion in the levels of saturated and monounsaturated fatty acids was observed. During larval development the proportion of saturated fatty acids continued to decrease. The proportion of monounsaturated fatty acids with 18-atom and shorter carbon radicals also declined, while the proportion of trienoic and tretraenoic acids increased. Among polyunsaturated fatty acids the levels of pentaenoic and docosahexaenoic acids depleted. After the transition to exogenous feeding the only level to increase is the proportion of tetraenoic acids at the expense of acids 18:4 (n 3) as well as 20:4 (n 3) and 22:4 (n 6). In general, after the transition to exogenous feeding the proportion of the linoleic family increased and that of the linolic family decreased both in neutral and polar lipids. The ratio n 3/n 6 increased respectively.

Table 5 - Changes of fatty acid composition of neutral lipids in *A. baeri*, (% of total amount)

	stage age								
Fatty acid	4	5	18	29	35	36	45	10 d. exogen. feed.	20 d. exogen. feed.
saturates									
12:0	0,2	-	-	0,2	0,4	0,2	0,3	0,2	0,1
14:0	0,7	0,4	0,5	0,3	0,5	0,2	0,3	0,3	0,5
15:0	0,4	0,4	0,5	0,3	0,5	0,2	0,2	-	0,1
16:0	12,6	14,8	12,9	13,4	15,8	9,9	8,4	7,3	6,9
17:0	0,4	0,6	0,6	0,8	1,5	0,4	0,3	0,1	0,3
18:0	2,2	1,8	2,2	2,1	2,1	2,3	1,9	2,1	2,3
20:0	1,3	0,8	1,5	0,7	0,4	1,9	1,9	2,0	1,3

Table 5 cont.

monoenes									
16,1	4,2	4,0	4,4	3,2	4,5	2,4	2,0	1,5	2,1
17:1	1,6	2,0	2,5	2,1	2,1	1,3	1,0	0,6	0,9
18:1	21,9	26,3	25,1	23,6	30,1	18,8	17,8	16,7	16,0
19:1	-	0,9	0,4	0,5	0,7	0,6	1,3	-	0,4
20:1	3,1	2,3	1,0	1,8	2,1	3,3	3,2	4,0	3,5
22:1	3,1	2,4	2,1	1,7	1,4	4,0	5,9	5,1	3,7
24:1	1,8	-	0,4	1,9	0,1	0,8	1,4	3,7	3,2
dienoics									
18:2 n-7	1,2	1,5	1,5	1,5	1,3	0,7	1,2	0,2	0,3
18:2 n-6	6,6	10,0	9,4	9,2	9,4	7,3	6,0	2,4	3,8
20:2 n-7	0,9	0,3	1,1	0,6	0,9	1,0	0,8	2,0	1,7
20:2 n-6	1,1	1,2	1,3	1,0	1,1	2,3	1,5	1,9	1,8
22:2 n-7	1,8	0,8	-	0,6	-	0,6	1,4	1,9	1,5
22:2 n-6	1,5	0,8	0,8	1,0	-	0,8	1,7	1,5	0,5
trienoics									
18:3 n-6	0,1	0,2	0,3	0,3	0,2	0,7	0,5	0,7	0,6
18:3 n-3	0,9	0,7	1,1	0,8	0,9	1,9	1,2	0,7	0,7
20:3 n-6	0,8	0,7	1,4	0,5	0,9	1,3	1,1	2,5	1,6
20:3 n-3	0,9	0,7	0,6	1,2	0,7	1,0	2,8	3,0	2,5
22:3 n-6	1,0	0,4	0,5	0,5	-	1,0	1,3	-	0,8
22:3 n-3	0,9	0,6	0,7	0,9	0,2	1,3	1,1	1,4	0,9
tetraenoics									
18:4 n-6	1,9	1,6	2,4	2,7	2,2	2,4	2,3	1,0	1,0
18:4 n-3	1,1	0,6	0,6	0,7	1,1	1,0	1,1	7,1	9,1
20:4 n-6	2,3	3,0	3,0	3,1	2,8	3,7	4,4	1,5	1,2
20:4 n-3	1,5	0,6	0,7	0,5	1,0	1,0	1,2	-	2,9
22:4 n-6	2,0	0,3	0,6	0,7	0,1	1,4	0,6	3,5	1,3
22:4 n-3	1,6	0,1	0,6	0,5	0,4	0,7	1,9	1,0	1,1
pentaenoics									
20:5 n-3	2,2	2,2	1,6	1,6	0,9	2,4	2,0	4,4	3,8
21:5 n-3	-	-	0,6	-	-	2,1	1,9	-	0,6
22:5 n-6	0,7	0,2	0,5	0,5	0,1	0,9	0,9	1,8	1,8
22:5 n-3	1,5	1,4	0,6	0,4	0,4	1,7	1,7	1,2	0,5
22:6 n-3	4,4	4,3	4,9	4,6	2,2	4,3	2,9	2,4	1,7
TOTAL									
Saturates	17,8	18,8	18,4	17,8	20,7	15,1	13,3	12,1	10,0
Monoenes	35,7	38,1	35,9	34,8	41,1	30,3	33,0	31,8	30,5
Polyenes	36,7	33,2	35,0	33,5	27,8	41,5	41,5	41,9	41,7
n-3	15,0	9,0	12,0	11,2	7,8	17,4	17,8	21,2	23,8
n-6	17,9	19,4	20,2	19,5	16,8	21,8	20,3	16,8	14,4
n-3/n-6	0,8	0,5	0,6	0,6	0,5	0,8	0,9	1,3	1,7

DISCUSSION

Eggs and larvae of sturgeons are supplied with large energy reserves. The lipid contents of sturgeon eggs is much higher as compared to a wide range of fish species (Sidorov, 1983 ; Gershanovich et al., 1987). Such a high quantity of lipids is characteristic only of the eggs of salmonids (Sidorov, 1983). Our results suggest that by the transition of exogenous feeding, up to 60 % of lipids are utilized by sturgeons. It is somewhat higher than estimated earlier and close to the ratio documented for salmonids (Shatunovsky, 1980). The increase of the level of monounsaturated fatty acids throughout development indicates that reserve lipids supply substrate for the energy metabolism to a lesser extent than for the formation of new structures. It is in correspondence with a previous assumption that glycogen in eggs serves as a main source of metabolic energy (Shatunovsky, 1980), since the level of energy metabolism during embryogenesis is not high. Low rates of reserve lipid utilization are observed in the eggs of cod but the embryogenesis of this species occurs at a lower temperature (Fraser et al., 1988). Increase of lipid content between blastula and gastrula stages is also observed in Coregonid species (Nefedova et al., 1983). Changes in lipid content of developing eggs are reported in cod, but in this species they are related to changes in the quantity of PC, which is viewed as the structural lipid (Lapin et al., 1981 ; Fraser et al., 1988). So it could be established that lipid catabolism during egg development is not great and that the water content increase is primarily responsible for the depletion in lipid proportion during the egg development of sturgeons.

High quantities of lipids are utilized after hatching. A marked depletion in the levels of saturated and monounsaturated fatty acids in neutral lipids between stages 35 and 36 is a testimony to the high utilization of metabolic energy during the hatching.

So lipid metabolism during embryogenesis provides the energy expenditure and processes of new organs and tissues formation using only reserves stored in eggs. That's why the lipid quantity decrease throughout this life stage. After the begining of the exogenous feeding the food lipids serve as source for organ and tissue growth. This explains the increase of lipid amount in the body. But because of predominant protein growth in the young sturgeon the lipid quantity increase proportionaly to dry weight raised to the power less than 1 (Gershanovich et al., 1987).

The large quantity of lipids in sturgeon eggs and larvae results from the high content of TAG, the main class of lipid reserves (Lapin et al., 1981). In sturgeons the proportion of this class is at higher levels than in the majority of fish species. In sea fish it accounts for 40 % in herring (Shatunovsky, 1980), 13 % - in Atlantic halibut (Falk-Petersen et al., 1986), below 9 % in cod (Fraser et al., 1988). It is evident that the lower the temperature of the egg development the lower the content of TAG in marine fish. In diadromous and freshwater fish the proportion of this lipid class is usually higher than in marine fish, as for example, in steelhead (Sidorov, 1983), although in some species the TAG content is equal to marine ones as for example in Coregonids (Nefedova et al., 1983 ; Sidorov, 1983). However the TAG proportion in these species is lower than in sturgeons.

Low ratio of PC and PE is characteristic of sturgeon eggs. In eggs of Atlantic halibut this ratio is about 10 (Falk-Petersen et al., 1986), in cod - 8 (Fraser et al., 1988), in Coregonids 6 (Nefedova et al., 1983), while in sturgeons the ratio is close to 3. The depletion in the levels of PC throughout the development of sturgeons is in correspondence with that described for cod (Fraser et al., 1988). It may strengthen the conclusion that this class of phospholipids is a source of metabolic energy and phosphate, choline and free fatty acids, as the essential elements of the metabolic process. The large energy reserves of sturgeon eggs and larvae represented by TAG will not apparently require such a high quantity of PC versus PE as in other species. A marked depletion of TAG in the larval development suggests that major lipid proportion is utilized for energy metabolism.

The composition of fatty acids in sturgeons is rather peculiar. If high proportions of saturated and monounsaturated fatty acids could be easily explained by

the high quantity of TAG characterized by high saturation rates, the low ratio of n 3/n 6, especially of polar lipids, seems to be unusual. It is much lower than in marine fish (Tocher et al., 1985 ; Falk-Petersen et al., 1986), anadromous steelhead (Cowey et al., 1985) and even freshwater species, in the latter the ratio approaches 4. It could be explained by rather low levels of 20:5 (n 3) and 22:6 (n 3) fatty acids, the proportion of which even in neutral lipids of Atlantic halibut is much higher (Falk-Petersen et al., 1986). On the other hand, 18:2 (n 6) and 20:4 (n 6) exibit a higher ratio in sturgeons especially in polar lipids. This situation may be the result of a relative preference for warm water by sturgeons. Comparative studies with coldwater species revealed that a higher temperature has brought about the depletion in the levels of docosahexaenoic acid (Bolgova et al., 1983).

CONCLUSION

High lipid content and the large quantity of reserve lipids (TAG) are characteristic of sturgeon eggs. PC dominates in strutural lipids. The ratio of PC and PE is not high (about 3). Total lipids of eggs contain predominantly monounsaturated and saturated fatty acids. Polar lipids exhibit extremely high quantities of arachidonic and docosahexaenoic acids. A total of 50 fatty acids has been identified. Thirteen acids account for 90 % of the total number. Low ratio n 3/n 6 is characteristic of lipids in sturgeons. Throughout the egg development the small quantity of lipids is utilized, thus lipid content of post-hatch embryo is rather high. Developing larvae utilize approximately 60 % of lipids, predominantly reserve lipids (TAG). PC is the major component of catabolized structural lipids. It is natural that the proportion of structural lipids increases primarily at the expense of PE. From the first feeding onwards, the proportion of structural lipids continues to increase. The ratio n 3/n 6 increases both in neutral and polar lipids.

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ONTOGENETIC DEVELOPMENT OF STURGEONS : SELECTED PHYSIOLOGICAL EXAMPLES

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RESUME

Les aptitudes physiologiques des embryons et larves d'esturgeons diffèrent de celles d'animaux plus âgés. A partir d'observations sur les habitudes alimentaires naturelles, sur les caractéristiques du métabolisme et de la digestion et sur des études empiriques relatives aux besoins alimentaires, on montre que les stades précoces et tardifs de la vie des esturgeons se différencient par les besoins alimentaires. Les stades précoces sont plus sensibles aux conditions extrême de l'environnement et aux polluants. Ces différences peuvent être attribuées à des aptitudes physiologiques réduites sinon différentes des jeunes stades. Au fur et à mesure qu'ils grandissent, les esturgeons acquièrent les caractéristiques physiologiques des adultes. Ces aptitudes moindres durant les jeunes stades peuvent être compensées par 3 voies : par incorporation de la part de la femelle de composés spécifiques dans le jaune, par un comportement maternel consistant à déposer les oeufs dans un environnement propice et par le comportement des larves incluant les habitudes alimentaires et l'habitat.

Mots clés : esturgeons, ontogenèse, développement, physiologie, digestion, nutrition, température, pollution.

ABSTRACT

Embryonic and larval sturgeon differ from older fish in several physiological capacities. Based on observations of natural feeding habits, metabolic and digestive characteristics, and empirical studies of nutrient requirements, the early and later life stages of sturgeon differ with respect to nutrient requirements. Early life stages also exhibit lower tolerances for environmental extremes and pollutants. The differences can be attributed to either reduced, or at least different, physiological capacities of the early life stages. As sturgeon grow they acquire the adult physiological characteristics. Despite the reduced capacities of the early life stages, they can be compensated for in three ways : by maternal investment of specific components into the yolk ; by maternal behaviors involving deposition of the eggs into suitable environments ; and by larval behaviors, including specific feeding habits and habitat selection.

Key-words : sturgeons, ontogeny, development, physiology, digestion, nutrition, temperature, pollution.

INTRODUCTION

Ontogenetic development consists of an intricately orchestrated series of events that are initiated when the egg and sperm are united. Of great interest to developmental biologists is understanding how expression of the genetic material present within the individual cells of multicellular organism varies over time, and from cell to cell.

The patterns of gene expression during the ontogeny can usually be related to the environmental conditions normally encountered by each life history stage during a species phylogeny. Perhaps the most dramatic examples can be drawn from comparisons of vertebrates before they hatch, or are born, when they are relatively quiescent and dependent on nutrient reserves provided by the mother, with the posthatch, or postpartum, stages when levels of activity increase manifold and there is the onset of exclusive dependency on external foods.

Exemplary of such ontogenetic changes are the differences between the hemoglobins present in larval and adult stages of sturgeon (Chikhachev and Tsvetnenko, 1980).

As a result of the patterns of gene expression, there is a matching of physiological characteristics to the conditions that have prevailed during a species phylogeny.

To date, the majority of developmental research has focused on vertebrates other than fish, due principally to the small size of larval fish. However, the rapid expansion of the aquaculture industry and an increasing awareness of the environment has led to an increase in the number of studies being performed with fish. Most of the efforts have been devoted to species spawning large, demersal eggs, such as the chondrosteans. This reflects two considerations : many commercially important species spawn large, demersal eggs ; and the difficulties associated with studying small, pelagic eggs and larvae.

Corresponding with the increasing culture of the sturgeons, the majority of developmental research focuses on the physiological parameters which have application to the propagation and culture of sturgeons. This paper reviews the growing literature of the ontogenetic changes in physiological characteristics.

The focus is on three areas, each with great impact on sturgeon culture : nutrient requirements, digestive physiology, and responses to changes in environmental conditions.

NUTRIENT REQUIREMENTS

Rapid, economical growth of fish, obviously the principal objective of a culturist, can be attained only by the availability of suitable feeds. In turn, the formulation of optimal feeds is dependent on a thorough understanding of nutrient requirements. Although nutrient requirements of older, larger sturgeon are now being established (Stuart and Hung, 1989 ; Moore et al., 1988 ; Hung et al., 1987), the early life stages, particularly those prior to metamorphosis, pose three problems which make nutrition studies difficult at best. Foremost is the small size of first feeding sturgeon which necessitates the use of small particle sizes that are subject to high rates of leaching. Second, food location and ingestion by sturgeon, regardless of age, is not immediate, thereby compounding the leaching problems. Hence, by the time the feed is ingested, unknown quantities of the original nutrients have been lost preventing accurate identification of the requirements. As a result, nutrient requirements established using feeding studies will not reflect the actual needs of the animal. Instead, such "requirements" will overestimate actual requirements because of the need to include excess nutrients in the diet in order to compensate for leaching losses prior to ingestion. Finally, even though it is now possible to rear larval sturgeon on formulated feeds, their highly selective feeding habits can limit the use of purified and semipurified feeds.

Despite the above difficulties, patterns of yolk utilization, studies of metabolism, and observations of natural feeding habits do allow for speculation about some aspects of the nutritional physiology during the early stages of sturgeon development.

The embryonic period (from fertilization to hatch)

During the first half of the embryonic period rates of oxygen consumption remain relatively constant, with a gradual increase during the second half up to hatch (Dabrowski and Kaushik, 1986), suggesting increasing metabolic activities. This contrasts with the constant post-fertilization increase in oxygen consumption typical of most other species (Boulekbache, 1981).

During embryonic development protein seems to be principal energy source. Whereas levels of protein in developing sturgeon (yolk and tissues) gradually decline at a constant rate throughout development up to yolk exhaustion, lipid levels do not change during embryonic development but begin to decline steeply after hatch (Wang et al., 1987). Corroborating these findings are the metabolic data of Dabrowski and Kaushik (1986) and Khakimullin and Molodyky, (1985) for the Siberian sturgeon

(*A. baeri*). Corresponding with the constant decline in protein, rates of ammonia excretion remained relatively constant during the embryonic and larval phases. The two peaks in ammonia excretion at fertilization and hatch were respectively attributed to the post-fertilization hydration of the chorion and to the release of accumulated nitrogenous wastes at the time of hatch when the chorion is shed.

Based on rates of ammonia excretion, Dabrowski and Kaushik (1986) estimated the amount of yolk protein utilized for energy production during the embryonic period would not exceed 13% of the original protein stores. Since protein catabolism may result in the production of other metabolites (e.g., urea, ornithine and other amino acids), the data of Dabrowski and Kaushik (1986) may underestimate the contribution of protein toward energy production during the embryonic period, if other nitrogenous wastes were being formed. However, the potential error is probably slight in light of the small amounts of the alternative nitrogenous wasteproducts.

Larval non-feeding phase

Similar to most fish species, sturgeon larvae are not fully developed at hatch and must undergo further development and differentiation before external foods can be ingested and digested. During this period of posthatch development the larvae remain dependent on yolk reserves. Coinciding with the release from the eggs and advent of free swimming, levels of activity and oxygen consumption increase dramatically after hatch (Dabrowski and Kaushik, 1986). It is during this time sturgeon are extremely sensitive to oxygen deficits (Khakimullin, 1987).

Curiously, although the decline in protein remains constant throughout development (Wang et al., 1987), the post-hatch period, particularly during the first half of the larval non-feeding phase, is characterized by rates of ammonia excretion that are higher than during embryonic development (Yarzombrek and Maslennicova, 1971 ; Dabrowski and Kaushik, 1986). This is suggestive of greater protein catabolism, yet it is not paralleled by a greater rate of decline in the amounts of protein. The reason for the elevated excretion of ammonia remains unexplained. As the yolk becomes depleted during the second half of this phase, rates of ammonia excretion decline until the extrusion of the melanin plug and onset of active feeding when nitrogen excretion once again increases (Dabrowski and Kaushik, 1986).

Together, the metabolic data and changes in proximate composition imply during embryonic development a larger proportion of the low energy requirements are met by the catabolism of protein. After hatch, when metabolism increases due to increased activity (i.e., swimming) and increasing quantities of metabolically active tissues, a larger proportion of the energy requirements are provided by the yolk lipids. This strategy would spare yolk proteins for tissue synthesis. Of the yolk lipids, lipoproteins are probably utilized more so than lipids associated with the oil globule. This speculation is based on data from the rainbow trout (Nakagawa and Tsuchiya, 1972) and observations of oil globule retention within the gut of larval sturgeon.

The above descriptions indicate the general trends of nutrient utilization during the period of yolk dependency are now partially understood. Furthermore, they are similar to the patterns of nutrient and energy utilization during early development of other fish species (Heming and Buddington, 1987). However, studies of the metabolic characteristics of fish embryos and yolk-dependent larvae remain scarce. This leaves several important questions unanswered. For example, what is the relative importance and differential utilization of the various classes of lipids (e.g., phospholipids, triglycerides, cholesterol) and proteins (e.g., proteins, glycoproteins, lipoproteins) present in the yolk. Furthermore, the potential role carbohydrates might play in the metabolism of early life stages has not been adequately considered. Though the quantities of carbohydrate existing at any one time during development are not substantial, this does not preclude the possibility of continual production and utilization of carbohydrate originating from other nutrients via gluconeogenic pathways. If so, carbohydrates may prove to be a metabolite of some, though yet unknown, importance. This needs to be addressed in greater depth in light of the parallel increases in levels of activity, oxygen consumption, and increased activities of enzymes

associated with glycolysis and gluconeogenesis, indicative of increased carbohydrate metabolism which have been observed during development of other larval fish (reviewed by Boulekbache, 1981). Furthermore, enzyme systems associated with carbohydrate metabolism are known to exist as early as gastrulation in developing sturgeon hybrids, and persist throughout development (Chikhachev and Tsvetnenko, 1983).

Perhaps what may prove to be one of the more interesting and potentially most productive areas of research will involve the role of maternal nutrient investment into the eggs. It is common to observe significant interbrood variability with respect to size and composition of eggs (Krivobok and Storozhuk, 1970), rates of oxygen consumption, ammonia excretion (Dabrowski and Kaushik, 1986), growth and survivorship (Wang et al., 1985). Though the age and size of the female is known to influence the quality and quantity of eggs produced by sturgeons and other species, the influence of maternal nutrient status on egg quality and subsequent growth and survival of the embryos and larvae is less well known. The potential influence of the maternal diet on egg composition, however, is evident from several studies (Watanabe et al., 1984, Mommsen and Walsh, 1988, Knox et al., 1988). Briefly, by manipulating the maternal diet, it is possible to influence the composition of the eggs. This can have profound influences. Leray et al., (1985) demonstrated rainbow trout broodstock fed diets deficient in essential fatty acids produced eggs of lower quality, based on higher mortality and developmental abnormalities. Similar findings were reported by Watanabe et al. (1984) for Red Sea bream fed diets with different levels of protein and phosphorus. Hence, information for maternal nutrient investment into the eggs may prove instrumental in the development of feeds formulated specifically for broodstock.

The larval feeding phase and later development.

As the yolk is depleted, larval sturgeon, as with other fish species, must begin to ingest external foods to satisfy increasing energy demands, and to provide nutrients for growth. Corresponding with their high energy and nutrient requirements, virtually all larval fish are carnivorous, regardless of adult feeding habits, and thereby consume a relatively nutrient dense diet. First feeding larval sturgeon are zooplanktivorous with a diet consisting primarily of cladocerans and copepods (Baranova and Miroshnichenko, 1969).

In light of the difficulties associated with establishing nutrient requirements by feeding studies (see above), the proximate composition of the natural larval foods might provide some initial insights as to larval nutrient requirements. The high rates of growth and survivorship of larval sturgeon fed zooplanktivorous diets (Milstein, 1972) indicate such diets satisfy nutrient and energy requirements. Therefore, when one considers zooplankton often contain high quantities of lipid (sometimes over 50% of the dry weight ; Tessier and Goulden, 1982) and the high rates of yolk lipid utilization prior to yolk depletion, it is conceivable that larval sturgeon diets should contain up to 50% fat (Buddington and Christofferson, 1985).

Coinciding with rapid growth, rates of protein metabolism are also high during the early external feeding phase (Fauconneau et al., 1986). The rates of metabolism, however, are acutely sensitive to quality of protein. As a result, larval sturgeon fed diets containing lower quality protein exhibit higher rates of nitrogen excretion, lower growth, and lower survival. Hence, it can speculated that diets formulated for larval sturgeon should be prepared with high quality proteins.

DIGESTIVE PHYSIOLOGY

Sturgeons undergo three phases of digestive system development, with all species exhibiting a similar pattern (Korzhuev and Sharkova, 1967 ; Plotnikov and Proskuryakov, 1984 ; Buddington, 1985 ; Buddington and Doroshov, 1986 ; and reviewed by Buddington and Christofferson, 1985).

Phase I : Differentiation of the digestive system

When sturgeon hatch they possess a primordial digestive system. During the period of yolk nutrition, the digestive system differentiates, developing many of the adult characteristics. When exogenous feeding is initiated sturgeon possess a functional stomach capable of producing both acid and pepsin. This differs dramatically from other fish species which are "stomachless" when the first foods are ingested and do not acquire gastric capabilities, if they possess a functional stomach as adults, until they metamorphose to the juvenile stage.

Phase II : The larval feeding phase

Once external foods are consumed, whether they be live foods or prepared diets, there is a rapid increase in the concentrations of all digestive enzymes. Of particular note are the high concentrations of amylase and lipase, being higher during the larval feeding phase than at any other stage of development. Though the high lipase activity coincides with the ingestion of lipid-rich zooplankton, the reasons for high amylase activity are unexplained. At conclusion of the larval feeding phase, when the larvae metamorphose and assume the adult feeding habits, activities of both amylase and lipase decline. Throughout the larval feeding phase there is a steady increase in the secretion of trypsin and chymotrypsin into the intestine, and pepsin and acid by the stomach.

Despite the presence of increasing concentrations of proteases, the data of Dabrowski et al. (1987) indicate the ability to digest protein, particularly low quality protein, may be limited. Unfortunately, even though sturgeon larvae are large compared to larvae of other fish, it has not yet been possible to determine the digestive capacities of larval sturgeon.

Phase III : Post-metamorphosis

After juvenile sturgeon have fully assumed the adult feeding habits, the digestive physiology remains relatively constant thereafter. For the white sturgeon (*A. transmontanus*), the digestive enzyme complement (Buddington and Doroshov, 1986) and the rates of nutrient transport by juveniles (Buddington et al., 1987) correspond with this species' omnivorous feeding habits.

The lack of dietary influences on the patterns of digestive enzyme secretion described above demonstrates the existence of a genetically hard-wired program of digestive physiology development. Moreover, the developmental program coincides with the evolutionary zooplanktivorous diet and therefore "preadapts" the larvae for anticipated changes in dietary inputs. It is possible that by understanding the changes in digestive physiology during ontogenetic development we may gain still further insights into the possible nutrient requirements of developing sturgeon and thereby be better able to design larval diets with nutrient compositions that track the genetically programmed changes in digestive secretions.

RESPONSES TO CHANGES IN ENVIRONMENTAL CONDITIONS

An organism's physiology is intricately interrelated with the prevailing environmental conditions. Consider how the physiological functions of ectotherms are profoundly influenced by changes in environmental temperatures. Hence, ectotherms exposed to changing temperatures, as well as other environmental conditions, must be able to adapt a multitude of physiological processes if they are to survive. Such adaptation is evident from the manifold adaptations fish exhibit when switched to environments that differ with respect to salinity, temperature, pH, or other parameters. Only by possessing the ability to compensate for changing environments, are fish able to enter new environments, or adapt to seasonal changes.

However, not all species are able to tolerate changes in environmental conditions. For examples, many stenohaline and stenothermal species are unable to adapt to changes in salinity and temperature. Such species typically have been restricted throughout their phylogeny to environments with constant conditions (e.g., deep-sea benthic fishes). Hence, there has been little selection for the abilities to adapt to changing conditions.

Similar to other ectothermic species, rates of development for sturgeons are dependent on water temperatures (Igumnova, 1985 ; Wang et al., 1985 ; Detlaf et al, 1981 ; Nikol'skaya and Sytina, 1978 ; Detlaf and Ginzburg, 1954).The temperature dependence of development includes physiological characteristics, such as the formation of and secretion by the interrenal gland (Barannikova et al., 1981). Although environmental conditions influence patterns of development, the early life stages of fish apparently lack, or at least have reduced abilities relative to older fish, to physiologically adapt to changes in environmental conditions. Exemplary of this is the ability of adult desert pupfish (Cyprinodon nevadensis) to survive temperatures ranging from 2 to 44°. Yet, successful embryonic development is restricted to a range of 22 to 34° (Shrode and Gerking, 1977).

Compounding the restricted temperature ranges, early life stages of most fish, including sturgeon, are less motile than older stages. They are thus less able to leave marginal habitats for more favorable conditions. Female pupfish apparently compensate for the limited temperature range and poor swimming abilities of the early life stages by restricting spawning activities to periods when suitable temperature conditions exist (Shrode and Gerking, 1977). The inability to leave marginal habitats is particularly true for the immotile eggs of the sturgeons. Once sturgeon eggs are spawned, fertilized, and become attached to a substrate, development of the embryo proceeds under the prevailing conditions, whatever they might be. Even after hatch, larval sturgeon are capable of only limited swimming. After metamorphosis sturgeon do acquire sufficient swimming abilities to enable them to leave one habitat for another. Hence, either the early life stages of sturgeon must be capable of adapting to changing environmental conditions, or the female must select and spawn within a habitat that is suitable for development of the early life stages.

In the following sections I review three sources of evidence which indicate the physiological responses of embryonic and larval sturgeon to changes in environmental conditions differ from those of older fish. Two sources involve the influence of temperature, the best studied environmental parameter, and the third source describes the influence of environmental contaminants.

Adaptation to temperature changes

Though chondrosteans are restricted to the northern hemisphere, representatives can be found in habitats ranging from freshwater to marine systems, from temperate to subarctic. Despite the wide diversity of adult habitats, spawning is restricted to freshwater, and usually within a temperature range of 14-16° (Detlaf et al., 1981, Wang et al., 1985). If spawning occurs outside of this range, survivorship declines. The ability to tolerate wider temperature ranges, and fluctuations within the tolerated range, increases as the sturgeon develop (Igumnova, 1985). Figure 1 presents a theoretical illustration of how the temperature range of sturgeons might increase during development. Hence, female sturgeons, like female pupfish, apparently limit spawning activities to periods when temperature conditions are suitable for development of the early life stages.

Older fish are able to compensate for changes in environmental temperature by two principal mechanisms : (1) behavioral strategies leading to the exploitation of more favorable habitats ; (2) development of physiologic characteristics matched to prevailing environmental conditions. In contrast, larval, and especially embryonic, fish may not be able to use these two adaptive mechanisms. Because of limited swimming abilities the early life stages are less able to behaviorally escape unfavorable environments. Furthermore, recent studies suggest early life stages of sturgeon lack the capacities to physiologically adapt in response to changes in environmental conditions.

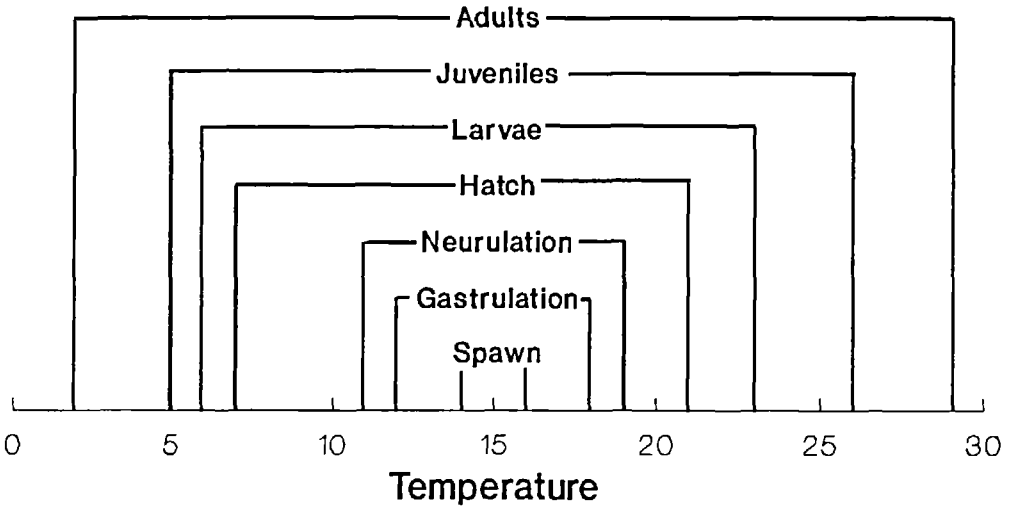


Figure 1 - Hypothetical tolerated temperature ranges for the different life history stages of a temperate water sturgeon.

One of the best studied examples of temperature adaptation is the ability of ectotherms to alter membrane phospholipid composition in response to changes in water temperature (Hazel, 1984). For the rainbow trout, as temperatures declines, the ratio of unsaturated to saturated fatty acids increases. The converse occurs with increasing water temperatures. This adaptive capability maintains a constant membrane fluidity throughout a range of temperatures, the homeoviscous response (Sinensky, 1971), and allows for the retention of the manifold and essential functions of cell membranes.

In contrast to the ability of adult trout to alter membrane phospholipid composition, Buddington et al., (1990) found the early life stages of white sturgeon (*A. transmontanus*), particularly the pre-neurulation stages, to have reduced capacities to regulate membrane phospholipid composition in response to changes in incubation temperature. Changes in lipid composition, though, are known to occur during early development of *A. guldenstadti* (Isuev and Magomedov, 1988). Although the fatty acid composition of membrane phospholipids of white sturgeon also change during development at natural spawning temperatures, when embryos are exposed to colder temperature the changes in composition were opposite to those predicted if homeoviscosity was to be maintained (Buddington et al., 1990). For example, the percentage of unsaturated relative to saturated fats decreased in the membrane phospholipids of embryonic sturgeon incubated at a temperature lower than normal (8° as opposed to 14°). These apparently non-adaptive responses coincided with higher mortality, particulary prior to neurulation, thereby agreeing with the findings of (Wang, 1984). In contrast, after hatch the ratio of unsaturated to saturated increased in the fish exposed to 8° and mortality was negligible, conforming with the anticipated homeoviscous response of other other fish (Hazel, 1984). The results of Buddington et al. (1990) are summarized in Table 1.

The reasons for the lower regulatory capacities of the early stages remain unknown. Although the costs of not being able to adapt are evident if environmental conditions should change, the developing fish do benefit by channelling energy and resources into growth, not adaptation.

Table 1 - Composition of membrane associated phospholipids at fertilization (eggs), at neurulation (N), at hatch (H), and one week after hatch (PH) of developing white sturgeon (*Acipenser transmontanus*) incubated at different, but constant, temperatures (from Buddington et al., 1990). A temperature of 14° is representative of natural conditions. Data are presented as means and standard errors and represent the percent of phospholipids represented by saturated and unsaturated fats, the unsaturation index (U.I.), and the average chain length. Also presented as arrows are predicted differences relative to the 14° treatment, if the developing sturgeon were able to regulate phospholipid composition to maintain membrane homeoviscosity. Results are based on analyses of samples originating from three separate broods. Due to low survival at 8°, material from only one brood was available at hatch and at posthatch.

Stage	Incubation Temp (C°)	Saturated	Unsaturated	U.I.	Chain length
Eggs	14	33.8 ± 1.3	66.2 ± 1.3	2.30 ± 0.06	18.62 ± 0.08
	Expected 8	↓	↑	↑	↑
	20	↑	↓	↓	↓
N	8	43.8 ± 2.5	56.2 ± 2.5	1.68 ± 0.11	18.21 ± 0.09
	14	37.8 ± 4.3	62.2 ± 4.3	2.11 ± 0.23	18.77 ± 0.22
	20	41.0 ± 1.8	59.0 ± 1.8	1.94 ± 0.19	18.52 ± 0.24
H	8	34.4	65.6	2.49	18.7
	14	26.5 ± 5.6	73.5 ± 5.6	2.95 ± 0.31	19.36 ± 0.38
	20	29.9 ± 4.1	70.1 ± 4.1	2.81 ± 0.20	19.23 ± 0.30
PH	8	30.7	69.3	2.55	19.01
	14	35.8 ± 1.2	64.2 ± 1.2	2.47 ± 0.08	18.85 ± 0.14
	20	37.4 ± 1.4	62.6 ± 1.4	2.33 ± 0.06	18.62 ± 0.09

Yolk utilization and temperature

The relationship between temperature and development is also evident from studies of changes in the composition of developing sturgeon incubated at different temperatures. Within the natural range of incubation temperatures, there is no influence of temperature on the pattern of yolk utilization (Wang et al., 1987). However, temperatures outside of the natural range lead to changes in the patterns of utilization. When eggs are incubated at elevated temperatures survival is reduced, and the larvae that do hatch have lower levels of lipid and protein, lower caloric content, and weigh less than eggs incubated at lower temperatures (Wang et al., 1987 ; Dabrowski and Kaushik, 1986).

Tolerance to pollutants

An area of research that will require further work involves the influence of environmental contaminants on development. Although managers of hatcheries are often able to manipulate most of the water conditions, it is much more difficult to control the quality of the water. Yet pollutants are posing increasing problems in hatchery and natural settings.

Pollutants can influence development in three principal ways. First, they can lead to the death of the developing fish. Second, abnormalities of development can result from exposure to a pollutant. Finally, development can be delayed. The actual response is dependent on the type pollutant and the dose. It should be noted, the

specific stages that are most sensitive to specific pollutants varies between species of teleost, and type of pollutant (Westernhagen, 1988).

Though there have been several studies of the responses of different species of sturgeon to a variety of pollutants (e.g., Zemkov and Teplyy, 1979), much less is known about how pollutants might influence development. Tikhonova and Shekhanova (1983) demonstrated early life stages of *A. guldenstadti* are more sensitive to carbamates than older fish with the most critical stages of development at or before gastrulation. Regardless of species or pollutant, resistance increases with further development, such as the increasing resistance of *A. guldenstadti* to carbamates as they grow. Development of the sturgeon digestive system is also known to be sensitive to pollutants (Schmal'gauzen, 1972). Specifically, exposure to phenol delays digestive system development which could have profound influences on the digestive capabilities at first feeding.

CONCLUSIONS

Previous studies of sturgeon development have focused on the patterns of development. Future studies need to address the mechanisms underlying the patterns of development (e.g., synthesis of new proteins, activation of existing proteins, and the differential expression of genetic information over time and from cell to cell). Also requiring attention are the signals which trigger the patterns of development (e.g., external signals, such as changes in environmental conditions, or internal signals such as hormones ; Diamond, 1987).

Sturgeon provide an interesting opportunity to study ontogenetic development. Similar to other species, rates of sturgeon development and patterns of yolk utilization are temperature dependent. Unlike other fish, sturgeon undergo holoblastic cleavage, possess an intraembryonic yolk endoderm, and have a functioning stomach at the onset of active feeding. Hence, care must be exercised when extrapolating results obtained from other species to how environmental conditions might influence the development of sturgeons.

During the embryonic period, especially up to gastrulation, sturgeon are most sensitive to environmental conditions. Corresponding with this, highest levels of mortality occur during early development of sturgeon. With increasing development, sturgeon acquire the ability to adapt to changes in environmental conditions and exposure to environmental contaminants, correspondingly, mortality associated with environmental perturbations decreases. Improvement of the hatchery management of sturgeons will be enhanced by increasing our understanding of sturgeon development, the physiological characteristics of the different stages of development, and the associated influences of environmental conditions. Another area requiring attention is the design of diets formulated specifically for the different life history stages of sturgeon, including the broodstock. Conventional methods of quantifying nutrient requirements and efforts to develop diets specifically for the different life stages of sturgeon will be aided by a better understanding of the digestive and metabolic physiology of actively feeding sturgeon, as well as the nutrient characteristics of the natural foods.

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NUTRITION AND FEEDING OF HATCHERY-PRODUCED JUVENILE WHITE STURGEON (*Acipenser transmontanus*) : AN OVERVIEW

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RESUME

Un aliment purifié a entraîné une croissance égale ou légèrement supérieure chez l'esturgeon blanc impubère (de 10 à 200 g) à celle obtenue avec un aliment sec pour salmonidé couramment utilisé dans l'élevage de l'esturgeon en Californie. Cette formulation a été utilisée pour déterminer les besoins de l'esturgeon en cholestérol, lécithine, choline, et protéines. Nous n'avons trouvé aucun besoin en cholestérol et en lécithine, par contre 0,17-0,32% de choline et 40% de protéine brute de l'aliment distribué étaient nécessaires pour assurer une bonne croissance du jeune esturgeon blanc. La formulation avait également pour but de déterminer l'aptitude de l'esturgeon à utiliser des protéines et des glucides différents. Un mélange de protéines (caséine:glutène de blé:blanc d'oeuf 62:30:8) et la caséine se sont révélées être les meilleures protéines, alors que le glucose et le maltose étaient les meilleurs glucides. Une étude supplémentaire basée sur cette formulation a été conduite pour déterminer le niveau optimal de consommation pour l'esturgeon blanc impubère. Dans la phase de croissance entre 15 et 150 g, la consommation optimale de l'esturgeon blanc s'est avérée être de 2,0% du poids vif par jour à une température de 20°C.

Mots clés : *Acipenser transmontanus*, alimentation, éléments nutritifs, besoins.

ABSTRACT

A purified diet has been shown to support growth of juvenile white sturgeon (10 to 200 g) similar to or slightly better than a dry commercial salmonid feed commonly used in California sturgeon farms. This purified formulation was used to determine for the cholesterol, lecithin, choline, and protein requirements of sturgeon. We found no dietary requirement for cholesterol and lecithin, but 0.17-0.32% of choline and 40% of crude protein were required by juvenile white sturgeon for good growth. The formulation was also used to determine the ability of sturgeon to utilize different proteins and carbohydrates. A protein mixture (casein:wheat gluten:egg white 62:30:8) and casein were the best proteins, whereas glucose and maltose were the best carbohydrates. An additional study was conducted using this formulation to determine the optimum feeding rate of juvenile white sturgeon. The optimum feeding rates for 15 to 150 g white sturgeon was 2.0% of their body weight per day at 20°C.

Key-words : *Acipenser transmontanus*, feeding, nutrients, requirements.

INTRODUCTION

Before 1900, the annual sturgeon catch in the USA was estimated to be 6.8 million kg on the East Coast and 4.6 million kg on the West Coast (Rostlund, 1952). By the turn of the century, extensive commercial sturgeon fisheries had been established on both Coasts, primarily for the export of flesh and caviar to Western Europe. Soon after that the U.S. sturgeon fisheries declined rapidly as a result of overfishing and poor fishery management (Doroshov, 1985). From 1920 to 1950 the decline of wild sturgeon stocks was further aggravated by destruction of their spawning grounds and habitats by damming of rivers and eutrophication of their environment (Craig and Hacker, 1940 ; Skinner, 1962). Stocks of at least three important species of sturgeon, the Atlantic, lake, and white sturgeon, were almost depleted.

The mean annual catch of white sturgeon in the Pacific Northwest from 1968 to 1983 was 108,000 kg, whereas Atlantic sturgeon in the southeast coastal region from 1971 to 1982 was 73,000 kg (Appendix 2, Conte et al., 1988). These reported landings may represent conservative estimates because landings from smaller fisheries are not

included. Nevertheless, this limited data showed how severe was the loss of wild sturgeon stocks in the USA. This loss is of particular concern today, because of the growing popularity of sturgeon as a foodfish and increasing desire to develop the recreational and commercial sturgeon fisheries.

Early attempts to propagate sturgeon artificially in the USA were unsuccessful (Leach, 1920). The first successful artificial spawning of wild sturgeon outside the USSR was achieved with white sturgeon in 1979 by Dr. S. I. Doroshov, Department of Animal Science, University of California, Davis (Doroshov et al., 1983). Since then there has been a resurgence of interest to establish sturgeon culture for mitigation of natural resources and for commercial foodfish production. The hatchery technology has expanded rapidly and a comprehensive sturgeon hatchery manual has been published (Conte et al., 1988).

At present, California has about 47 registered white sturgeon growers, and 11 farmers working to develop domesticated broodstock. Several farmers are marketing 3.5-5.5 kg white sturgeon as foodfish two to three years of age. A projected 140,000 kg of cultured white sturgeon will be entering the U.S. market in 1988-89 with a wholesale value of 1.3-1.8 million dollars. Most cultured white sturgeon currently are processed as either dressed, gutted with head and tail attached, steak, or as a saddle (Dr F.S. Conte, pers.com.). Processing yields were : 85-96% eviscerated, 53-65% dressed, 61-74% steaked, 54-61% skin-on fillets, and 32-50% skinless fillets in sturgeon between 0.25 and 3 kg (Price et al., 1989). The proximate compositions of carcass and edible portion of these fish were : 72-80% moisture, 12.0-21.3% protein, 0.4-7.4% fat, and 1.0-1.7% ash (Hung et al., 1987a ; Price et al., 1989).

Very little information is available on the nutrient requirements and optimum feeding rates of white sturgeon, especially under hatchery conditions (Lane, 1985). The lack of information on sturgeon nutrient requirements has been cited as a major limiting factor for further development of sturgeon aquaculture (Conte et al., 1988). The lack of a suitable commercial sturgeon feed has forced farmers to use salmonid feeds for their sturgeon. This feeding practice is known to be inadequate because prolonged feeding of white sturgeon with salmonid feeds sometimes has resulted in poor growth and abnormal symptoms including scoliosis, ataxia, and other complications which are suspected of being nutritionally related (personal observation).

Development of a suitable purified diet

Nutritional requirements of a species of fish are often determined with a purified diet specifically developed for the species (Castell and Tiews, 1980). The purified diet should at least provide the fish with a growth rate comparable to those fed the best available commercial feed. We conducted our first experiment to compare the growth performance of juvenile white sturgeon fed a purified test diet (similar to 79F, Conklin et al., 1980) to those fed a commercial salmonid dry diet (Hung et al., 1987b).

Percent body weight increase (%BWI = 100 x (final weight - initial weight)/initial weight) of sturgeon fed the purified diet (344±46%) for eight weeks was as good or slightly better than those fed the salmonid diet (210±39%) (recalculated from Hung et al., 1987b). Therefore, this purified diet (SPD) is suitable for nutritional and feeding studies of juvenile white sturgeon.

Lecithin and choline requirements

The SPD diet contained two unusual ingredients, 0.5% cholesterol and 8.0% refined-soy lecithin. A second experiment was conducted to determine the effects of replacing the 0.5% cholesterol or 8.0% refined-soy lecithin with an equal amount of the oil mixture on growth of white sturgeon (Hung et al., 1987b). Eight-week %BWI of sturgeon fed SPD diet with its cholesterol replaced by an equal amount of the oil mixture (SPD-C) was 174±19%. This was not different from those fed the SPD (183±31%). Sturgeon fed a diet with the refined-soy lecithin replaced by an equal amount of the oil

mixture (SPD-L) grew significantly ($P<0.05$) less (23±3%) than those fed either the SPD or SPD-C diets both of which contained 8% refined-soy lecithin. These results suggested that sturgeon did not require cholesterol but may require refined-soy lecithin for good growth.

Lecithin is known to be required by some crustaceans but not by finfish, whereas choline is known to be required by many species of finfish (NRC, 1981 ; 1983 ; Millikin, 1982). Careful examination of the SPD diet revealed that the vitamin premix (Hung et al., 1987b) used did not contain any choline supplement but the 8.0% refined-soy lecithin, however, contained 0.24% choline. A subsequent study (Hung and Lutes, 1988) was conducted using a 2 x 2 factorial design (0 or 8% refined-soy lecithin and 0 or 0.8% choline chloride supplements) to determine the essential requirement of these two supplements in juvenile white sturgeon.

Eight-week %BWI and percent feed efficiency (%FE) showed very similar pattern. They were significantly ($P<0.05$) affected by each of the two supplements and the interaction was also significant (Hung and Lutes, 1988). The lower growth (Figure 1) and feed efficiency of fish fed the diet without refined-soy lecithin and choline compared to those fed diets with either or both supplements suggest these two supplements may be required by sturgeon for good growth. There was, however, little or no additive growth-promoting effect of the two supplements. This suggests only one of the above supplements was required. Choline is a moiety common in both choline chloride and phosphatidyl-choline which was present in the refined-soy lecithin. This suggested that the growth promoting effect of refined-soy lecithin was mostly resulted from the choline in phosphatidyl-choline rather than the refined-soy lecithin per se.

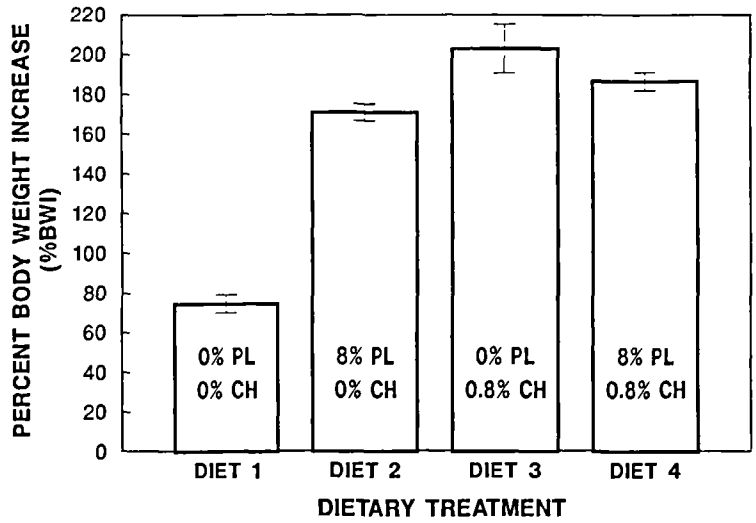


Figure 1 - Percent body weight increase of sturgeon fed diets with 0 or 8 % refined-soy lecithin (PL) and 0 or 0.8 % choline chloride for 6 weeks.
Values are means ± SE for 3 replicates per treatment.

In a follow up experiment, sturgeon were fed for eight weeks one of the eight diets which were supplemented with 0 or 8% refined-soy lecithin and 0, 0.2, 0.4, or 0.8% choline chloride (Hung, 1989). Eight week %BWI (Figure 2), %FE, and total plasma lipid levels had a very similar pattern. They were significantly ($P<0.05$) affected by each of the two supplements and the interactions were also significant for the measurements. Sturgeon fed the diet without both supplements had an extensive, diffused fat vacuolation and fatty cyst formation in their liver. In the absence of refined-soy lecithin

supplement, the growth, feed efficiency, and total plasma lipid levels were affected significantly ($P<0.05$) by the choline chloride supplements. Results in this experiment suggested that there is no requirement for refined-soy lecithin, but 0.17-0.32% choline/kg diet (30 to 60 mg/kg body weight/day) was required by juvenile white sturgeon for good growth. Choline requirement of sturgeon did not seem to be affected by the dietary methionine because our diets contained adequate amount of methionine (1.14%).

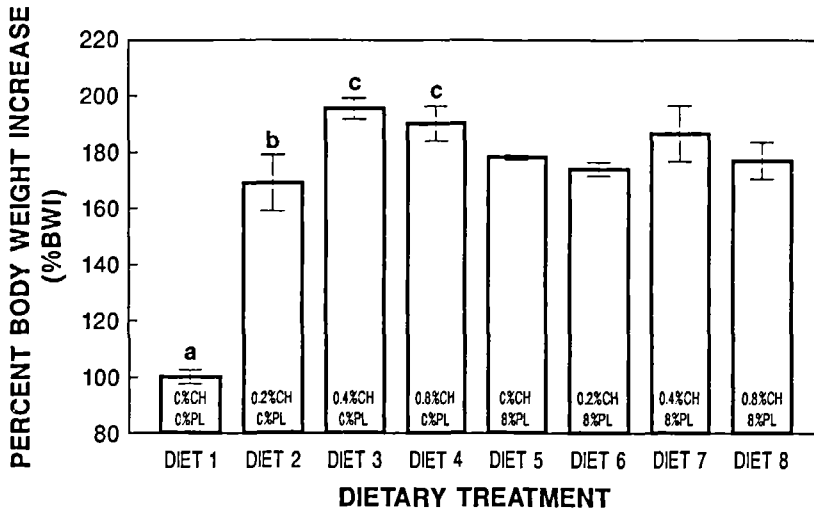


Figure 2 - Percent body weight increase (%BWI) of sturgeon fed diets with different levels of refined-soy lecithin (PL) and choline chloride (CH) for 8 weeks.
Values are means ± SE for 3 replicates per treatment. Only means for sturgeon fed diets 1 to 4 were compared and means with different superscripts are significantly different ($P < 0.05$). There was no significant difference between sturgeon fed diets 5 to 8.

Choline requirement of juvenile white sturgeon is similar to common carp (Ogino et al., 1970), higher than lake trout (Ketola, 1976), and much higher than channel catfish (Wilson and Poe, 1988). The different choline requirements may have resulted from the different experimental diets and conditions in these studies. Alternatively, the difference may have resulted from the different ability of these fish to synthesize choline from methionine by the S-adenosylmethionine pathway as observed in other animals (Kuksis and Mookerjea, 1984). This alternative is further supported by a recent report (Wilson and Poe 1988) showing that channel catfish did not require choline if adequate methionine was included in the diet.

Protein requirement and quality

Protein is the most expensive ingredient in fish feeds because fish usually require a very high level of good quality protein (NRC, 1981 ; 1983). Protein deficiency is known to have a profound effect on the growth of animals. Therefore, information on protein requirement of juvenile white sturgeon is very important for the development of a cost-effective sturgeon feed.

An eight-week growth experiment was conducted to determine the dietary protein requirement by juvenile white sturgeon (Moore et al., 1988). In this experiment, juvenile white sturgeon were fed one of the eight purified diets which contained from

20.0 to 52.7% crude protein. Protein of these diets was supplied with graded amounts of the control protein (casein : wheat gluten : spray-dried egg white, 62:30:8).

Eight-week %BWI, %FE, protein deposited (%PD), and energy retained (%ER) showed a high degree of agreement and were equally sensitive to the different levels of dietary protein. They were significantly ($P<0.05$) affected by the dietary protein content. Protein requirement of juvenile white sturgeon on an as fed basis was estimated from the percent body weight increase using second order polynomial regression analysis (Zeitoun et al., 1976). The regression analysis (Figure 3) showed that the minimum range of protein requirement was between 36.5 and 40.5%. Based on these results, a 40% crude protein on an as fed basis was recommended for juvenile white sturgeon.

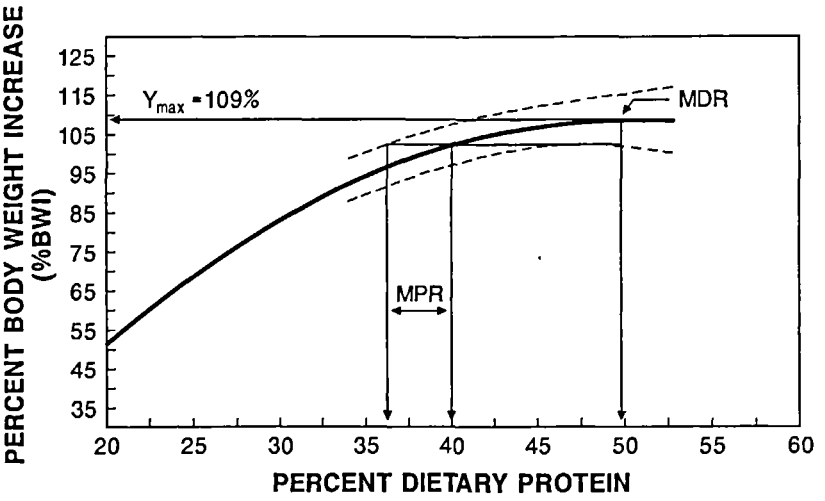


Figure 3 - Second order polynomial relation (solid curve line $Y = -51.89 + 6.49X - 0.07X^2$) and 95 % confidence limits (dashed curved lines) of percent body weight increases of sturgeon and percent dietary protein.
MPR = minimum range of protein requirement, MDR = maximum of dose response curve.
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Protein requirement of a fish is determined usually by feeding the fish with graded levels of a high quality protein. Casein, egg albumin (egg white), fish muscle, and fish meal have been used as high quality proteins to determine the requirements of several species of fish (NRC, 1983 ; Wilson and Halver, 1986). The quality of our control protein mixture is unknown ; thus, it is impossible to compare the protein requirement of white sturgeon to other species of fish. Furthermore, the lack of information on the quality of the commonly used feed proteins makes formulation of a cost-effective sturgeon feed difficult.

A growth trial was conducted to determine the performance of white sturgeon fed purified diets with different proteins (Stuart and Hung, 1989). These diets were isoenergetic, isonitrogenous, and contained one of eight different proteins as the sole protein source. Eight-week %BWI, %FE, %PD, and %ER of sturgeon were significantly ($P<0.05$) affected by the different dietary proteins. A high degree of agreement also existed between these measurements as observed in the previous study (Moore et al., 1988). Among the four measurements, %ER (Figure 4) appeared to be the most sensitive index because it gave the greatest degree of separation between the different proteins. The higher sensitivity of %ER over %PD was attributable mainly from the greater

differences in body lipid than protein of sturgeon fed the different proteins. Based on the %ER, their quality was : control = casein > defatted shrimp meal > defatted herring meal > soybean concentrate > egg white > gelatin > defatted zein meal.

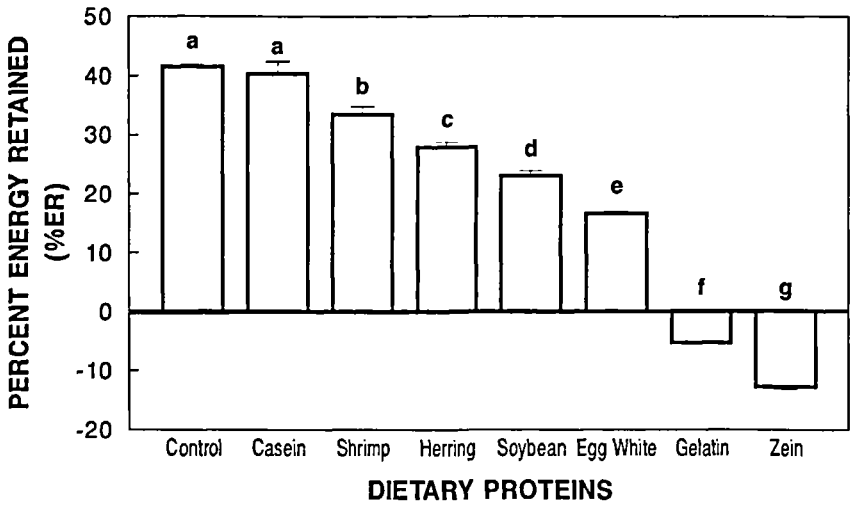


Figure 4 - Percent energy retained of sturgeon fed different proteins for 8 weeks.
Values are expressed as mean ± SE for 3 replicates per treatment. Means with different superscripts are significantly different (P<0.05).

The quality of our control protein was as good or better than those proteins used to determine the requirement of other species of fish. Based on these results, a 40% dietary crude protein, on an as-fed basis, from a good quality source is recommended for the optimum growth of juvenile white sturgeon. The 40% protein requirement of white sturgeon is similar to most other fishes but lower than Japanese eel, puffer fish, red sea bream, and smallmouth bass (NRC, 1981 ; 1983). The protein requirement of juvenile white sturgeon is similar to the 36-42% reported for Siberian sturgeon (*Acipenser baeri*) (initial body weight 90 to 150 g) (Kaushik et al., 1989).

Ability of sturgeon to utilize different carbohydrates

The ability of fish to utilize different types and levels of carbohydrates is different among species (Millikin, 1982). No information existed on the ability of white sturgeon to utilize different carbohydrates. There is also no information on the maximum levels of different dietary carbohydrates without depressed growth in juvenile white sturgeon. This information is essential for the development of a cost-effective commercial sturgeon feed because carbohydrates are the cheapest dietary energy source. Adequate levels of dietary carbohydrate are known to spare the more expansive dietary protein and lipid.

We recently completed an experiment to compare the ability of juvenile white sturgeon to utilize different carbohydrates (Hung et al., 1989a). In this experiment sturgeon were fed isonitrogenous diets containing 27.2% of one of the eight carbohydrates. The eight carbohydrates were glucose, fructose, maltose, sucrose, lactose, dextrin, raw cornstarch, or cellulose.

Eight-week %BWI, %FE, %PE, and %ER were significantly (P<0.05) affected by the different dietary carbohydrates (Hung et al., 1989a). Again percent energy retained (Figure 5) appeared to be the most sensitive index to assess the ability of sturgeon to utilize different dietary carbohydrates. The greater sensitivity of this measurement was

also resulted from the greater differences in the body lipid of sturgeon fed the different carbohydrates.

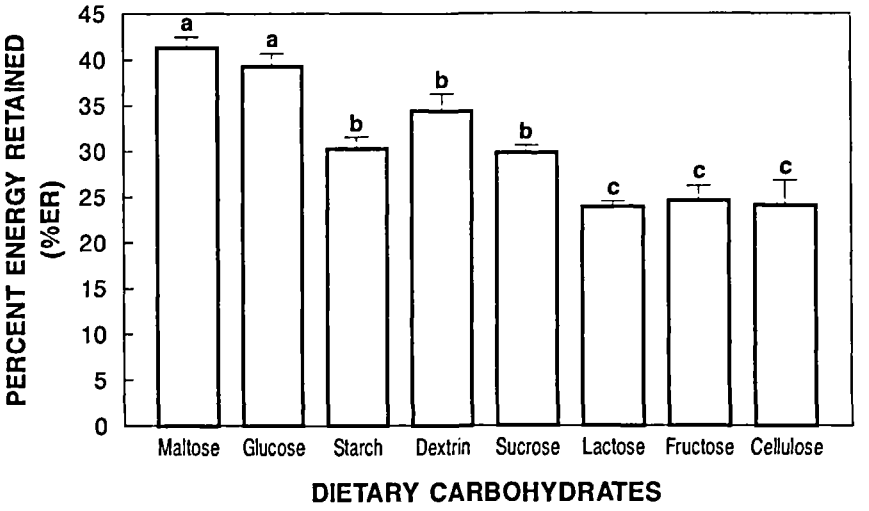


Figure 5 - Percent energy retained of sturgeon fed different carbohydrates for 8 weeks. Values are expressed as mean ± SE for 3 replicates per treatment. Means with different superscripts are significantly different ($P<0.05$).

Based on the %ER, the ability of sturgeon to utilize different dietary carbohydrates in decreasing order was : glucose = maltose > raw cornstarch = dextrin = sucrose > lactose = fructose = cellulose. The poor utilization of either raw cornstarch or dextrin by sturgeon may have been the result of the low α-amylase activity similar to that of rainbow trout (Spannhof and Plantilow, 1983). The poor ability of white sturgeon to utilize fructose may have been the result of poor absorption as observed in channel catfish (Wilson and Poe, 1987). The poor utilization of either sucrose or lactose was primarily due to the low intestinal sucrase and lactase activities in the sturgeon (Hung et al., 1989 a).

Histological examination (Hung et al., 1989b) showed that sturgeon fed either the glucose, maltose, dextrin, or raw cornstarch diets had normal livers histologically, whereas sturgeon fed either the sucrose, lactose, or fructose diets had a moderate condensation of the hepatocyte cytoplasm and a decreased number of glycogen containing intracytoplasmic vacuoles. The distal intestines of sturgeon fed either the sucrose, lactose, or fructose diets contained a significantly ($P<0.05$) higher luminal water content. In these sturgeon, the cytoplasm in the mucosal epithelial cells of the distal intestines were occupied by a few large, irregular, and nonglycogen-containing vacuoles. The increased luminal water content in the distal intestines of sturgeon fed either sucrose, lactose, or fructose diets was similar to disaccharide-induced diarrhea observed in higher vertebrates (Monson et al., 1950 ; Morris et al., 1977 ; Mayes, 1988). The sucrose, lactose, or fructose induced diarrhea in juvenile white sturgeon was resulted from the low intestinal sucrase, lactase, or low absorption of fructose, respectively.

Growth responses of sturgeon fed either glucose, maltose, dextrin, or raw cornstarch were similar to those reported for chinook salmon (Bulher and Halver, 1961), except that salmon utilize sucrose better than sturgeon. The better growth in the sucrose-fed chinook salmon may have resulted from the higher sucrase than maltase activities in these fish, whereas the opposite is true in white sturgeon (Hung et al., 1989a).

We observed better growth rates in white sturgeon fed either maltose or glucose than those fed the dextrin or starch diets. This suggested that α-amylase activity may be the limiting factor for the digestion of dextrin and raw cornstarch. Other investigators reported that common carp (Furuichi and Yone, 1982) and channel catfish (Wilson and Poe, 1987) fed either starch or dextrin had better growth than those fed glucose, whereas red sea bream fed these carbohydrates displayed no growth differences (Furuichi and Yone, 1982).

Our results cannot be compared directly with those of Kaushik et al., (1989) who fed Siberian sturgeon with 38% of either crude, gelatinized, or extruded starch. These authors showed that Siberian sturgeon can utilize gelatinized and extruded starch better than crude starch. This observation agreed with our results that carbohydrate utilization of white sturgeon decreased with increasing molecular weight or increased with increasing hydrolysis.

The different ability of various species of fish to utilize different carbohydrates may have been the result of methodology or species differences in the present and previous studies. The important methodology differences include diet formulations, water temperatures, and feeding strategies. The important species differences include digestion, absorption, hormonal response, metabolic efficiency, and gastrointestinal anatomy and physiology of different species of fish.

Optimum feeding rates of juvenile white sturgeon

Information on the optimum feeding rates is important to fish culturists because overfeeding leads to feed waste and, thus, increase feed costs. Overfeeding also leads to poor water quality which is known to increase other production costs by decreasing growth rate and feed efficiency but increasing the susceptibility of fish to diseases and mortality. Poor water quality also reduces the carrying capacity of a farm and, thus, reduces its production efficiency. There is, however, very little information on the optimum feeding rates of sturgeon.

We have conducted a growth experiment (Hung and Lutes, 1987) to determined the optimum feeding rate of juvenile white sturgeon at 20°C. We selected this water temperature because it is common in most of the California sturgeon farms. In this study, sturgeon were fed from 0.5 to 4.0% (at 0.5% increments) body weight per day (BW/day) of a purified diet.

Eight-week %BWI and feed gain ratio were significantly ($P<0.05$) affected by the different feeding rates (Hung and Lutes, 1987). The percent body weight increase showed an asympotitic curve similar to those growth curves derived from the nutrient requirement studies of mammals (Robbins et al., 1979) and fishes (Zeitoun et al., 1976). The optimum feeding rate based on the percent body weight increase (Brett, 1979) was 2.0% BW/day.

Potential of white sturgeon aquaculture

Results from the above studies demonstrate that body composition of white sturgeon was closely related to the growth rate. Sturgeon with good growth usually had higher body lipid and lower moisture contents than those with poor growth. Body protein and ash were less affected unless growth was severely depressed. These relationships have also been observed in largemouth bass (Nimi and Beamish, 1974) and Tilapia aurea (Winfree and Stickney, 1981).

Feed efficiencies higher than 100% were common in juvenile white sturgeon fed a good diet at or near the optimum feeding rate under our experimental conditions. This may have resulted from the higher body moisture (74-80%) and lower lipid (6-9%) contents of juvenile white sturgeon (Hung et al., 1987b ; Hung and Lutes, 1987 ; Moore et al., 1988 ; Hung 1989 ; Stuart and Hung, 1989, and Hung et al., 1989a) as compared with those of hatchery- raised channel catfish (66-72% moisture and 10-19% lipid) (Garling and Wilson, 1976) and salmonids (65-78% moisture and 11-29% lipid) (Wood et

al., 1957). The best percent protein and energy retained of white sturgeon in our laboratory were slightly lower than the best values obtained from juvenile rainbow trout, common carp (Takeuchi et al., 1978 ; 1979), and channel catfish (Garling and Wilson, 1976 ; Wilson and Poe, 1987). This further indicated that the unusually high percent feed efficiency was an artifact of more body water and less lipid in the juvenile white sturgeon as compared with other species of fish.

The growth and feed efficiency of sturgeon reared at 20°C, however, are comparable to those of channel catfish and better than rainbow trout of similar sizes reared at their respective optimum temperatures. This favorable characteristic suggests a high potential for commercial production of white sturgeon. White sturgeon culture also providing an alternative for locations too warm for salmonids and too cold for catfish.

Results from this and previous studies demonstrate a rapid growth and high feed efficiency of white sturgeon under hatchery conditions. Based on these results and experience in our laboratory, it should be possible to raise white sturgeon from hatch to one kilogram size within a year in a production facility when a good diet, optimum feeding rate, and adequate water temperature are provided. These favorable production characteristics are as good as, if not better than, those of Atlantic salmon and rainbow trout (Storebakken and Austreng, 1987a ; b). These characteristics also suggest a high potential for commercial production of white sturgeon.

Future studies needed for successful sturgeon culture

White and lake sturgeon are still relatively new aquaculture species. Sturgeon cultured to date are F1 offspring from the wild female spawners because reproduction of F1 females in captivity has not been achieved (Doroshov and Binkowski, 1985 ; Conte et al., 1988). Therefore, currently cultured sturgeon stocks in the USA are different from salmonids and catfish which have been subjected to genetic selection for domestication (Gjedrem, 1983, Smitherman et al., 1983 ; Refstie, 1986). Furthermore, being a primitive chondrostean that diverged approximately two million years ago (Sewertzoff, 1923) from the modern teleosts, wild sturgeon possesses very unique biological characteristics. These include longer life-span (may live over 100 year), late sexual maturation (ca. ten years in males and 15 years in females), and long interval between spawning (2-3 year in males and 4-5 years in females). Therefore, the nutrient requirements and culture techniques of these fish are expected to be different from the modern teleosts, such as salmonids, catfish, and carps. These differences make extrapolation of nutrient requirements and culture techniques from the modern teleosts to sturgeon questionable.

Very limited information, however, is available on the optimal environmental conditions (Cech et al., 1984 ; Ruer et al., 1987), feeding rates (Hung and Lutes, 1987, Hung et al., 1990), and culture techniques (Monaco et al., 1981 ; Smith et al., 1981 ; Smith and Dingly, 1984 ; Doroshov, 1985 ; Conte et al., 1988) of white, lake, shortnose, Atlantic, and Siberian sturgeon during their growout phase. Information on diet formulation (Barrucand et al., 1979 ; Buddington and Doroshov, 1984 ; Lindberg and Doroshov, 1986 ; Hung et al., 1987b), nutrient utilizations (Stuart and Hung, 1989 ; Hung et al., 1989a), and nutrient requirements (Hung and Lutes, 1988 ; Moore et al., 1988 ; Kaushik et al., 1989 ; Hung, 1989) of cultured sturgeon is also limited. Therefore, further studies on the nutrient requirements and utilization as well as determinations of the apparent digestibility coefficient, digestible energy, and metabolizable energy values of commonly used fish feed ingredients are needed before a cost-effective sturgeon feed can be developed. Better understanding of efficient feeding practices, management strategies, and other production technique are also needed before efficient growout of sturgeon is possible.

The nutrient requirements and utilization by sturgeon larvae and domesticated broodstocks are expected to be very different from the growout juvenile because the very fast growth rate of the larvae and high fecundity of the broodstock. There is, however, limited information on the larval development (Binkowski and Doroshov,

1985 ; Buddington and Doroshov, 1986a ; Wang et al., 1987 ; Conte et al., 1988) and nutrition (Dabrowski et al., 1985 ; 1987 ; Fauconneau et al., 1986 ; Lutes et al., 1990). Information on general (Smith and Dingly, 1984 ; Binkowski and Doroshov, 1985 ; Conte et al., 1988), gastrointestinal (Buddington, 1985 ; Buddington and Doroshov, 1986b ; 1987) and reproductive biology and physiology (Smith et al., 1980 ; Doroshov, et al., 1983 ; Doroshov and Lutes, 1984 ; Binkowski and Doroshov, 1985 ; Lutes et al., 1987 ; Conte et al., 1988) of sturgeon is also limited. This information is urgently needed to ensure the successful culturing of sturgeon because of the high mortality during their larval stage and failure to complete the reproductive cycle in the F1 captive female broodstock.

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LE METABOLISME DES XENOBIOTIQUES CHEZ L'ESTURGEON : ACTIVITES ENZYMATIQUES ET INDUCTION

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RESUME

L'activité des enzymes de fonctionnalisation et de conjugaison a été mesurée dans le foie, le rein postérieur et les branchies d'esturgeons pesant en moyenne 400 g. Ces mesures ont été obtenues par incubation in vitro de fractions subcellulaires (microsomales ou cytosoliques) à partir de différents substrats. L'effet de la ß-naphtoflavone et du phénobarbital, inducteurs-types des systèmes enzymatiques, a été étudié sur les enzymes hépatiques, rénales et branchiales. Au niveau du foie, les taux de cytochrome P450 et les activités des enzymes qui lui sont associés sont du même ordre de grandeur que ceux mesurés chez la truite. Seule l'UDP glucuronyltransférase a une activité très inférieure à celle des salmonidés. Les niveaux enzymatiques extrahépatiques, bien que généralement 2 à 3 fois plus faibles que dans le foie sont parfaitement détectables. Parmi les inducteurs testés, seule la ß-naphtoflavone a un effet net, quoique tardif, sur les enzymes du métabolisme des xénobiotiques, alors que le phénobarbital est inactif.

Mots clés : *Acipenser baeri*, métabolisme, xénobiotiques, activités enzymatiques, induction enzymatique.

ABSTRACT

Activities of oxidative and conjugative enzymes were measured in the liver, posterior kidney and gills of sturgeon weighing ca. 400 g. The enzyme systems were investigated in vitro, incubating subcellular fractions (microsomes or cytosol) with various substrates. Induction of these enzyme systems by treatment of fish with ß-napthoflavone or phenobarbital was investigated on liver, kidney and gill fractions. Sturgeon hepatic preparations exhibited cytochrome P450 levels and cytochrome P-450 dependent activities nearly comparable to those of the trout. In contrast, UDP-glucuronyl transferase activity was considerably lower than in salmonids. These activities were easily detected in some extrahepatic organs, although they were 2-3 times lower than in liver. Whereas phenobarbital failed to cause an induction of the xenobiotic metabolizing system, the effect of ß-naphtoflavone on these enzymes was clearly demonstrated, eventhough it occurred belatedly.

Key-words : *Acipenser baeri*, metabolism, xenobiotics, enzyme activities, enzyme induction.

INTRODUCTION

Dans la majorité des cas, pour se débarrasser d'une substance étrangère (ou xénobiotique) un organisme vivant devra la métaboliser, c'est-à-dire lui faire subir une ou plusieurs réactions chimiques destinées à accroître sa polarité et de ce fait la rendre plus facilement excrétable. Il apparait donc que la persistance d'un composé dans un organisme dépendra, entre autre, de la capacité de cet organisme à effectuer ces biotransformations. Chez le poisson, pour qui le potentiel des enzymes intervenant dans le métabolisme des xénobiotiques est limité par rapport aux vertébrés supérieurs, on observe généralement une persistance élevée de certains contaminants tels que les PCB ou les organo-mercuriels. Ces phénomènes ont conduit quelques auteurs à avancer, pour certaines espèces sensibles à la pollution telles que les corégones, l'hypothèse selon laquelle la régression des populations naturelles serait corrélée à une faible capacité des enzymes du métabolisme des xénobiotiques (Lindström-Seppä et al., 1981). Outre cet aspect, il faut ajouter que l'on assiste souvent chez des poissons exposés à des polluants divers à une stimulation ou "induction" des systèmes enzymatiques susceptibles de prendre en charge ces composés et donc à un accroissement du potentiel de biotransformation de l'individu contaminé.

Connaissant la déficience de certaines enzymes du métabolisme des xénobiotiques chez des poissons primitifs tels que les lamproies (Lech et al., 1975) et sachant que les seules données que nous ayons sur l'esturgeon, poisson moins évolué que les téléostéens, font état d'activités à peine mesurables (Buhler et al., 1968), il était légitime de nous interroger sur les systèmes enzymatiques d'Acipenser baeri. Parmi ces systèmes, certains dits de fonctionnalisation sont liés au cytochrome P450 et sont chargés d'hydroxyler la molécule exogène. Les autres, appelés de conjugaison, combinent une molécule ou un radical endogène avec la substance exogène (ou avec un de ses métabolites) pour former une molécule moins liposoluble.

Le but de notre étude a été de mesurer l'activité des enzymes de fonctionnalisation et de conjugaison dans le foie, le rein postérieur et les branchies d'esturgeons (*A.baeri*) pesant en moyenne 400 g. Ces mesures ont été obtenues par incubation in vitro de fractions subcellulaires (microsomales ou cytosoliques) avec les substrats suivants : benzo(a)pyrène, 7-éthoxycoumarine, styrène oxyde, p-nitrophénol, 1-chloro-2, 4-dinitrobenzène. L'effet de la ß-naphtoflavone et du phénobarbital, inducteurs-type des sytèmes enzymatiques a été étudié sur les enzymes hépatiques, rénales et branchiales.

MATERIELS ET METHODES

Produits chimiques

Le 8-[^{14}C] styrène oxyde et le 4-nitro-[U-^{14}C]-phénol utilisés proviennent d'Amersham (France) et le [^{3}H]-benzo-a-pyrène du CEA Saclay (France).

Le 4-nitrophénol et le benzo(a)pyrène proviennent de Fluka AG (Suisse), l'acide UDP-glucuronique, le glutathion réduit, la 7-éthoxycoumarine et le cytochrome c de Boehringer (France) et la 5,6-benzoflavone ou ß-naphtoflavone de Sigma (France).

Animaux

Les esturgeons, âgés de 2 ans et demi et pesant 400 g. environ ont été fournis par le CEMAGREF et proviennent de la pisciculture de Donzacq (Landes, France). Les truites qui servent de référence dans cette étude ont un poids moyen de 300 g et proviennent de la même pisciculture. Pendant la période expérimentale, les poissons sont maintenus à jeûn et stockés au Laboratoire en eau recyclée à 12 ± 2°C en attendant d'être sacrifiés. Les tests d'induction ont été pratiqués sur des esturgeons recevant par injection intrapéritonéale soit du phénobarbital dissout dans du chlorure de sodium 0,9 % à la dose de 20 mg/kg trois jours consécutifs avant l'abattage, soit une dose unique de 100 mg/kg de ß-naphtoflavone dissoute dans de l'huile de maïs. Dans ce cas, les esturgeons sont sacrifiés 3, 15 et 30 jours après l'injection. Un groupe d'esturgeons ne recevant que de l'huile de maïs sert de lot témoin.

Préparation des microsomes

Les esturgeons sont assommés, puis le foie, le rein postérieur et les branchies sont rapidement prélevés, pesés et plongés dans du tampon phosphate de potassium 0,1 M pH 7,4 contenant 1,15% de KCl.

Les différentes étapes du fractionnement cellulaire sont ensuite réalisées à + 4°C : les trois types de tissus sont broyés et homogénéisés avec un homogénéisateur de Potter à piston en téflon dans 4 volumes de tampon phosphate de potassium 0,1 M pH 7,4 contenant 1,15 % de KCl et un inhibiteur de la trypsine pour bloquer toute activité protéasique. Le surnageant post-mitochondrial obtenu en centrifugeant l'homogénat à 11000 g pendant 20 min. est centrifugé à 105000 g pendant 1 h. (ultra-centrifugeuse Kontron T 1045) ; le cytosol et les microsomes ainsi séparés pour les différents organes sont conservés à -80°C.

La même technique est utilisée pour la préparation des microsomes de foie de truite.

Dosages biochimiques

Le culot microsomal est homogénéisé au Potter dans du tampon phosphate pH 7,4. Les concentrations en cytochrome P450 sont déterminées selon la procédure d'Omura et al. (1964) modifiée par Johannesen et al. (1978). Toutes les mesures spectrales sont réalisées sur un spectrophotomètre Kontron Uvikon 810.

L'activité NADPH-cytochrome C réductase est suivie en spectrophotométrie selon le protocole décrit par Mazel (1972). La mesure de la benzo(a)pyrène-hydroxylase (AHH) se fait en radioactivité grâce à la technique de De Pierre et al. (1975) modifiée par Van Cantfort et al. (1977). La 7-éthoxycouramine déethylase (ECOD) est mesurée en suivant l'apparition de 7-hydroxycoumarine fluorescente (Aitio, 1978).

L'activité époxyde hydrolase (EH) est mesurée avec le 8 ^{14}C-styrène oxyde comme substrat, selon la procédure décrite par Oesch et al. (1971).

La détermination de l'UDP glucuronyl transférase (UDPGT) se fait en radioactivité avec du 4-nitro-[U-^{14}C]-phénol comme substrat selon la méthode de Tukey et al. (1978). L'activité de la glutathion transférase cytosolique (GSHST) est suivie en spectrophotométrie comme décrit par Habig et al. (1974). Les teneurs en protéines sont mesurées selon les techniques de Gornall et al. (1949) pour les microsomes et de Lowry et al. (1951) pour les cytosols avec l'albumine sérique bovine comme référence. Toutes les mesures de radioactivité sont effectuées sur un compteur à scintillation liquide Packard Minaxi 4430.

RESULTATS

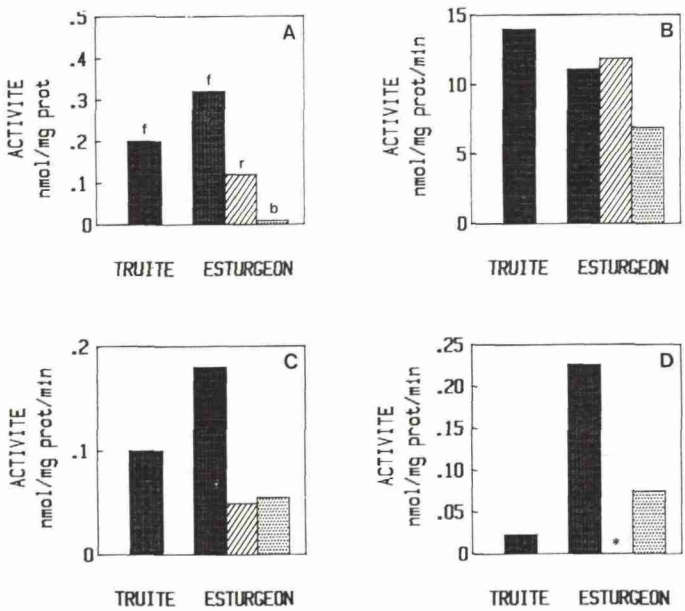


Figure 1 - Activités des enzymes de fonctionnalisation dans le foie (f),le rein postérieur (r) et les branchies (b) d'esturgeon. Comparaison avec les activités enzymatiques correspondantes chez la truite.
A : *taux de cytochrome P450.* B : *activité de la NADPHcytochrome c reductase mesurée à 25°C,* pH 7,6. C : activité de l'AHH mesurée à 30°C, pH 7,5. D : activité de l'ECOD mesurée à 30°C, pH 7,4.
Les résultats proviennent de mesures faites sur des lots de 10 animaux.* : Absence de mesure en raison du manque de microsomes.

Les préparations microsomales obtenues à partir du foie d'esturgeon contiennent des quantités appréciables de cytochrome P450 et des enzymes qui lui sont associés (Fig. 1).

Les activités de ces enzymes sont pour la plupart comparables, ou parfois même nettement supérieures (ECOD, Fig. 1 D) à celles mesurées dans des conditions identiques chez la truite. Comme celà a déjà été montré pour d'autres espèces de poissons, les activités enzymatiques extrahépatiques sont plus faibles que celles mesurées dans le foie. Cette remarque s'applique également aux enzymes qui ne sont pas cytochrome P450-dépendantes, telle que la GSHST ou l'EH (Fig. 2). L'UDPGT est le seul cas dans notre étude où l'activité enzymatique s'avère nettement plus faible chez l'esturgeon que chez la truite (Fig. 2 B).

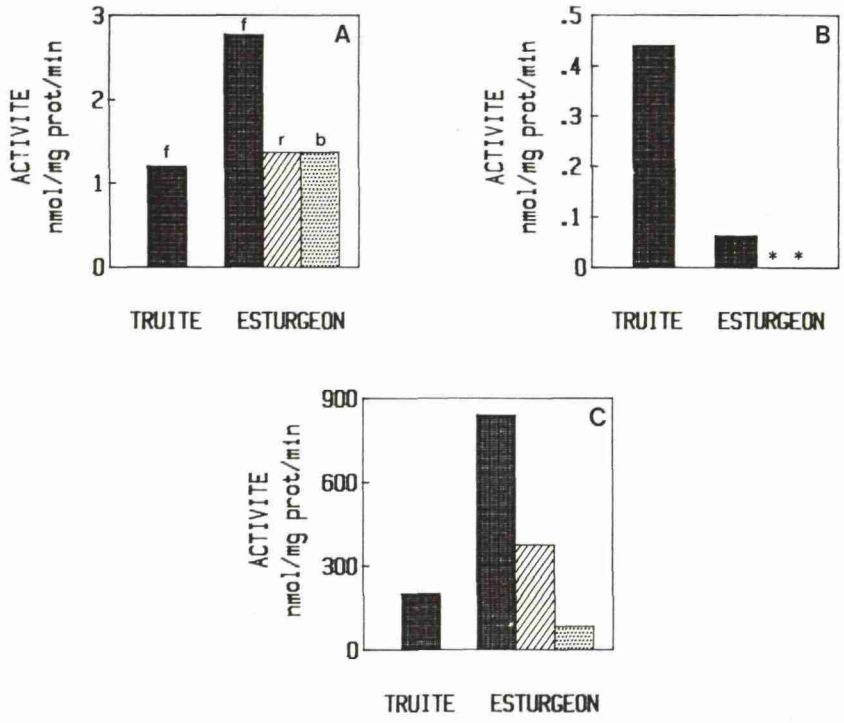


Figure 2 - Activités de l'époxyde hydrolase (EH) et des enzymes de conjugaison dans le foie (f), le rein postérieur (r) et les branchies (b) d'esturgeon. Comparaison avec les activités enzymatiques correspondantes chez la truite.
A : EH, mesurée à 30°C et pH 9. B : UDPGT, mesurée à 25°C et pH 7,4. C : GSHST, mesurée à 25°C et pH 7,4.
Les résultats proviennent de mesures faites sur des lots de 10 animaux.
* : Absence de mesure en raison du manque de microsomes.

Le traitement des poissons par une injection unique de ßnaphtoflavone n'entraine pas d'effet sur le taux de cytochrome P450 du foie au cours des 15 premiers jours (Fig. 3). En revanche, après un mois, la teneur en cytochrome P450 dans le foie des animaux traités est 2,3 fois supérieure à celle mesurée chez les témoins. Il n'a pas été observé de modification spectrale du cytochrome P450 lors du traitement à la ß-naphtoflavone ni de variation du taux de protéines microsomales.

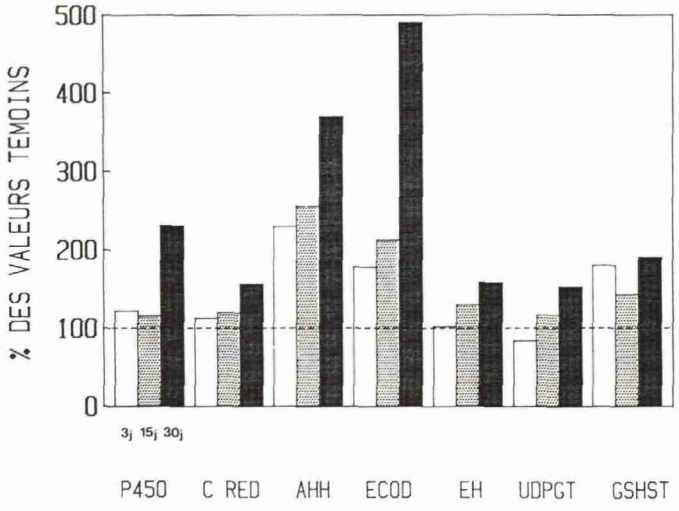


Figure 3 - Action de la ß-naphtoflavone sur les activités enzymatiques du foie d'esturgeon. Les analyses ont été effectuées sur des échantillons regroupés provenant de 4 animaux, 3, 15 et 30 jours après l'injection de ß-naphtoflavone.

Pour toutes les enzymes testées (Fig. 3), les niveaux d'activité les plus élevés sont observés un mois après l'injection de ß-naphtoflavone, et pour la plupart d'entre elles, les activités en fin d'expérimentation sont 1,5 fois plus élevées que le niveau de base. Toutefois, une augmentation nette et constante apparait pour l'AHH et l'ECOD ; ces enzymes voient leur activité multipliée par 1,8 et 7,8 respectivement au bout de 3 jours et par 4,9 et 12,8 respectivement en un mois.

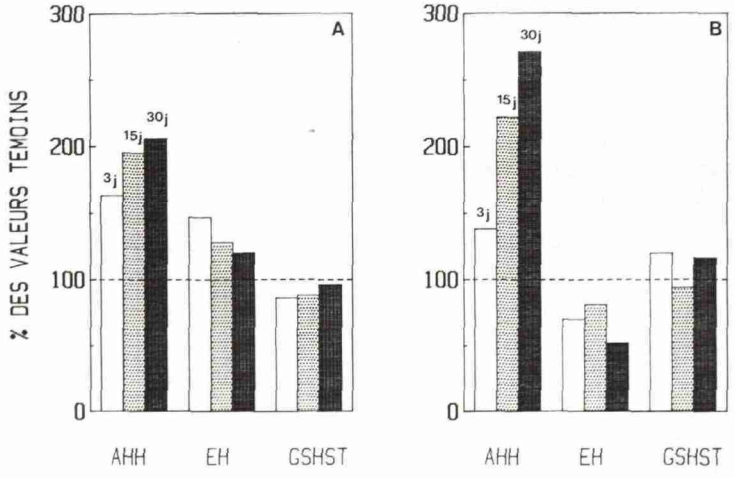


Figure 4 - Action de la ß-naphtoflavone sur les activités enzymatiques de rein postérieur (A) et de branchies (B) d'esturgeon.
Les analyses ont porté sur des échantillons regroupés provenant de 4 animaux abattus 3, 15 et 30 jours après l'injection de ß-naphtoflavone.

Le traitement à la ß-naphtoflavone ne semble pas avoir d'effet inducteur sur l'EH et la GSHST du rein ou des branchies (Fig. 4) mais augmente l'activité de l'AHH d'un facteur 2 dans le rein et d'un facteur 2,7 dans les branchies.

Le phénobarbital en revanche, ne provoque aucune stimulation sur les systèmes métaboliques analysés dans cette étude.

DISCUSSION

Contrairement aux premières études menées par Buhler and Rasmusson (1968) chez *Acipenser transmontanus*, ce travail démontre qu'*Acipenser baeri* possède dans la majorité des cas un niveau d'activité aussi important que celui observé chez *Salmo gairdneri*. La métabolisation de certains substrats tels que le benzo(a)pyrène, l'éthoxycoumarine ou le styrène oxyde est même plus intense chez l'esturgeon que chez la truite. Il semble donc que ce poisson possède l'équipement enzymatique nécessaire à la métabolisation des substances exogènes susceptibles de contaminer sa nourriture ou son environnement.

Parmi les enzymes que nous avons testées, l'UDPGT est la seule pour laquelle l'esturgeon présente un taux nettement inférieur à celui mesuré chez la truite. Cette caractéristique pourrait néanmoins avoir des conséquences importantes sur la sensibilité de l'esturgeon aux xénobiotiques détoxifiés par cette voie.

Il existe, en effet, une relation directe entre la toxicité de certaines substances et la capacité de l'organisme cible à les conjuguer à l'acide glucuronique. Lech et al. (1980) ont démontré qu'en inhibant l'UDPGT chez la truite, on augmentait fortement la toxicité du 3-trifluorométhyl-4-nitrophénol à l'égard de cette espèce.

Dans le cas de l'esturgeon, nos résultats ne suffisent pas à conclure à une déficience en UDPGT, il existe en effet de nombreuses formes de cette transférase. Gregus et al. (1983) ont montré chez plusieurs espèces que les mesures d'UDPGT pouvaient varier considérablement en fonction du substrat choisi ; il est donc possible que d'autres valeurs que les notres soient obtenues en utilisant un autre xénobiotique que le p-nitrophénol. Par ailleurs, une étude récente sur le devenir du molinate chez *Acipenser transmontanus* a montré que le métabolite majeur de cet herbicide était le 4-hydroxymolinate glucuronide, ce qui prouve que l'UDPGT s'exprime parfaitement *in vivo* chez cette espèce. La mesure de la glucuronidation *in vitro* et *in vivo*, à partir de différents contaminants ou médicaments permettrait donc de compléter nos données et d'appréhender de façon plus précise les capacités de cette voie métabolique chez l'esturgeon.

L'induction des enzymes microsomales et cytosoliques est abondamment décrite chez le mammifère. C'est le moyen mis en oeuvre par l'animal pour augmenter son potentiel de détoxification vis-à-vis des substances exogènes lors d'une contamination massive ou fréquente par ces composés.

Chez le poisson, la plupart des études portent sur les monooxygénases hépatiques et démontrent que plusieurs types d'agents chimiques sont capables d'augmenter le potentiel métabolique des animaux traités en agissant sur le cytochrome P450 ; (Lech et al. 1982). Beaucoup de ces agents sont des hydrocarbures aromatiques polycycliques ou encore des polychlorobiphényls. Contrairement aux mammifères, les poissons ne présentent pas ou très rarement une induction significative au phénobarbital ; cette caractéristique s'applique également à l'esturgeon puisque nous n'avons obtenu aucune augmentation d'activité enzymatique avec cet inducteur.

En revanche, avec la ß-naphtoflavone, inducteur fréquemment utilisé chez la truite, nous avons pu observer certaines modifications d'activité. Les augmentations les plus nettes ont été relevées dans le foie avec l'AHH et l'ECOD alors que dans les reins et les branchies, pour lesquels il n'y a pas eu de mesure de l'ECOD, c'est l'AHH qui a été davantage stimulée ; notons également une nette progression du taux de cytochrome P450 un mois après l'induction. Si l'on compare ces résultats avec ceux décrits chez la truite par Elcombe et al. (1979) et par Andersson et al. (1985) ou chez le poisson-chat

par Tate (1988), il apparait chez toutes ces espèces que l'AHH et l'ECOD sont les enzymes répondant le mieux à une induction par la ß-naphtoflavone.

Deux points caractérisent cependant l'esturgeon ; il semble tout d'abord que l'amplitude de cette induction soit plus faible que chez les autres espèces. Ainsi, chez la truite l'activité de l'AHH est multipliée par 40 lors d'une injection intrapéritonéale de 100 mg/kg de ß-naphtoflavone (Elcombe et al., 1979), alors qu'elle n'augmente que d'un facteur 4 chez l'esturgeon. Il est peu probable que la faible réponse de l'esturgeon soit due à nos conditions expérimentales (animaux à jeûn depuis 5 jours au début de l'expérience et donc privés de nourriture depuis 5 semaines pour les individus abattus à 30 jours) puisque Andersson et al. (1985) ont pu montrer qu'un jeûne de 6 semaines n'affectait aucunement l'activité enzymatique des foies de truites, induites à la ß-naphtoflavone ou pas.

Le second point concerne la lenteur avec laquelle cette induction se manifeste ; chez le poisson l'induction maximale est généralement observée entre 3 et 7 jours après l'injection de ßnaphtoflavone. Ce phénomène décroit ensuite plus ou moins rapidement suivant les enzymes (Andersson et al. 1985). Notre étude chez l'esturgeon a montré que pour toutes les enzymes hépatiques testées, le maximum d'activité avait été enregistré en fin d'expérimentation c'est-à-dire un mois après l'injection de ß-naphtoflavone. Il faut noter que chez le poisson, la température de l'eau joue un rôle important sur le délai d'apparition du maximum d'induction. Ainsi, Andersson et al. (1985) ont observé chez des truites traitées à la ß-naphtoflavone que la réponse maximale à cet inducteur s'obtenait dès 24 h chez des animaux vivant à 17°C mais pas avant 3 jours à 5°C. De même, *Archosargus probatocephalus* répond plus lentement à l'induction au méthylcholanthrène à 14°C qu'à 26°C (James et al., 1980).

Notre étude, menée à la température moyenne de 12°C a révélé une certaine inertie de l'esturgeon vis à vis d'inducteurs du type ß-naphtoflavone. Il est cependant difficile de savoir si cette caractéristique peut avoir des conséquences sur la survie de cette espèce dans un environnement fortement pollué. Il est bien connu que l'induction des systèmes enzymatiques peut modifier la toxicité de nombreux produits chimiques. En fonction des étapes de biotransformation celà peut conduire à une toxicité accrue (cas d'une production d'un intermédiaire réactif au cours du métabolisme) ou à une élimination plus rapide de l'agent exogène (voie classique de la détoxification). Ainsi, lors du métabolisme du benzo(a)pyrène, on observe chez la majorité des espèces, la formation d'un métabolite, le benzo(a)pyrène 7,8dihydrodiol-9,10-époxyde (Melius, 1984) qui réagit fortement avec l'ADN et engendre un processus de mutagénèse, puis de cancérogénèse. Chez le saumon coho, l'induction des enzymes associées au cytochrome P450 augmente plus de 50 fois la liaison à l'ADN des métabolites du benzo(a)pyrène (Varanasi et al., 1980). En revanche, l'induction a un effet protecteur vis-à-vis de l'aflatoxine B1 chez la truite en favorisant la production et l'élimination de métabolites non-cangérigènes (Hendricks & al., 1977).

Il ressort de cette étude que l'esturgeon possède un équipement enzymatique équivalent à celui de poissons évolués tels que la truite. Deux points le caractérisent cependant : une faible capacité de glucuronidation et un très long temps de réponse aux inducteurs de type ß-naphtoflavone.

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EVOLUTION OF SEX STEROID LEVELS IN MALES AND FIRST TIME MATURING FEMALES OF THE SIBERIAN STURGEON *(Acipenser baeri)* REARED IN A FRENCH FISH FARM

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RESUME

Les niveaux circulants d'androgènes (A), d' oestrone (E1) et d'oestradiol (E2) ont été mesurés tous les trois mois pendant deux ans et demi sur des esturgeons sibériens *Acipenser baeri* élevés dans une pisciculture expérimentale française. Les profils obtenus chez les femelles ont montré que l'évolution de l'ovogénèse pouvait être corrélée aux concentrations d'androgènes dans le plasma mais non à celles des oestrogènes. Un changement du régime alimentaire effectué en début d'expérimentation, suggère une influence de l'alimentation sur les niveaux circulants d'oestrogènes. La vitellogénèse qui s'est déroulée normalement chez les femelles, s'est accompagnée d'une augmentation des androgènes dans le plasma, mais d'aucune augmentation des taux d'oestrone ni d'oestradiol. La spectrométrie de masse a confirmé l'absence de doses d'oestradiol supérieures à 1ng/ml chez les femelles en vitellogénèse. Les androgènes ont disparu et ont atteint un niveau très bas au moment de l'ovulation. Les données endocriniennes, histologiques et ultrastructurales indiquent que l'évolution des ovocytes du Stade II au Stade V dure 3 à 4 ans dans les conditions thermiques particulières de la pisciculture expérimentale (17°C).

Mots clés : stéroïdes sexuels, *Acipenser baeri*, phytostéroïdes, gamétogénèse, aliment.

ABSTRACT

Androgens (A), estrone (E_1), and estradiol (E_2) were measured every three months for two years and an half in a population of Siberian sturgeons *Acipenser baeri* bred in an experimental French fish farm. The profiles obtained in females showed that the evolution of the oogenesis could be correlated to androgens levels but not to those of estrogens. A change in the diet of the fish, at the beginning of the experiment, suggested an influence on estrogens profiles. Vitellogenesis was normal in females and correlated with a rise in plasma androgens, but not in estrone nor in estradiol levels. Mass spectrometry analysis confirmed the absence of plasma levels superior to 1ng/ml of estradiol in vitellogenic females. Androgens decreased to a very low level at the time of ovulation. Radio-immunoassay (RIA), histological and ultrastructural data indicated that oocyte transformation from Stage II to Stage V took 3 to 4 years in the specific thermic conditions of the experimental farm (17 $\pm$ 1°C).

Key-words : sex steroids, *Acipenser baeri*, phyto steroids, gametogenesis, food.

INTRODUCTION

The exploitation of wild stocks and farming of sturgeons are of economic interest throughout the world. A French research project has been set up to study the reintroduction of the wild species *Acipenser sturio*, and to evaluate the zootechnical potentialities of the Siberian sturgeon *Acipenser baeri* for culture in fish farms. Siberian sturgeon were imported for this purpose from the USSR into France in 1974 and 1982. Since 1981, this species has been succesfully reproduced each year in France (Williot et al., 1982a; Williot et al., 1982b) even if percentage of ovulating female is relatively low. Fish are now available for scientific investigation.

The breeding of sturgeon requires greater knowledge of their reproductive cycle. Such studies can be based on morphological or ultrastructural parameters (Pelissero et al., 1985 ; Le Menn et Pelissero, 1990) or on endocrine data. Many studies have dealt with the influence of gonadotrophic hormone (GTH) on gonads development

(Barannikova et al., 1981 ; Barannikova and Fadeeva, 1982 ; Barannikova et al., 1984 ; Barannikova et al., 1985) but hardly any data are available on the sex steroid cycle. The only available studies concerned with sex steroids in sturgeon are those of Sangalang et al., (1971) on two testosterone measurements and by Lutes et al., (1987) on the evolution of a progestagen during maturation. This lack of information is probably due to the length of the sturgeon reproductive cycle. At Donzacq fish farm, with water at a constant 17 ± 1°C, gametogenesis was shortened and sexual maturity was obtained in 5-years-old males and 7-years-old females i.e. half the time necessary in the USSR. In such thermic conditions the study of paramaters such as sex steroids, vitellogenin (Pelissero and Le Menn, 1988) or other endocrine data becomes possible.

This study presents results obtained by sex steroid measurements with sampling performed every three month for two years and an half on male and female Siberian sturgeons bred in the INRA experimental fish farm, CEMAGREF hatchery of Donzacq. Sex steroid measurements and histological and ultrastructural data are tentatively related to oocyte developmental stages.

The change in the brand of fish diet used during the experiment seems to indicate an influence of the composition of this diet on the steroid levels in fish plasma. Two different effects are described involving animal and vegetal protein sources in the diet composition.

MATERIALS AND METHODS

Animals

The sturgeons used in this study were imported from the USSR in 1982 to the INRA experimental fish farm, CEMAGREF hatchery of Donzacq (Landes, France). Fish were maintained in tanks (40 metres long and 6 metres wide) at a density of about 1200 kg of wet weight per tank. The average fish weight was 4.5 kg at the beginning of the experiment and 6.7 kg at the end. Six hundred and fifty 4-years-old fish were sexed by biopsy of the gonad associated with histological control. 120 females and 30 males were used among them for the experiment.

Blood from all 150 sturgeons was sampled every three monthes to ensure that each fish received the same treatment. On each occasion a statistically representative sample of 7 ramdomly chosen males and females were examined by gonad biopsy in order to investigate correlations between blood data and oocyte stage.

For each sex steroid measurement (presented in Fig. 1 and Fig. 2) 15 plasma samples were randomly chosen from the 120 samples of females and 15 others from the 30 samples of males without particular correlation with the spermatogenesis or oogenesis stage determination.

For all the experiments the fish were fed with commercial trout diet but the brand used was changed at the beginning of the experiment (January 1988).

Gonadal stage

The gonadal stage were determined by gonad biopsy on male and female sturgeons associated with histological control. For all the experiment, males had already spermatozoa in their testis and the gonadal stage was simply defined as the percentage of spermatozoa in reference to other germinal cells in testis. For females various electron microscopy investigations were also performed and oogenesis stage were defined as presented in Le Menn and Pelissero (1991).

Blood sampling

Blood samples were collected from the caudal vein using an heparinized syringe. The samples were kept at 4°C before centrifugation at 10,000g for 5 min to separate the plasma which was stored at -20°C until needed for radio-immunoassay (RIA).

Plasma treatment

Sex steroids were treated according to Fostier et al., 1978. They were extracted from the 500µl plasma samples by two successive extractions in 2ml ethyl acetate - cyclohexane (1-1) mixture (91% of recovery).

The extrated fraction was then evaporated to dryness, dissolved in 200µl ethanol and stored at -20°C until chromatographic separation. Sex steroid separation was performed on a 8cm x 0.5cm Sephadex LH 20 (Pharmacia) column in a dichloromethane - methanol (95-5) mixture and the separated fractions evaporated to dryness were then dissolved in 200µl ethanol and stored at -20°C until the RIA manipulation.

The collecting efficiency of the extraction and the separation is evaluated by addition and incubation (2 hours at room temperature) of 1000 cpm of labelled sex steroid to several standard plasma samples before all manipulations (recovery efficiency: estrone 70% ; estradiol 60% ; testosterone 80%). Just before the assay the plasma samples were dissolved in 500µl of gelatinized (0.1%), phosphate buffer (0.01M) saline (0.15M), pH 7.25 (RIA buffer).

Plasma steroid levels were compaired to a sex steroid standard range. Standards were obtained from Steraloid Ltd (England).

The immunological reaction was performed against antibody to estrone and to estradiol from Steranti comp. (England) (anti-estrone ref. E 001 ; anti-estradiol ref. E 002) and against an anti-testosterone immunserum provided by Dr Fostier (INRA Laboratory of Fish Physiology, Rennes). The cross reactions of Steranti estrone antibody is of 1.4 % with estradiol and the Steranti estradiol antibody cross react at 0.85 % with estrone. The testosterone immunserum from Dr Fostier (Rennes) had a crossreactivity of 100% with testosterone, 31% with 11-ketotestosterone, 14% with 4-androstenedione and 40,5% with 5-dihydrotestosterone as such is considered as an anti-androgen immunserum. Percentage of linkage was calculated by competition with labelled sex steroids from Amersham France ((2,4,6,7) ^{3}H estrone ; (2,4,6,7) ^{3}H estradiol ; (1,2,6,7) ^{3}H testosterone)).

Results were expressed after a log - logit transformation : the intra-assay variations (n=10) is 11.5% for estradiol, 9.9% for estrone, 8.2% for androgens ; the inter-assay variation (n=6) is 13.8% for estradiol, 13.2% for estrone, 12.1 for androgens. The lower sensitivity is more or less 1 ng/ml for all the steroids measured since a dilution to 1/20 is practiced in each case. Below, the assay appears distored.

RIA in the diets

The sex steroids in the commercial diet for trout used for our fish were measured as described above on sonicated material in solution in RIA buffer. The extraction procedure is presented in Pelissero et al., (1989). Their composition is unknown for commercial reason but the mean estradiol levels are 350 ng/100g for Diet 1 and 50 ng/100g for Diet 2 measured on 4 different batches.

Mass spectrometry analysis

The gas chromatography - mass spectrometry analysis (GC -MS analysis) was performed in the CESAMO at the University of Bordeaux I on a VG Micromass 16F. Molecules were separated before mass analysis on a SE 52 capillary column (50m ; 0.31mm) from Hewlett Packard adapted on a split injector. The carrier gas was helium and a thermal program applied to the different samples raised the temperature from 250°C to 310°C within 30 min. This technique employed with fragmentography technique has a 1 ng detection limit.

The sample analysed in Fig. 3 was collected from a Stage IV female plasma extracted by RIA technique, evaporated to dryness and conjugated to tri-methyl- silyl ether (TMS ether) before analysis. The injected volume correspond to one ml of extracted plasma.

RESULTS

The evolution of oogenesis stage from Stage II to Stage V takes about 2 years and an half since at the beginning of the experiment in June 1987 females were in Stage II, reaching Stage III in December 1987, Stage IV in June 1988 and Stage V in December 1989. The large variability in oogenesis stages could be connected to the duration of oogenesis in this species and the absence of genetic selection in this population.

Fig. 1 gives the estrone, estradiol and testosterone levels in male plasma. Throughout the experiment, males were in spermatogenesis and had large quantities of spermatozoa in their gonads. In males, the gonadal stages were determined by the evaluation of the percentage of spermatozoa in volume in reference to the other germinal cells calculated on 10 different testicular lobes. The estrogen levels were just as high in the first three sampling, before the change of the diet and the estradiol levels in males were not significantly different from those of females (30 ± 14.5 ng/ml in males ; 80 ± 20.8 ng/ml in females in June 1987). These estrogens levels then decreased rapidely to reach values close to 1 ng/ml (detection limit of our assay). Androgen levels were always very high, with more or less individual variability. The higher value was obtained in the fourth sampling just after the diet change with 880 ng/ml. The androgen levels in males are significantly lower in March 1989 than in March 1988.

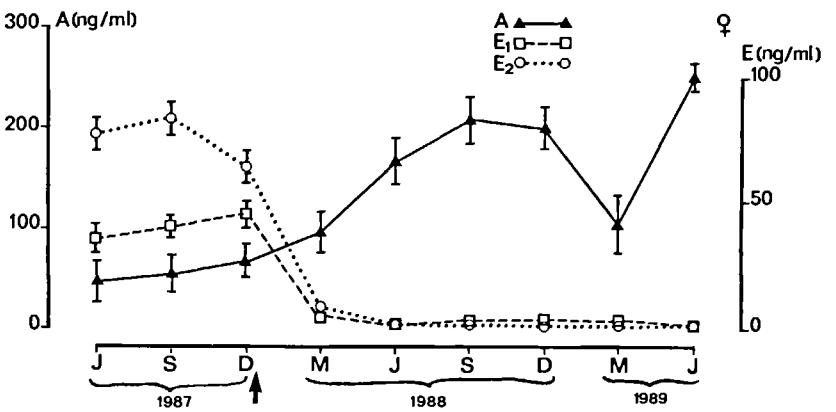


Figure 1 - Evolution of androgens (A), estrone (E1) and Estradiol (E2) plasma levels in 5, 6, 7 years-old males of Siberian sturgeon bred in fish farm. The arrow indicates the moment of the change from diet 1 to diet 2. Bars are SE calculated on 15 different data for each point.

Fig. 2 gives the estrone, estradiol and androgen profiles for females. A great individual variation was noticed in hormone levels especially for androgens. After the first three samples, collected before the diet change, and containing a large amounts of estradiol (80 ng ± 20.8 ng/ml) and estrone (40 ± 8.6 to 45 ± 10.2 ng/ml), the estrogen levels declined to values closed to the detection limit (average 1ng) and their evolution was no longer correlated with oocyte development. Estradiol levels decreased until the end of the experiment while estrone levels increased slightly (from 1 ng to 3.5 ng/ml) during summer 1988 and decreased later. Androgen levels were relatively low at the beginning of the experiment (40 ± 15 ng/ml to 68 ± 12 ng/ml before the diet change) and rose progressively after the March 1988 sampling to reach value close to 90 ng/ml. This increase was interrupted in March 1989 ; perhaps because of the presence of Stage V females reaching ovulation. In this female plasma, the androgen levels were very low (about 1ng/ml) and the mean value obtained for 15 different females decreased. In June 1989, the androgen level increased because the more advanced females showed atretic features among the next oocyte batches at Stage III or IV i.e. in vitellogenesis.

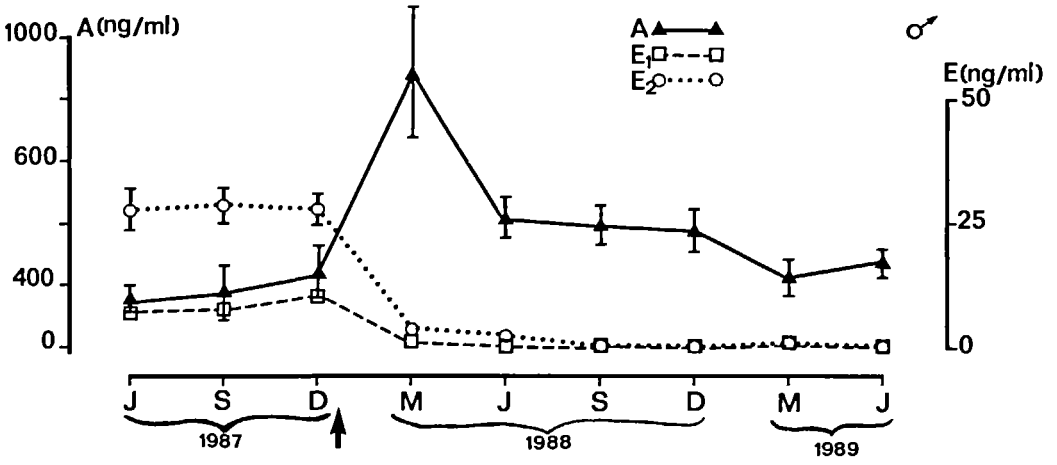


Figure 2 - Evolution of androgens (A), estrone (E1) and Estradiol (E2) plasma levels in 5, 6 and 7 years-old females of Siberian sturgeon bred in fish farm. The arrow indicates the moment of change from diet 1 to diet 2. Bars are SE calculated on 15 different data for each point.

Results of GC - MS analysis are presented in Figure 3, 4 and 5. The gas chromatography profile obtained from Stage IV female plasma (Fig. 3) was caracterised by a first peak at 91 scan number corresponding to phtalate dioctyl contaminant probably contained in extraction solvent. Two smaller peaks then appeared. The second at 143 scan number was testosterone as could be verify in (Fig. 4) on the fragmentogram obtained for typical molecular ion of TMS-testosterone mass spectrum (mass m/z: 360) (Fig. 5). The fragmentographic analysis does not allow the identification of TMS estradiol 17ß. The 180 scan number compound was not identified. After these smaller peaks one very large peak saturating the chromatography column appeared at 412 scan number. This was identified as cholesterol thanks to its mass spectra compared to the TMS-cholesterol reference mass spectra (molecular ion at mass m/z: 458) (unpresented results). This large peak of cholesterol overlaped one of cholestadien and another of tocopherol (spectra not shown).

DISCUSSION

The results presented here on sex steroid profiles have not been previously reported in bred sturgeons. The main reason is that sexual maturity in sturgeons usually occurs very late (about 14 years for males and 16 years for females of *A. baeri* in nature) (Akimova et al., 1979). This long period of maturation causes problems for breeding and scientific investigations. In this study we take the advantages of the specific thermic conditions of the experimental fish farm (17 ± 1°C) which provokes a more rapid sexual development of the fish.

In males

Androgen levels were just as high but could be in agreement with those obtained in other fish such as Atlantic and Sokeye salmons (Schmidt and Idler, 1962), or rainbow trout (Scott and Sumpter, 1983) and also with those reported on the Atlantic sturgeon (Sangalang et al., 1971). They have been correlated with the percentage of spermatozoa in comparison to other testicular germ cells (Pelissero, 1988). For all the experiments and even at the beginning, spermatozoa were generally present in large amount in gonads 75% to 100%. This is not surprising since sexual maturation in males

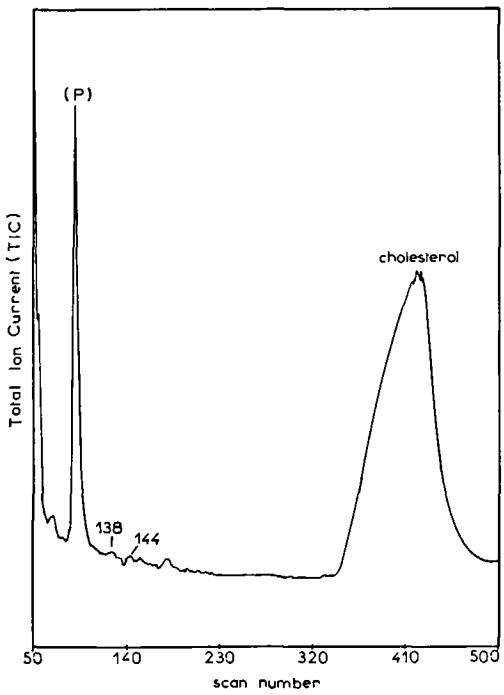


Figure 3 - Gas chromatography analysis of stage IV female total plasma derivative extract

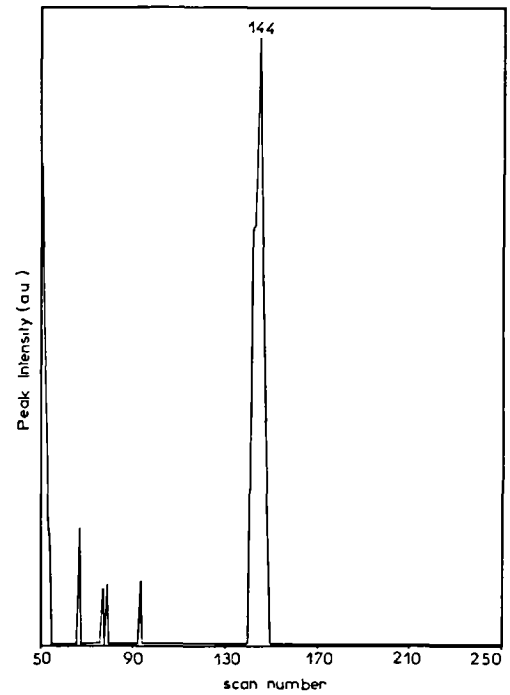


Figure 4 - TMS-testosterone fragmentogram from female plasma chromatogram corresponding to 143 scan number

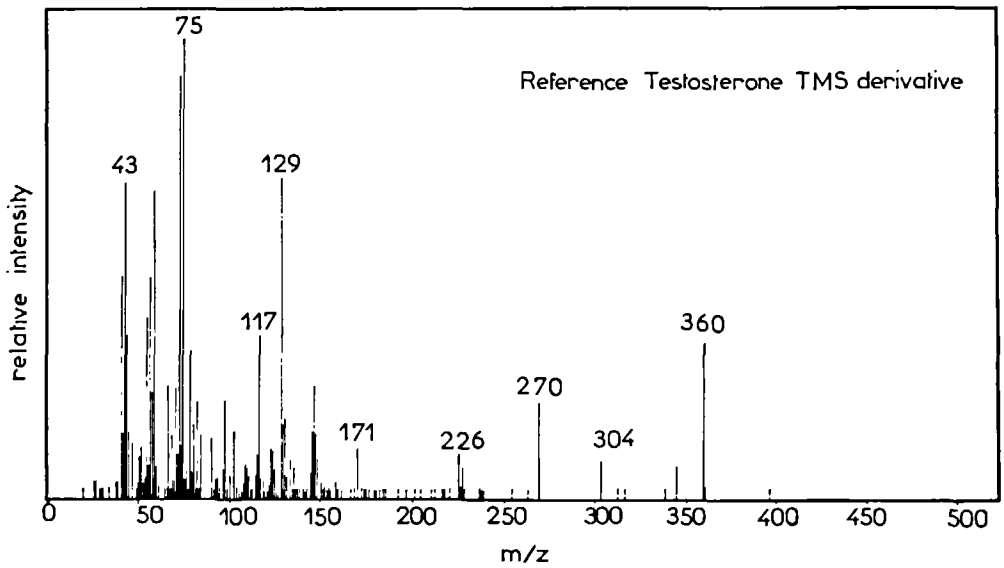


Figure 5 - standard TMS-testosterone mass spectrum compared in this study to the mass spectrum of 143 scan number peak

is reached two years earlier than in females. In March 1987, males had already reached a high developed stage. The first notable rise of androgens in March 1988 could be related to spermiation since the reproductive season of Siberian sturgeon in France is in spring. It could also be due to the diet change but this transitory effect remains unclear. Spermatozoa were then progressively eliminated and the spermatozoa stock is progressively reconstituted. The percentage of spermatozoa in reference to other germinal cells in testis rose again in June 1988. A decrease in androgens was observed in March 1989 probably because the reproductive season already occured for males at the time of the sampling. This advance in time could be correlated to the exceptionally warm and dry winter in France which could have an influence on water temperature in Donzacq (± 1°C). It could also be interpreted as a shift of the breeding season caused by the absence of marked thermal variations. The estrogen levels noted in males before the diet change were very high comparing to that observed for other species (about 30 ng/ml). After the diet change they fall down definitively and were in accordance with data previously reported by other authors (Schreck and Hopwood, 1974; Billard et al., 1978; Idler et al., 1981). This seems to indicate an influence of the diet brand on estrogen levels in males most of all if one considers that classically males do not exhibit high plasma estrogen levels. The estrogen amount measured in the two diets shows that the first one contains more or less 7 fold more estradiol than the second one. The influence on the plasma estrogen levels could perhaps be related to those data.

In females

Androgen levels were high relatively to that observed in other fish species (Campbell et al., 1976; Scott et al., 1980 a, b; Wingfield and Grimm, 1977). However increased androgen levels throughout the oogenesis was observed, since testosterone is one of the precursor of estrogens (Campbell et al., 1976). In March 1989 the androgens decreased. Several females reached maturation and had no more than 1ng/ml plasma androgens (assay detection limit). Since other females were still in stage IV the mean value was high, although the individual variation was large. In June 1989 the androgen levels increased again which could be explained by the fact that the more advanced females presented atretic features among Stage III and IV oocytes and recommenced vitellogenesis for the next oocyte batches. Estrogen levels have never been measured in cultured sturgeons. Although RIA data on estradiol and estrone levels were unexpected. The strong decrease in those levels at the time of diet change confirms the influence of diet brand on hormone plasma levels. The definitive decrease of estrogen levels after diet change was also unexpected since estrogen plasma levels classically rose during vitellogenesis. Androgen levels are usually higher but of the same range than those of estrogen since they are metabolically linked. For some authors the range is high (about ten ng/ml) (Wingfield and Grimm, 1976; Yaron et al., 1978; Billard et al., 1978; Scott and Sumpter 1980; Fostier et al. 1983), while for other it is low (about 1ng/ml) (Yaron and Levavi-Zermonsky, 1986; Ouchi et al., 1988; Greeley et al., 1988). As far as we know, Mc Gregor et al. (1981) were the first to find large difference of range between androgen and estrogen levels in *Scomberomus cavalla* (30 ng/ml for androgens and only 5 ng/ml for estradiol during gametogenesis). They also present estrogen profiles in *Pomatomus saltator* with no correlation with oocyte development. These authors found these results unexpected and could not explain them. Since no data is yet available on measurements in wild Siberian sturgeon no definitive conclusion could be reached. There is however no correlation between estradiol levels and oocyte development and a great difference of range between androgen and estrogen levels ; this result could be considered as the expression of an artificial phenomenon.

The fall in estrogens occured just after the change in the diet administered daily to the fish. Before the change from Diet 1 to Diet 2, males and females had high estradiol levels (80 ± 20.8 ng/ml for females, 30 ± 14.5 ng/ml for males). On another fish population fed with Diet 1 we found that after 10 years of diet administration estradiol levels reached 26.7 ± 4.7 ng/ml in males which was higher than in females (Pelissero and Le Menn, 1988). A measure of steroid in the first diet showed large amounts of estradiol (375 ± 34 ng/100g) (Pelissero and Le Menn, 1988; Pelissero et al., 1989a). Then we

assumed that the unexpected estradiol levels in males could be due to dietary contamination since a change from Diet 1 to Diet 2 (only 47 ± 9 ng/100g), induced a significant decrease of the estradiol levels. This influence played a role in vitellogenin synthesis (Pelissero et al., 1989) but also could act on growth and energy dissipation in males.

The new diet is probably responsible for the phenomenon observed. Despite commercial secrecy we have acertained that in this new diet a part of the animal protein, provided by fish meals has been replaced by vegetal protein provided by soya. In Diet 1 the treatment of fish meals is different. This explains the difference observed in estradiol amounts in the diets. But the use of soya as fibers is known to bring isoflavonic phytoestrogens. These molecules could act directly or after being transformed in estrogenic compounds like "equol" by the intestinal tract bacteria (Axelson et al., 1983). They could then be implicated in many reproductive problems in mammalian species (Tang and Adams, 1980, in rat; Thompson et al., 1984, in macaques; Kaziro et al., 1984, in sheep; Adlercreutz et al., 1987, in man). Equol has been identified by GC - MS in sturgeon plasma (Pelissero et al., 1989b). This isoflavonic derivate and also lignans and phytoestrogens such as enterolactone, enterodiol and O-desmethylangolensin identified in large quantity in vegetarian women urine (Adlercreutz et al., 1986 a, b, 1987), has been shown to act on estradiol metabolism in man (Adlercreutz et al., 1986b; Adlercreutz et al., 1987). Its main known effect is to decrease the percentage of plasma free estrogens by inducing the synthesis of sex hormone-binding globulin (SHBG). Two other works from Goldin and coworkers (Goldin et al., 1982; Goldin et al., 1986) showed that diets caracterised by fiber-rich and low-fat composition could decrease estrogen levels in blood and urine in man by interfering partly with the biliary estrogens reabsorption. It also could act in other ways on estrogen metabolism. These effects are perhaps less complex in fish but since any data is not yet available, the hypothesis of a soya effect on estradiol metabolism could thus explain the results found here.

The GC - MS analysis failed to identify 17ß estradiol in vitellogenic females confirming the low plasma level of this hormone during vitellogenesis. It also pointed up the large amount of cholesterol in sturgeon plasma. This has already been demonstrated in various salmonid species (Farrell and Munt, 1983) and according to these authors it might not be correlated directly to diet. Deb and Bhattacharya (1986) demonstrated that the plasma levels of cholesterol change during reproductive cycle and seem to be involved in ovarian steroidogenesis as principal precursor. Large amount of cholesterol could perhaps be also linked to the gametogenesis in sturgeon.

It appears, that in fish farm conditions, sex steroid profiles in sturgeon do not evolve in a way similar to that observed in other fish species. They are dependent on the diet brand used for the fish. When androgen levels vary in relative accordance with that known in other fish, estrogen levels and especially estradiol ones, are not correlated to oocyte development. They constantly decrease after the change from Diet 1 to Diet 2. These levels obtained by RIA do not seem to retard vitellogenesis nor oocyte development. This suggests a dietary influence on sex steroid profiles in the cultured sturgeons. This effect may result of the presence on isoflavonic phytoestrogens in commercial complete diets. These data point out that the classical diet for trout is not well adapted to sturgeon. It will be of interest to obtain a special sturgeon diet taking into acount the dietary particularity of *Acipenser baeri* (Ruban and Sokolov, 1986 ; Sokolov et al., 1986).

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VITELLOGENIN : POTENTIAL VEHICLE FOR SELENIUM BIOACCUMULATION IN OOCYTES OF THE WHITE STURGEON (*Acipenser transmontanus*)

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RESUME

La concentration en sélénium (fluorimétrie) dans les ovocytes et le sérum d'esturgeons blancs en vitellogenèse, capturés dans la baie de San Francisco et dans la rivière Sacramento, est relativement élevée et variable. Elles varient de 3,06 à 29,26 µg/g et de 5,39 à 9,33 µg/g (poids sec) respectivement dans les ovocytes et le sérum. Les diverses composantes moléculaires du vitellus et du sérum ont été obtenues par filtration sur gel (Sepharose 6B-100) et chromatographie échangeuse d'ions (DEAE-Sephacel). Pour deux composants des plaquettes vitellines, la lipovitelline et la phosvitine, la concentration en sélénium est de 3,14 à 11,22 µg/g et de 3,17 à 90,31 µg/g respectivement ; tandis que dans l'immunoglobuline on en a trouvé de 0,76 à 6,70 µg/g. Dans la vitellogénine, les teneurs sont comprises entre 2,78 à 4,11 µg/g. Nous avons mis en évidence une relation linéaire significative entre, d'une part les concentrations en sélénium de l'oeuf entier et celles de l'immunoglobuline et de la phosvitine séparemment et, d'autre part entre celles de l'immunoglobuline et de la phosvitine ($P<0,05$). Les données indiquent que le sélénium est transporté par les protéines plasmatiques puis incorporé dans les plaquettes vitellines, la phosvitine et la lipovitelline. Nous faisons l'hypothèse que les séléno-amino acides s'incorporent à la vitellogénine puis passent dans les composants du vitellus.

Mots clés : esturgeon blanc, *Acipenser transmontanus*, sélénium, vitellogénine, protéines vitellines.

ABSTRACT

Adult vitellogenic white sturgeon caught in the San Fransisco Bay and Sacramento River have relatively high and varied concentrations of selenium in the oocytes and plasma as determined by fluorimetry. Ova concentrations ranged from 3,06-29,26 µg/g and plasma from 5,39-9,33 µg/g (dry weight). Egg yolk and plasma were resolved into their molecular components using gel filtration (Sepharose 6B-100) and ion exchange (DEAE-Sephacel) chromatography. Yolk platelet components, lipovitellin and phosvitin, exhibited selenium concentrations of 3,14-11,22 µg/g and 3,17-90,31 µg/g, respectively ; whereas, egg immunoglobulin selenium ranged between 0,76-6,70 µg/g. Vitellogenin selenium concentrations ranged from 2,78-4,11 µg/g. There were significant linear relationships between the selenium levels of whole ova and both egg immunoglobulin and phosvitin, and between the selenium content of egg immunoglobulin and phosvitin ($P<0,05$). The data indicate that selenium carried by plasma proteins is incorporated into the yolk platelets, phosvitin and lipovitellin. The potential incorporation of the selenoaminoacids into vitellogenin and transfer to the yolk molecules are suspected.

Key-words : white sturgeon, *Acipenser transmontanus*, selenium, vitellogenin, yolk proteins.

INTRODUCTION

Selenium is an essential micronutrient that can be highly toxic to the developing embryo at elevated tissue concentrations (Ohlendorf et al., 1986). Ecological studies indicate that fish reproduction is affected by selenium bioaccumulation (Lemly 1985, Gillespie and Baumann, 1986). Potentially, selenium can be incorporated into the vitellogenin (VG) molecule and subsequently into the egg yolk. This may influence ovarian development and survival of the progeny. The likelihood of such a mechanism is high considering vitellogenin properties (Plack et al., 1979; Shackley, 1981) and reported selenium incorporation from blood to milk proteins in goats and cows (Allen et al., 1981).

Within the past several years the increasing selenium concentration in the San Joaquin River and upper San Fransisco estuary have caused concern. The water quality criterion for selenium to protect freshwater life is 35 µg/L (USEPA, 1980). However, the San Joaquin River basin areas, such as the Kesterson Refuge and San Luis Drain, range from 140-1,400 µg/L (Tanjii et al., 1986). The white sturgeon is anadromous, migrating annually between the river and bay areas to forage and spawn. Due to the sturgeon's longevity and demersal feeding habits, selenium may accumulate beyond safe levels in selected tissues. The concentration of selenium in developing sturgeon ovaries from the San Fransisco Bay ranged from 1,0-71,8 µg/g on a dry weight basis (Kroll and Doroshov, unpublished observations). Due to the complex composition of vitellogenin molecule, the phosphate, lipid, and protein components can incorporate and carry potentially toxic molecules into the developing oocytes. Studies have proven that VG carries cations such as copper and cadmium by phosphate interactions (Shackley, 1981) and DDT (2,2-bis(p-chlorophenyl)-1,1,1-trichloroethane) in the lipid residues of the molecule (Plack et al., 1979). However, to our knowledge, no study has determined the route(s) of selenium accummulation in the ovary.

The purpose of this preliminary study is to determine if selenium is being incorporated into the egg yolk platelets of the sturgeon, and to investigate quantitative distribution of selenium between the blood and egg protein compartments.

MATERIALS AND METHODS

Fish

Blood and vitellogenic oocytes were obtained from six white sturgeon (range : 18,46-113,38 kg, body weight) caught in the San Pablo Bay by gill net (obtained from California Department of Fish and Game) or by hook in the Sacramento river (obtained from Sea Farm of Norway, and The Fishery, Inc.). Females were at similar stage of ovarian development having oocytes at or near mature diameters (3,6-3,8mm).

Sample processing

Freshly dissected ovaries or ovulated eggs were rinsed with 0,9% NaCl, homogenized, and then filtered to remove egg membranes. Egg yolk proteins were isolated from this aqueous phase using a modified method described by Markert and Vanstone (1971) and Hara and Hirai (1978). The aqueous phase was centrifuged at 11000 x g for 30 min (4°C) and the middle yellowish layer was recentrifuged at 45000 x g for 60 min (4°C) to separate any remaining cell materials and lipids. The resulting solution was lyophilized and stored at -70°C. The platelet proteins from this crude egg yolk preparation was purified by reconstituting to 2-3 ml (with doubly distilled water) and resolving on gel filtration media.

Blood was collected from the caudal vein in heparinized vacutainers and immediately centrifuged (4000 x g, 10 min, 4°C). Plasma was stored at -20°C. Whole plasma was lyophilized for selenium analysis on a dry weight basis (µg/g).

Vitellogenin levels were determined by measuring alkaline labile protein phosphorus (ALPP) as previously described by Wallace and Jared (1968), De Vlaming et al. (1984), and Moberg et al. (in press).

Column chromatography

Vitellogenin (VG) and the yolk proteins, lipovitellin and phosvitin, were purified using gel filtration and ion exchange chromatography. Plasma (2-3 ml) or crude egg yolk proteins (2-3 ml) was loaded onto gel filtration column (Sepharose 6B-100), 90 x 2,6 cm, and resolved with an eluent buffer consisting of 0,020M Tris-HCl, 0,05% Na-Azide, and 0,35M NaCl (pH 8,0). Flow rate was maintained at 20,0 ml/hr and fractions of 6,0 ml were collected. The protein absorbance of each fraction was measured at 280 nm (Perkin-Elmer Lambda 3B Spectrophotometer).

Due to the low levels of vitellogenin it is masked by the neighboring proteins. Hence, it was necessary to collect the gel fractions at the VG region (400000 D) and then repurify by ion exchange chromatography (DEAE-Sephacel) using a linear NaCl gradient (0,0-0,3M NaCl, 0,05% Na-Azide, 0,050 M Tris-HCl, pH 8,0) as described by Norberg & Haux (1985). After thorough dialysis the VG was lyophilized and processed for selenium content, but only for 4 females. All other plasma and yolk components were purified using only gel filtration methods.

Identification of egg proteins-immunoglobulin (660000 D), lipovitellin (190000 D), phosvitin (64000 D) and vitellogenin (360000 D) was acccomplished by comparing to molecular weight standards : Thyroglobulin (669000 D), Apoferritin (443000 D), b-Amylase (200000 D), Alcohol Dehydrogenase (150000 D) and with data obtained in previous studies (Acton et al., 1971 ; Hara and Hirai, 1978 ; Plack et al., 1979). Moreover, VG was identified by determining the phosphorus content (ALPP) of purified proteins, since, VG is the major phospho-protein in the blood and contains >95% of plasma phosphorus (Kroll and Doroshov, unpublished observations).

Gel filtration fractions (2-3) at the center of each protein peak were pooled and dialyzed (Spectrapor 1) for 4 days in doubly distilled water (changed 3 times a day) to remove salts. Thereafter, the material was lyophilized, and stored at -70°C. Fractions containing vitellogenin for further purification on ion exchange matrix were dialyzed overnight and stored in a dessicator prior selenium analysis. All chromatographic preparations were conducted at 4°C.

Selenium analysis

Total selenium was determined by fluorimetry as selenite using methods described by Brown and Watkinson (1977), and Whetter and Ullrey (1978). Plasma (1000 µl), purified plasma proteins (5-10 mg), whole eggs (0,25-0,30 g),or purified egg yolk proteins (5-10 mg) were digested with 5,0 mls of concentrated HNO3 and 2,0 ml HCLO4. Digestion was carried out at 150°C for 1,5 hr and then at 210°C for 1,25 hr. Reduction of selenate to selenite was accomplished by adding 3,0 ml of 1,2M HCL to the cooled solution and returning to the block for 3-4 min (150°C). The solution was recooled and then 2,5 ml of EDTA (0,016M) was added.

This final solution was adjusted to pH 1,0 using cresol red indicator, 10M NH4OH and 1,2M HCl, respectively. Samples were diltuted quantitatively with 0,1M HCl and compared to selenite standards (0,001-0,010 ppm) and blanks using a fluorimeter (Perkin-Elmer 650-15) All reagents were of analytical grade and selenium free. Standard recovery was 98-100%.

Data were analyzed using descriptive statistics and least squares regression ($P<0,05$).

RESULTS

The concentrations of selenium in plasma, vitellogenin and egg protein components are presented in Table 1. All six females from which the samples were collected, were in vitellogenic state, as confirmed by the elevated plasma protein-phosphorus (e.g. ALPP was above the base level observed in nonvitellogenic females and males). Plasma of these fish contain 0,27± 0,02 µg/ml selenium ; whereas, the lyophilized plasma proteins contained substantially higher selenium concentrations (6,95 ± 0,56 µg/g). The ova selenium concentration was 12,42 ± 3,31 µg/g (dry weight).

Selenium distribution in the egg yolk protein fractions are shown in Figure 1 and Table 1. Among the egg yolk proteins, the highest concentations were found in phosvitin (25,56 ± 12,30 µg/g), although there was very high variation between individual fish (3,17-90,31 µg/g). Phosvitin had about four times the selenium concentration as compared to lipovitellin (6,75 ± 1,16 µg/g), and egg immunoglobulin had lowest value (2,75 ± 0,82 µg/g).

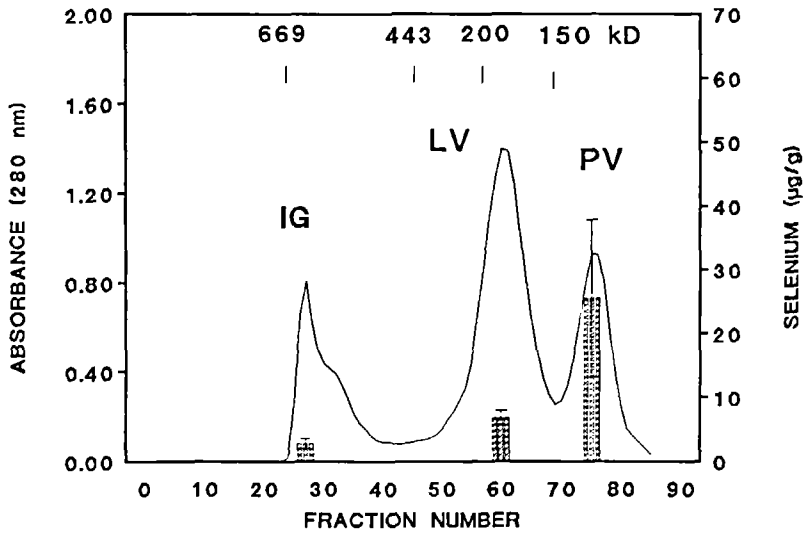


Figure 1 - Elution pattern and mean selenium content of white sturgeon egg yolk proteins (n=6). Selenium concentration is indicated by bars (x ± s.e.m.) and protein absorbance (280 nm) by a line. Proteins were resolved on Sepharose 6B-100 (2,6 x 90 cm). Flow was maintained at 20,0 ml/hr and fractions of 6,0 ml were collected. Molecular weight markers in kilodaltons (kD) are designated at the top of figure. Egg immunoglobulin, lipovitellin, and phosvitin are indicated by IG, LV, and PV, respectively.

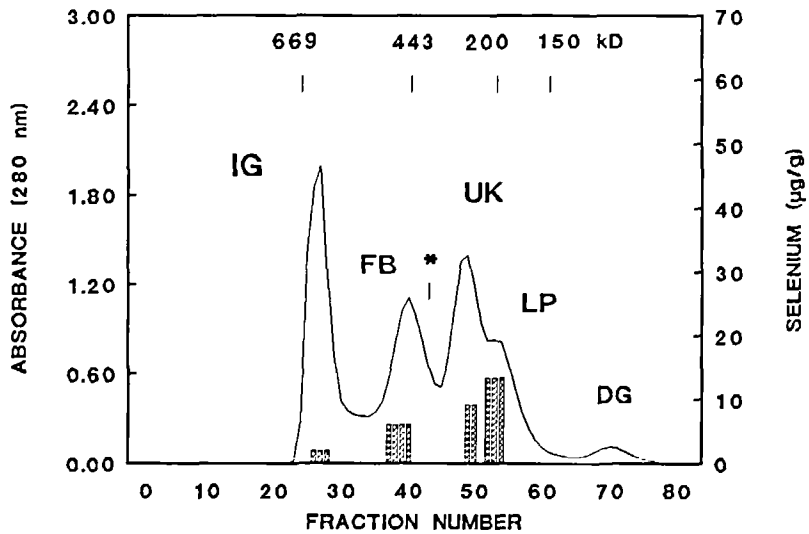


Figure 2 - Plasma elution pattern and selenium content of proteins from a vitellogenic white sturgeon (female # 2, Table 1). Proteins were resolved as described in Figure 1. Plasma peak identities are assumed as follows : immunoglobulin (IG), fibrinogen (FB), unknown (UK), lipoprotein (LP), and vitellogenin degradation (DG). The area designated with an asterisk corresponds to the vitellogenin region ; however, this sample contained very low levels of VG. Further purification was accomplished by pooling VG fractions and then resolving by ion exchange chromatography (DEAE- Sephacel).

Plasma protein profiles from a vitellogenic sturgeon display differing degrees of selenium concentration (Figure 2). The highest content appears in the protein at 200000 Daltons. The VG peak (400000 D) is not prominent due to the low levels in these females. Further purification required ion exchange chromatography.

In spite of the small sample size, there were statistically significant linear relationships between the selenium concentration in whole ova and both egg immunoglobulin and phosvitin. This indicates direct interactions between the platelet yolk components and whole ova , regarding selenium densities. The coefficients of correlation, regression F and regression coefficient values were significant at $P<0,05$. (Table 1). Vitellogenin re-purified on ion exchange matrix also contained substantial selenium concentrations, 3,39 ± 0,25 µg/g (Table 1).

Table 1 - Selenium concentrations in plama and yolk compartments of wild white sturgeon from the San Francisco Bay and Sacramento River

	Selenium concentrations								
Female	Wt (kg)	ALPP (µg/ml)	Plasma (µg/ml)	Plasma (µg/g)*	Whole ova (µg/g)	VG (µg/g)	IG (µg/g)	Lipovitellin (µg/g)	Phosvitin (µg/g)
1	113,38	22,00	0,19	6,18	3,06	na	0,93	4,69	4,17
2	18,58	17,00	0,32	9,33	9,64	na	3,61	9,74	28,42
3	20,72	45,00	0,28	7,89	12,44	4,11	1,94	5,35	11,94
4	18,46	50,00	0,25	7,28	11,64	3,10	2,58	11,22	15,33
5	21,00	90,00	0,28	5,39	8,46	3,58	0,76	3,14	3,17
6	26,30	70,00	0,33	5,63	29,26	2,78	6,70	6,38	90,31
mean	36,41	49,00	0,27	6,95	12,42	3,39	2,75	6,75	25,56
s.e.m.	14,09	10,40	0,02	0,56	3,31	0,25	0,82	1,16	12,30

(* - lyophilized whole plasma, na- insufficient sample)

Regression Analysis

Components (Y:X)	Regression Equation	R^2	F value
OV : IG	Y = 4,08X - 0,44	0,83	18,70
OV : PV	Y = 0,25X + 5,95	0,88	30,20
PV : IG	Y = 14,59X - 14,61	0,95	74,20

(Immunoglobulin (IG), whole ova (OV), vitellogenin (VG), and phosvitin (PV) all based on dry weight

DISCUSSION

The most probable site for selenium incorporation is the poly peptide backbone of vitellogenin. Since the chemical characteristics of selenium are similar to sulfur, it can be substituted into the sulfur amino acids : cysteine, cystine, methionine, and homocysteine (N.R.C.S.S., 1983). Although the composition of sturgeon yolk is not yet established, data on salmonids indicates that both VG and the yolk platelet molecules contain small, but appreciable percentages of these amino acids (Table 2). The phosvitin

molecule contains a greater number of possible substitution sites in rainbow trout and is composed mainly of protein, whereas lipovitellin has fewer sulfur groups and contains about 25% lipid by weight (Campbell and Idler, 1980). This may explain higher selenium concentrations in the phosvitin molecule. Moreover, if selenium is incorporated only at this level, the yolk molecules would have a finite level of saturation. Hence, the sensitivity of developing embryos and larvae to selenium bioaccumulation may depend on the magnitude of selenoaminoacid substitution and be species-specific.

Table 2 - Sulfur containing amino acids of vitellogenin, lipovitellin, and phosvitin in the rainbow trout (Hara & Hirai, 1978)

Amino Acid	Vitellogenin	Lipovitellin	Phosvitin
CYS/2	1,18	0,0	2,74
MET	2,55	2,55	3,03
Total	3,73	2,55	5,77

(result expressed as moles/100 moles of amino acid)

Linear relationships between the selenium burden of ova and either egg phosvitin or immunoglobulin, as well as correlation betweeen selenium in phosvitin and immunoglobulin, suggest potential incorporation of selenoaminoacids into the egg yolk protein compartments from vitellogenin molecules. The existence of such relationships provide indirect evidence that accumulation of selenium in yolk platelets is controlled by changes of selenium in plasma compartments, most likely, in vitellogenin and lipoprotein fractions. The role of the vitellogenin molecule as a vehicle for selenium transfer into the egg yolk requires further studies and clarification.

Unfortunately, little information is available on the identity of the major plasma proteins in fish. Acton et al. (1971) have determined that the protein at 660000 D is an immunoglobulin. The protein at 500000 D appears to be fibrinogen; its quantity varies with the addition of heparin (Kroll and Doroshov, unpublished observations). The region at 400000 D coincides with vitellogenin ; however, since VG is at very low levels compared to other proteins it is masked by the neighboring proteins. The protein at 200000 D appears to be a high density lipoprotein (Plack, 1979). However, these conjectures should be substantiated by a thorough chemical analysis of each protein.

Analysis of the major plasma proteins indicate that both lipoprotein and vitellogenin contain appreciable amounts of selenium (Figure 2). However, the role of plasma lipoprotein in vitellogenesis is not understood. Due to the low levels of vitellogenin in sampled females, our data is limited and does not permit evaluation of correlations between the selenium in VG and other components.

At present, the relationships between selenium levels in the yolk compartments and progeny survival is uncertain for fish. Studies with the bluegill provide some indication that ovarian selenium concentration correlates with reproductive failure (Gillespie and Bauman, 1986). Embryos resulting from ovaries containing 38,6 µg/g (dry weight) exhibited 65% mortalities. At the selenium ovarian concentration of 44,53 µg/g there was 100% mortality of hatched embryos. The dry weight value (used by authors) is based upon the assumption that the ovary contains 85% water ; however, it is known that the water content of ovaries vary considerably at different reproductive stages (e.g. immature, vitellogenic, post-spawn, etc...). Even though reliable data on critical ovarian concentrations are absent for other fish species, the range of selenium concentrations found in sturgeon ova (3-29 µg/g, Table 1) indicates abnormal conditions.

It is apparent from our study that selenium carried by plasma proteins is incorporated into the yolk platelets of the white sturgeon. This suggests that long term exposure to seleniferous water or food supply may be harmful to reproduction of this

valuable species. Information concerning effect of selenium accumulated in maternal yolk on the survival of the sturgeon embryo, is needed to clarify this effect. In addition, the bio-accumulation and turnover of selenium in the ovarian tissue of fish is not understood at the mechanistic level. More studies are required to interpret "normal" versus "toxic" levels of selenium in fish eggs and the sensitivity of different species to elevated yolk selenium, during embryonic development.

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ELISA FOR SIBERIAN STURGEON (*Acipenser Baeri* Brandt) VITELLOGENIN

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RESUME

Nous décrivons ici un dosage ELISA (enzyme-linked immunoassay) simple et spécifique pour la vitellogénine (VG) de l'esturgeon sibérien *Acipenser baeri*. Ce dosage est basé sur la compétition entre Vg soluble et la Vg adsorbée par les plaques de microtitration pour les sites de fixation de l'anticorps anti-Vg développé chez le lapin. Les complexes Vg-anticorps, adsorbés sur la plaque, sont alors révélés par la technique peroxydase-anti-peroxydase et lus à 492 nm. L'intensité de la réaction est proportionnelle à la quantité d'anticorps lié à la Vg adsorbée.

La sensibilité du test est d'environ 3 ng/puits avec une variation intra-dosage de 10 % (n=30) et une variation inter-dosage de 11 % (n=40) aux environs de 50 % de liaison.

On distingue 5 principales étapes :

1 Greffage de l'antigène : il est réalisé par une solution de Vg à 500 ng/ml dans du tampon carbonate 0.05 M pH 9.6.

2 Incubation avec l'anticorps spécifique : (dilution 1/20000) avec des dilutions sériées des échantillons ou du standard (Vg).

3 Incubation avec le second anticorps : IgG de proc anti-IgG de lapin dilué au 1/2000.

4 Complexes peroxydase-anti-peroxydase : dilué au 1/5000.

5 Révélation des complexes adsorbés par une solution d'ophénylènediamine et d'eau oxygénée dans un tampon citratephosphate 0.1 M pH 5.

Les données sont traitées selon deux modèles :

1 Bi/Bo = F (log(dose)) ou F (log (dilution)),

2 Logit (Bo-Bi/Bi-N) = F (log(dose)) ou F (log(dilution)).

Cette transformation (2) permet de réaliser une régression linéaire de type Y=AX + B par la méthode des moindres carrés. La comparaison des droites de régression est basée sur l'analyse de covariance après avoir, au préalable, testé l'homogénéité des variances par le test de Bartlet.

Nous observons le parallélisme entre la Vg et les dilutions de plasmas de femelles ainsi que la plupart des plasmas de mâles testés. Ces résultats prouvent donc la présence de VG chez les mâles élevés en pisciculture.

Mots clés : *Acipenser baeri*, ELISA, vitellogénine, plasma.

ABSTRACT

A specific and simple enzyme-linked immunoassay for the sturgeon (*Acipenser baeri*) vitellogenin (VG) is discribed. This assay is based on the competition between soluble VG and VG adsorbed on microtiter plates, for the rabbit anti-VG antibody binding sites. The adsorbed VG-antibody complexes are then revealed using the peroxidase-anti-peroxidase technique and read at 492 nm. The intensity of the reaction is proportional to the amount of the antibody linked to the adsorbed VG.

The sensivity was about 3 ng/well with an intra-assay variation of 10 % (n=30) and an inter-assay variation of 11 % (n=40) near 50 % of binding.

Five main steps can be defined :

1 Antigene-coating : VG was coated using a 500 ng/ml solution of VG in 0.05 M carbonate buffer pH 9.6.

2 Specific antibody incubation : diluted 1/20000, with serial dilutions of samples or of the reference preparation.

3 Second antibody incubation : swine IgG anti-rabbit IgG diluted 1/2000.

4 Peroxidase-anti-Peroxidase complex incubation dilutes 1/5000.

5 Revelation of the adsorbed complexes by o-=phenylene-diamine and hydrogen peroxide in citrate-phosphate buffer 0.1 M pH 5.

The data were expressed using two models :

1 Bi/Bo = F (log(dose)) or F (log (dilution)),

2 Logit (Bo-Bi/Bi-N) = F (log(dose)) or F (log(dilution)).

Transformation (2) allowed us to fit a least squares linear regression. Comparison of regression curves was based on covariance analysis after having tested the homogeneity of their variance by the Bartlet test.

Parallel displacement was verified with the standard preparation serial dilutions of females plasmas and also with most of the 14 males plasmas tested. These data provide an argument for the presence of VG in the plasma of reared male sturgeon

Key-words : *Acipenser baeri*, ELISA, vitellogenine, plasma.

INTRODUCTION

The variations of plasma vitellogenin (Vg) levels in fish are a good parameter for the knowledge of sexual cycle and vitellogenesis development.

Indirect methods have been used to estimate plasma Vg concentrations :

- quantitative immunoelectrophoresis (Goedmaker and Verbroom, 1974)
- alkali-labile phosphorus evaluation (Martin and Doty, 1949 ; Craik and Harvey, 1984)
- indirect semi-quantitative immunodiffusion (Plack et al., 1971).

Recent progress in radioimmunoassay (RIA) (Idler et al., 1979 ; Campbell and Idler, 1980) and enzyme linked immunosorbent assay (ELISA) technique (Nunez Rodriguez et al., 1989) allow now to measure plasma Vg levels and correlate them to oocyte stages during gametogenesis. These techniques have high sensitivity compared to the previous ones which are not reliable quantitative methods.

This paper deals with the development of an ELISA for Siberian sturgeon *Acipenser baeri* Vg and with a comparison of Vg plasma levels between two stocks during vitellogenesis.

MATERIAL AND METHODS

Siberian sturgeons were imported into France from USSR in 1982 and reared either in the INRA fishfarm-CEMAGREF hatchery of Donzacq (Landes) at a constant temperature of 17°C, or in a private fishfarm in Biganos (Gironde) in natural conditions of temperature (minimum : 6°C ; maximum : 18°C).

For this study 50 seven year-old fish were used :
18 females (oocyte stages : III,IV and V),
7 males,
from the two different stocks.

Fish were sexed by biopsy of the gonad followed by light microscopy. The different stages of oogenesis were identified by electron microscopy using a Jeol 100S microscope (Departement de Microscopie Electronique, Université de Bordeaux I) (Le Menn and Pelissero, 1991).

Sturgeon Vg was purified from plasma of 17 -estradiol treated male and female fish. Blood was collected by the caudal vasculature in heparinized tubes. After centrifugation (5 min at 10000 g) plasma was stored at -20°C in 1 ml aliquots. Purification was performed using a two-steps technique (6 sepharose gel filtration followed by DEAE trisacryl anion exchange chromatography).

Vg immunserum was obtained in rabbits according to the procedure used by Pelissero (1988). The antiserum was tested by radial immunodiffusion in 1.5 % agarose gel (Ouchterlony, 1949).

ASSAY PROCEDURE

This assay is based on the competition for the antibody between Vg in sample and Vg adsorbed on microtiter plates . The adsorbed antigen-antibody complexes are then revealed using the peroxidase anti-peroxidase technique and optical density is measured at 492 nm.

The assay procedure is similar to that described by Nunez Rodriguez et al. (1989) with some modifications : coating by 500 ng/ml of Vg ; specific antibody diluted 1/20,000 ; preincubation for 24 h at 37°C.

RESULTS

The sensitivity (Vg amount which gave 90% of binding) is 3 ng/well (i.e. 15 ng/ml) with an intra-assay variation 10% (n=30) and an inter-assay variation 11% (n=40) near 50% of binding.

Parallelism of 43 different standard curves was assessed by covariance analysis using the F-test on mean squares (Snedecor and Cochran, 1957). No statistical difference was observed between the different regression lines ($F_{obs} = 0.02 << F_{0.05} = 1.49$ with 42 ; 691 df).

Data concerning competition curves of various antigens are represented in Fig.1 : Parallel displacement was obtained with the standard preparation and serial dilutions of female plasma (Stage III,IV and V oocytes) and also with male plasma.

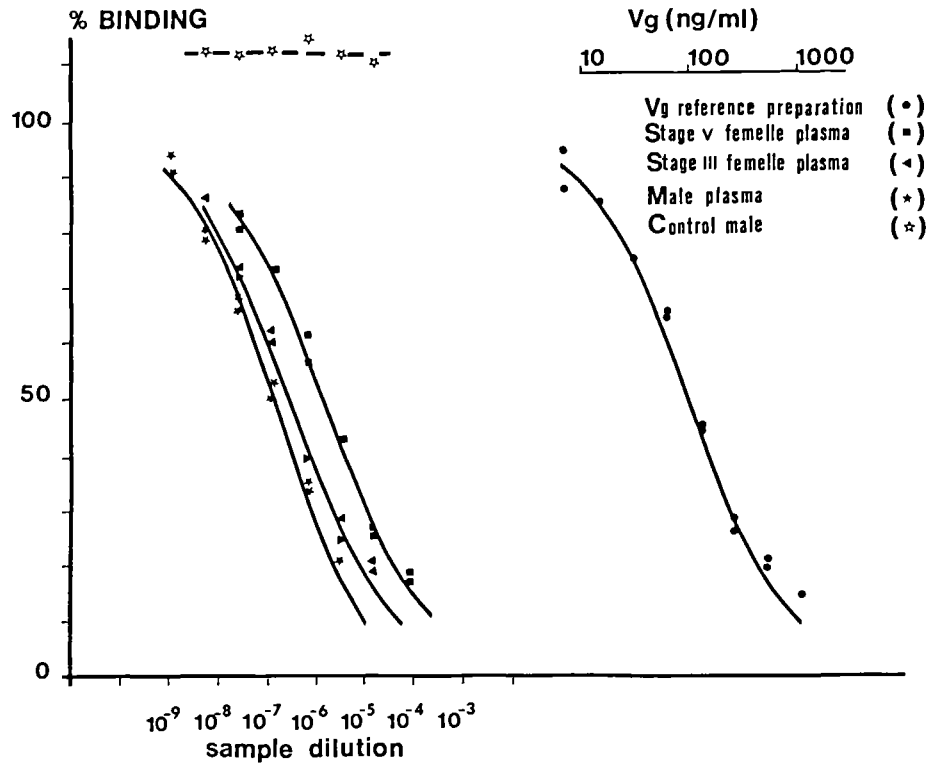


Figure 1 - Binding curves Bi/Bo = F (Log(Dose)) or F (Log(dilution))

This assay has been used to measure plasma Vg concentrations during the Siberian sturgeon reproductive cycle (table 1).

Fish issued from two fish farms were all undergoing their first sexual cycle and females which had never spawn presented different sexual development stages.

Table 1 - Plasma vitellogenin levels (mg/ml) measured in females (stage III, IV and V oocytes) and in males. Only two males among the 14 tested had non detectable Vg.

	DONZACQ	BIGANOS
Females (stage III oocytes)	X = 21.48 mg/ml SEM = 5.4 (n = 7)	X = 12.56 mg/ml SEM = 5.4 (n = 7)
Females (stage IV oocytes)	X = 25.98 mg/ml SEM = 5.8 (n = 7)	X = 27.57 mg/ml SEM = 5.1 (n = 7)
Females (stage V oocytes)	X = 10.59 mg/ml SEM = 3.7 (n = 7)	X = 59.03 mg/ml SEM = 42.1 (n = 2)
Males	X = 20.12 mg/ml SEM = 8.9 (n = 7)	X = 6.03 mg/ml SEM = 2.2 (n = 7)

DISCUSSION

The development of this ELISA now provides a direct reliable method for Vg measurement without involving any strong structural modification of the molecule. The good immunological reactivity and specificity is attested by the binding curve between specific antibody and control male (devoid of Vg), which shows that control males plasma does not compete for the antibody binding sites. On the contrary, the antibody recognizes in a similar way the standard Vg preparation and the native plasma proteins, as shown by the good parallelism between sample serial dilutions and standard (Fig.1).

We present here only preliminary results concerning Vg concentration in sturgeon plasma. Vg in female increases from the beginning of vitellogenesis (Stage III oocyte) to the Stage V in which starts oocyte germinal vesicle migration. However, the minimal Vg concentration observed here in sturgeon is 160 µg/ml in female with Stage III oocytes and reaches 100 mg/ml during Stage V . In the literature, Vg levels reported using RIA have never been inferior to 80-100 ng/ml in immature rainbow trout (*Salmo gairdneri*) but could reach 100 µg/ml in the same species mature fish (Sumpter, 1985) or 80 mg/ml in the brown trout (*Salmo trutta*) (Crim and Idler, 1978). In sole (*Solea vulgaris*) minimum Vg concentration were 100 mg/ml in female during the resting periode and it reached 10 mg.ml during the spawning period (Nunez Rodriguez et al., 1989).

The slight differences in sturgeon Vg levels between the two fish stocks can be related to the different water parameters and specially temperature which acts on metabolism, growth and sexual development of the fish (Craik, 1978).

In conclusion, the main interest of this study is to reveal and quantify the presence of Vg in males in concentrations as high as in stage III oocyte females. Only two males among the 14 tested had non detectable Vg. These animals were all reared in

fishfarms and fed with classical commercial fish diets. The occurence of Vg in male has been attributed to the presence of steroids in diets (Pelissero et al., 1988). Complementary studies tend to clearly suggest the possible action of these steroids on Vg synthesis (Pelissero et al, 1989). To definetely confirm these findings it would be interesting to check if Vg is detectable in wild fish with natural feeding conditions.

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HISTOLOGICAL AND ULTRASTRUCTURAL STUDIES OF OOGENESIS OF THE SIBERIAN STURGEON *Acipenser Baeri.*

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RESUME

L'ovogenèse de l'esturgeon sibérien *Acipenser baeri* est étudiée en microscopie photonique et électronique. Nous avons identifié 5 stades corrélés au stade physiologique des ovocytes.

Faisant suite au stade ovogonial, le stade I présente un index nucléoplasmatique élevé et le noyau contient un seul nucléole. Dans ces 2 stades précoces, les globules lipidiques ont déjà été identifiés. Le stade II est caractérisé par la multiplication des nucléoles et leur migration vers la périphérie du noyau. A ce stade, le nombre de globules lipidiques augmente dans le cytoplasme de l'ovocyte. A la fin de ce stade, les alvéoles corticaux commencent à être synthétisés. Le début du stade III correspond aux premières figures d'incorporation de la vitellogénine, parallèlement à l'élaboration de la zona radiata externa. C'est à ce stade que la zona radiata interna 1 est construite. Le stade IV est caractérisé par la première apparation de granules pigmentaires et par l'élaboration de la zona radiata interna 2. L'accumulation du vitellus augmente. Le stade V correspond au début de la migration du noyau vers la membrane ovocytaire. La zona radiata est achevée et le "jelly-coat", synthétisé par les cellules folliculaires, est déposé à la périphérie de la zona radiata externa. L'accumulation du vitellus s'achève à ce stade.

En conclusion, ces données ultrastructurales permettent la détermination précise des stades de l'ovogenèse. Le terme de prévitellogenèse ne peut être employé dans le cas d'esturgeons élevés en pisciculture étant donné que les réserves lipidiques sont déjà présentes dans les stades de l'ovogenèse précoce.

Mots clés : microscopie électronique, microscopie photonique, ovogenèse, *Acipenser baeri.*

ABSTRACT

Oogenesis of Siberian sturgeon *Acipenser baeri* is studied using light and electron microscopy. We have identified 5 stages correlated to the physiological state of oocytes.

Following the oogonial stage, Stage I presents an elevated nucleoplasmic index and the nucleus contains only one nucleolus. In these two early stages, lipid globules have been identified. Stage II is caracterised by the nucleoli multiplication and their migration towards the nuclear periphery. During that stage, the number of lipid globules increases in the oocyte cytoplasm. At the end of this stage, cortical alveoli begin to be synthesized. The beginning of Stage III corresponds to the first features of vitellogenin incorporation simultaneously with the elaboration of the zona radiata externa. During this stage, the zona radiata interna 1 is built. Stage IV is caracterised by the first apparation of pigment granules and the elaboration of the zona radiata interna 2. Yolk accumulation increases. Stage V corresponds to the beginning of nucleus migration towards the oocyte membrane. The zona radiata is completely synthesized and a jelly-coat is deposed at the perifery of the zona radiata externa by synthesis of follicular cells. Yolk accumulation end at this stage.

In conclusion, ultrastructural data allow oogenesis stages to be accurately determined. The term of previtellogenesis could not be used in the case of cultured sturgeon since lipidic reserves are already present in the early stages of oogenesis.

Key-words : electron microscopy, light microscopy, oogenesis, *Acipenser baeri.*

INTRODUCTION

Most studies of oogenesis in sturgeon species have been performed using light microscopy (Ginsburg et al., 1969; Caloianu-Yordachel, 1971; Kornienko, 1975; Trusov, 1975; Fedorova, 1976; Kondrat'yev, 1977; Akimova et al., 1979; Badenko et al., 1981;

Veshchev, 1982; Kijima and Maruyama, 1985). A few investigations used electron microscopy (Markov, 1975; Raikova, 1976; Cherr and Clark, 1982, 1984). We have not found studies dealing with *Acipenser baeri* gametogenesis, a sturgeon living in natural conditions in some Siberian rivers and lakes.

This species was introduced into France in 1974 and is reared in experimental fishfarms.

This paper deals with oogenesis of the Siberian sturgeon until the end of vitellogenesis. We underwent this study in order to be able to identifie different stages of the reproduction process and to determine the exact time for external hormonal intervention in order to induce artificial reproduction (Williot et al., 1982a,b).

MATERIAL AND METIIODS

Ovarian samples were taken from female sturgeon born in USSR in 1982, imported into France the same year and reared at the INRA fishfarm-CEMAGREF hatchery of Donzacq (Landes). Fish were raised at a constant temperature of 17°C and fed with artificial trout diets.

Biopsies were performed for light microscopy studies. Fixations were made with Bouin's fluid for 72 hours and tissue embedded in cytoparaffin. Serial sections 7 µm thick were stained with Groat eosin hematoxylin and periodic acid-Schiff. Immunocytochemical staining by peroxydase-antiperoxydase complex was used to show the incorporation of exogenous yolk into the oocyte at light microscopy level (Pelissero, 1988).

Every three months for three years some females, taken in the same aged batch, since 4 year-old to 8 year-old, were sacrificed for electron microscopy investigations. Gonads of stunned but live fish were perfused for 30 min with glutaraldehyde (3%) in sodium cacodylate buffer 0.04 M, $CaCl_2$ 0.05%, with an osmotic pressure of 300 mos at a pH of 7.4. The gonad was then cut in small pieces and immersed for 90 min in the same fixative. Pieces were washed three times for 15 min in a washing buffer of the same composition as the fixative fluid except that the glutaraldehyde was replaced by sucrose to obtain a similar osmolarity. Osmic post-fixation (2% OsO_4) in sodium cacodylate buffer 0.04 M for 30 min was followed by washing in graded ethanols for dehydration and immersed in propylene oxide. Samples were embedded in Epon 812. Semi-thin sections were stained with 0.1% toluidine blue and lipids were revealed by Azur blue 2 solution (Parry, 1973). Ultrathin sections were contrasted by uranyl acetate and lead citrate (Reynolds, 1963) and observed on a 100S JEOL (Département de Microscopie Electronique, Université de Bordeaux I).

RESULTS

We choosed a classification of oocyte stages based upon the distinction of the successive physiological steps in the process of the oocyte development.

Oogonia

Oogonia varied in size between 8 µm and 15 µm. Their nucleo-plasmatic ratio was greater than 1. They occured in clusters of 10 to 20 cells surrounded by the adipose tissue. Each cluster was enclosed by a common single layer of prefollicular cells with a basement lamina.

Ultrathin sections showed numerous lipid globules and mitochondrial agregates intermingled with the smooth endoplasmic reticulum. The mitochondrial agregates occured around masses of homogenous dense material which constituted the "ciment". Some intercellular bridges were observed.

Stage 1 follicle

When each oogonia gave rise to a Stage 1 oocyte, the follicular epithelium and the basement lamina invaginated the cluster and surrounded each oocyte (Figure 1b). The oocyte and its surrounding layers formed a follicle (Figure 1c).

We observed this stage for the first time in the gonad of some fish taken in a 1 -year old batch with a range of size about 40 cm and a range of weight about 800 g.

On semi-thin sections, a large centrally positioned round nucleus contained a few nucleoli close to the smooth nuclear envelope. The cell diameter varied between 20 µm to 80 µm. In the vicinity of the nucleus, a crown of lipid granules were observed. Thecal cells were present outside the basement lamina (Figure 1d).

The electron micrographs showed a large number of ribosomes in the cytoplasm intermingled with endoplasmic reticulum, mitochondria and lipid globules. Flattened follicular epithelium, closely adjacent to the oolemma exhibited ultrastructural features of metabolic activity (mitochondria, rough endoplasmic reticulum, Golgi complexes and coated vesicles). Coated pits were present on the outer follicular membrane against the basement lamina.

Stage 2 follicle

In this stage, the oocyte diameter varied between 80 µm to 120 µm.

It appeared for the first time in gonad of 3-year old fish with an average weight of 2 kg.

Semi-thin observations showed, with Azur blue coloration,an increase in the number of lipid globules (Figure 1e).The electron micrographs showed numerous small nucleoli located in the concavities of the wrinkles of the nuclear envelope near the inner nuclear membrane (Figure 1e,2c). In the cytoplasm, in the vicinity of the nuclear envelope, spots of electron-dense material similar to homogenous dense material linked with the mitochondrial agregates scattered throughout the cytoplasm as far as the outer periphery of the lipid globules crown (Figure 2a). The remaining part of the cytoplasm contained Golgi complexes with numerous Golgian vesicules. Some of those, closely adjacent to Golgi complex, exhibited pear-shaped features and seemed to discharge their content into a central smooth endoplasmic reticulum vesicle (Figure 2b). Numerous mitochondria were present in their vicinity. At the end of this stage, a few oocyte microvillosities protruded from the oolemma towards the follicular cells and the basement lamina exhibited a maximal thickness of about 10 µm (Figures 1e, 2b et 3).

Stage 3 follicles

Oocyte diameter varied between 120 µm and 600 µm. They appeared for the first time in the gonad of 3 1/2-year old female with a weight range of about 4 kg.

In light microscopy,a clear crown of lipid globules was observed in the medium cytoplasm. After PAS treatment, a very thin glucidic layer was exibited (Figure 1f). This was the first visualisation of the zona radiata located between the oolemma and the follicular epithelium. Small granules located in the peripheral ooplasm were PAS positive and corresponded to the cortical alveoli.

Cortical alveoli appeared on electron micrographies clustered of three or four near a Golgi complex and mitochondria. They contained fibrillar material. At the end of stage 2 follicule, numerous clusters of microvillosities appeared at the oocyte surface. Each cluster protruded into an hemispheric space between the oocyte surface and the follicular epithelium.

Follicular cells formed cytoplasmic bridges, still linked in places by desmosomes to the oolemma (Figure 3a,c). This space was filled with an electron-clear material showing a floculent extracellular matrix (Figure 3b). During the Stage 3 the follicle-oocyte junctions desappeared and the follicular epithelium remained entirely separated from the oocyte (Figure 4a). The oocyte microvillosities protruded vertically towards the follicular cells in the floculent extra-cellular matrix.

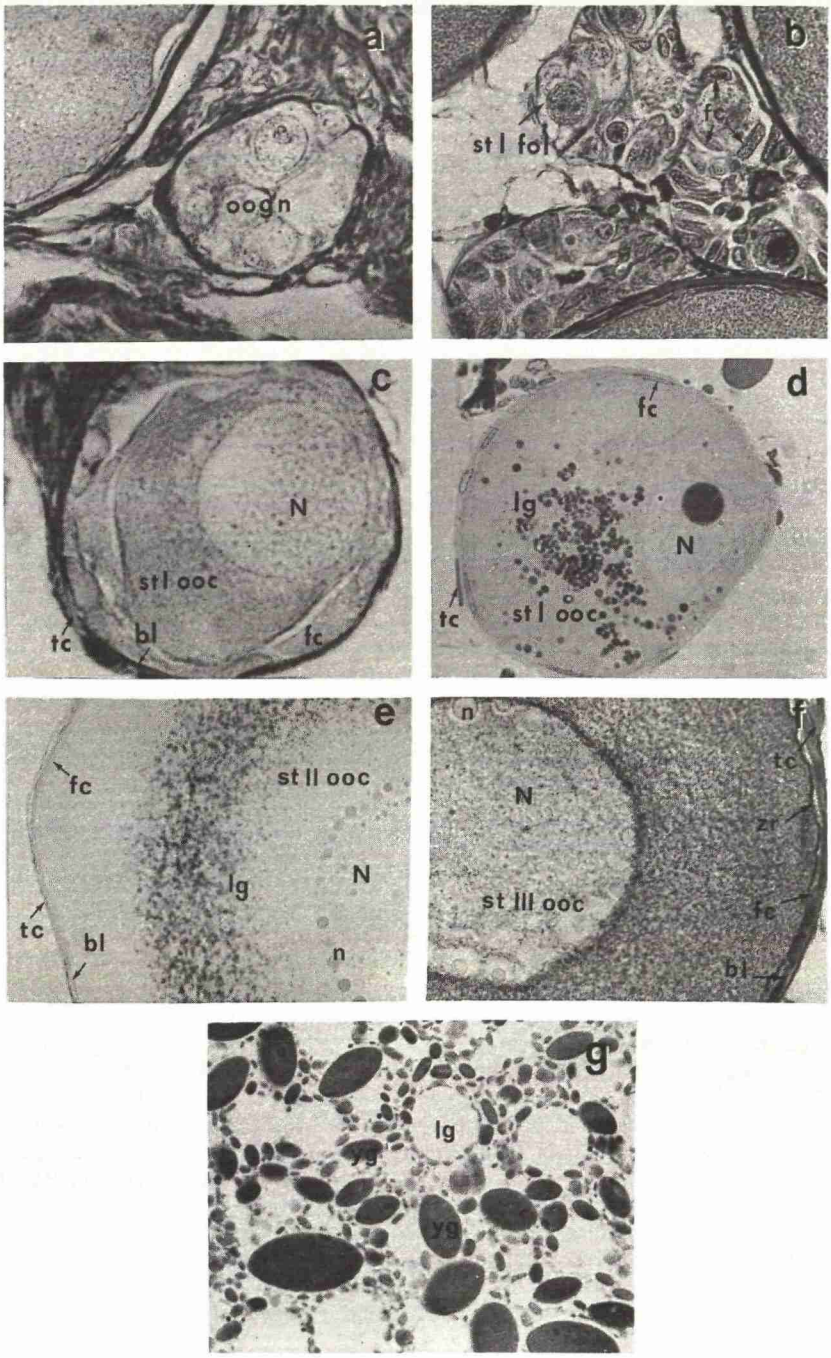


Figure 1- Photonic micrographies of ovarian follicle development. a- oogonie nest; b- early stage one follicle; c- stage 1 follicle; d- stage 2 follicle; e- old stage 2 follicle; f- stage 3follicle; g- stage 3 oocyte yolk. (bl: basement lamina, fc: follicular cell, lg: lipid globule, n: nucleolus, N: nucleus, ooc: oocyte, oog n: oogonial nest, thecal cell, yg: yolk globule).

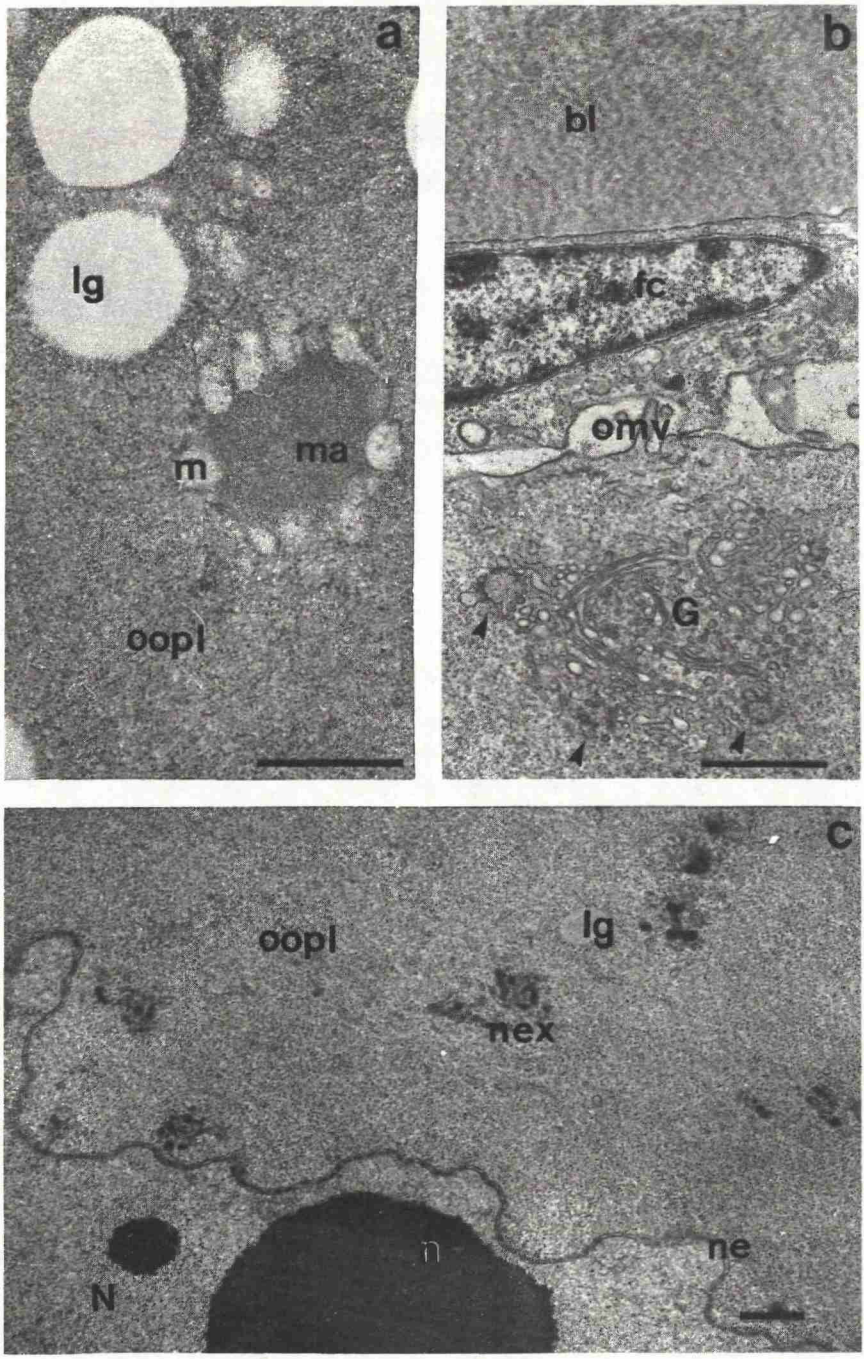


Figure 2- Early stage 2 follicle (electron micrographs). a- mitocondrial aggregate,b- peripheral ooplasm and follicular cells relationship, c- perinuclear area. (bl: basement lamina,fc: follicular cell, G: Golgi complex, lg: lipid globule, m: mitochondrial aggregate, n: nuleolus, ne: nuclear envelop, nex: nuclear extrusions, N: nucleus, omv: oocyte microvillosities, oopl: ooplasm, : cortical alveoli formation).

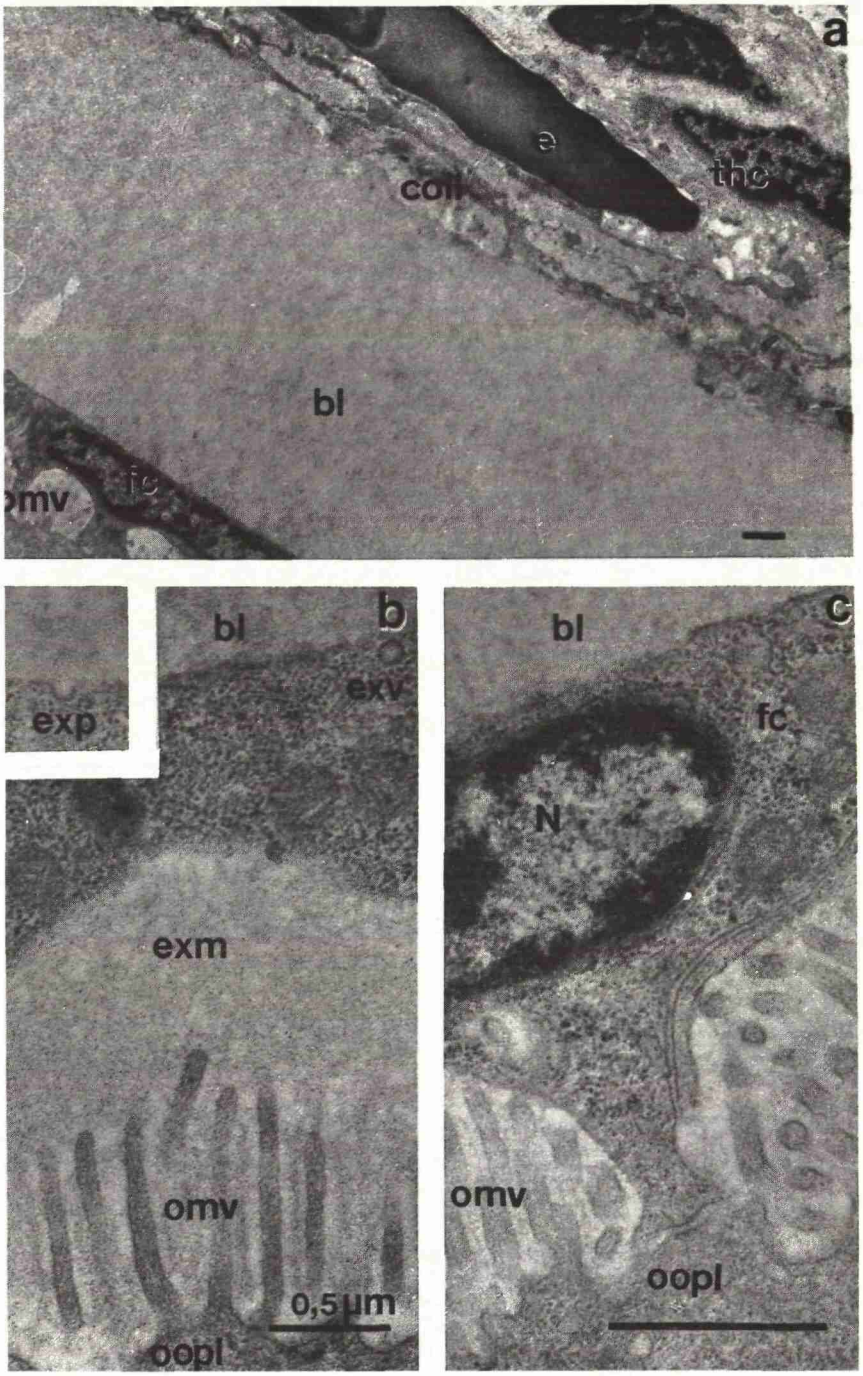


Figure 3- Stage 2 follicle (electron micrographies). a- follicle envelope; b,c- follicular cells and oocyte relationship. (bl: basement lamina, coll: collagen, e- erythrocyte, exm: extraoocyte matrix,exp: exocytotic pit, exv: exocytotic vesicle, fc: follicular cell, N: nucleus, omv: oocyte microvillosities, oopl: ooplasm, thc: thecal cell).

In this matrix, an electron-clear material, later becoming electron-dense was deposited around oocytes microvillosities thus building-up the zona radiata externa (Figure 4a,b). Perforations of this dense structure appeared as pores on tangential sections and as channels on transversal sections, through which the oocytes microvillosities passed (Figure 5e, f). Simultaneously, the first endocytotic features appeared between microvillosities on the oolemma which invaginated into pinocytotic pits coated with clathrin (Figure 4b). These pits gave rise to coated vesicles which fused to form the first yolk granules located in the peripheral ooplasm.

During Stage 3, the outer homogeneous layer constituting the zona radiata externa was gradualy displaced towards the follicular epithelium by succesive deposits of an inner electron-clear reticulated layer. This was the zona radiata interna 1 showing an highly organized parabolic fibrillar arrangement on oblique sections (Figure 5 e, g).

The basement lamina which was very thick at the previous stage (Figure 3a) decreased in thickness to 1 µm (Figure 5a).

Stage 4 follicle

This stage corresponded to the rapid vitellogenic growth phase leading to a considerable increase in oocyte diameter (900 µm to 2800 µm). It was found for the first time in 5-year old fish with a weight range of about 5 kg.

In light microscopy observations, yolk globules were first observed in the outer part of the cytoplasm, then in the whole cytoplasm intermingled with lipid globules. In the surrounding layers the basement lamina made so thin as to be not observable.

On semi-thin sections, yolk globules were ovoid in shape and lipid globules were spheric (Figure 1 g). The zona radiata interna exhibited two layers, a new zona radiata interna 2 with numerous narrow stratifications deposited under the zona radiata interna 1 (Figure 5 b, c).

Electronic micrographies indicated that the number of yolk globules increased considerably compared to those of the cortical alveoli and lipid globules. The yolk globules exhibited a crystalline part inside a granular matrix. Mitochondria and rough endoplasmic reticulum were scattered throughout these various inclusions. Pigment granules began to appear underneath the peripheral ooplasm and a very thick layer of mitochondria. These pigments showed a very electron-dense matrix inside a vacuola with dark granulations (Figure 6a).

Stage 5 follicle

At this stage events leading to maturation and ovulation began to occur. We observed such features in gonad of 6 year-old female with an average weight of 5.5 kg.

On semi-thin sections we observed that the nucleus migrated towards the periphery of the oocyte to the animal pole. This phenomenon took up to six months under the conditions of the Donzacq fish farm.

On ultra-thin sections performed at the end of this stage, just before the vesicule germinative break down, the microvillosities were retracted. The structure of the zona radiata was more dense (Figure 5 d). Follicular cells deposited on their outer surface, above the zona radiata externa, a new electron-clear spongy layer constituting the "jelly-coat". This layer partially entered the empty pores of the zona radiata externa. The thickness of the jelly-coat varied between 0.5 µm to 0.9 µm (Figure 6b).

DISCUSSION

Oogenesis in *Acipenser baeri* resembles oogenesis in numerous fresh water teleosts (Nagahama, 1983). Nevertheless some synthetic and structural particularities were worth noting.

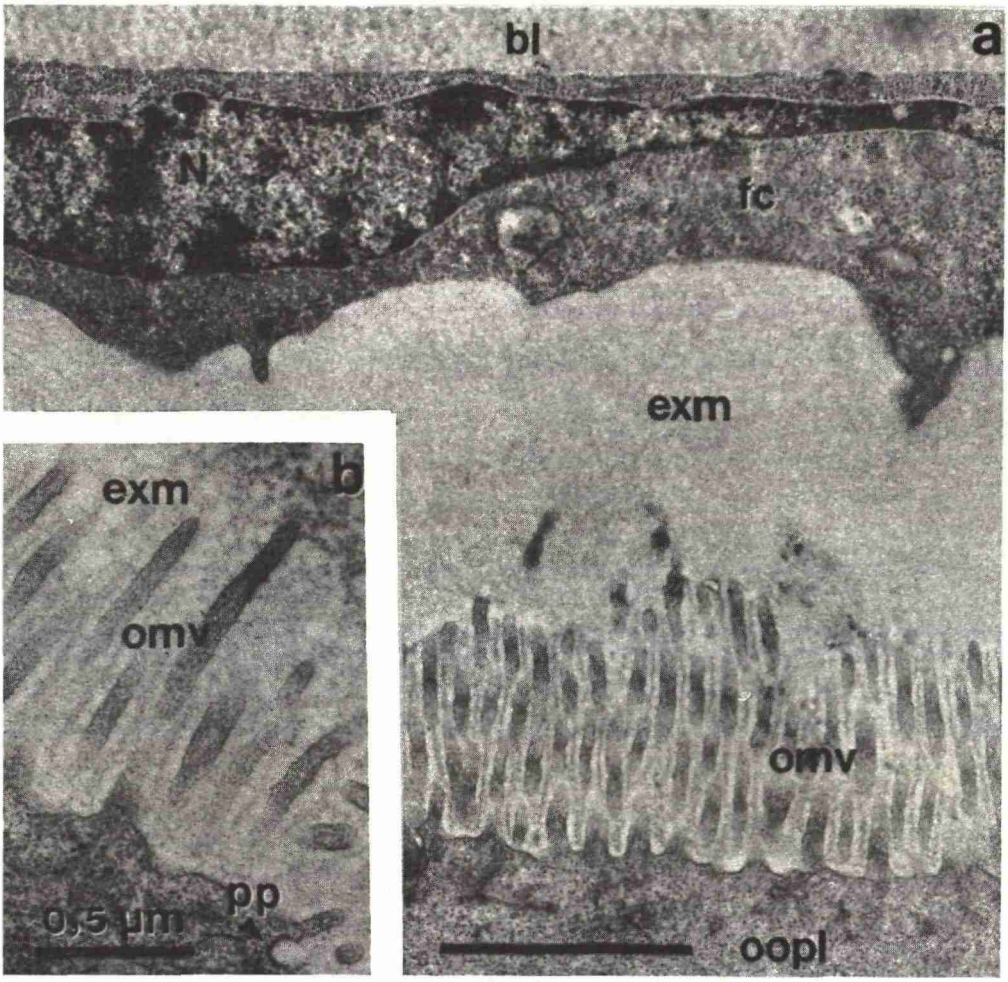


Figure 4- Early stage 3 follicle (electron micrographies). a- follicular cells and oocyte relationship; b -endocytosis at the basis of oocyte microvillosities. (bl: basement lamina, exm: extraoocyte matrix, fc: follicular cell, N: nucleus, omv: oocyte microvillosities, oopl: ooplasm, pp: pinocytotic pit).

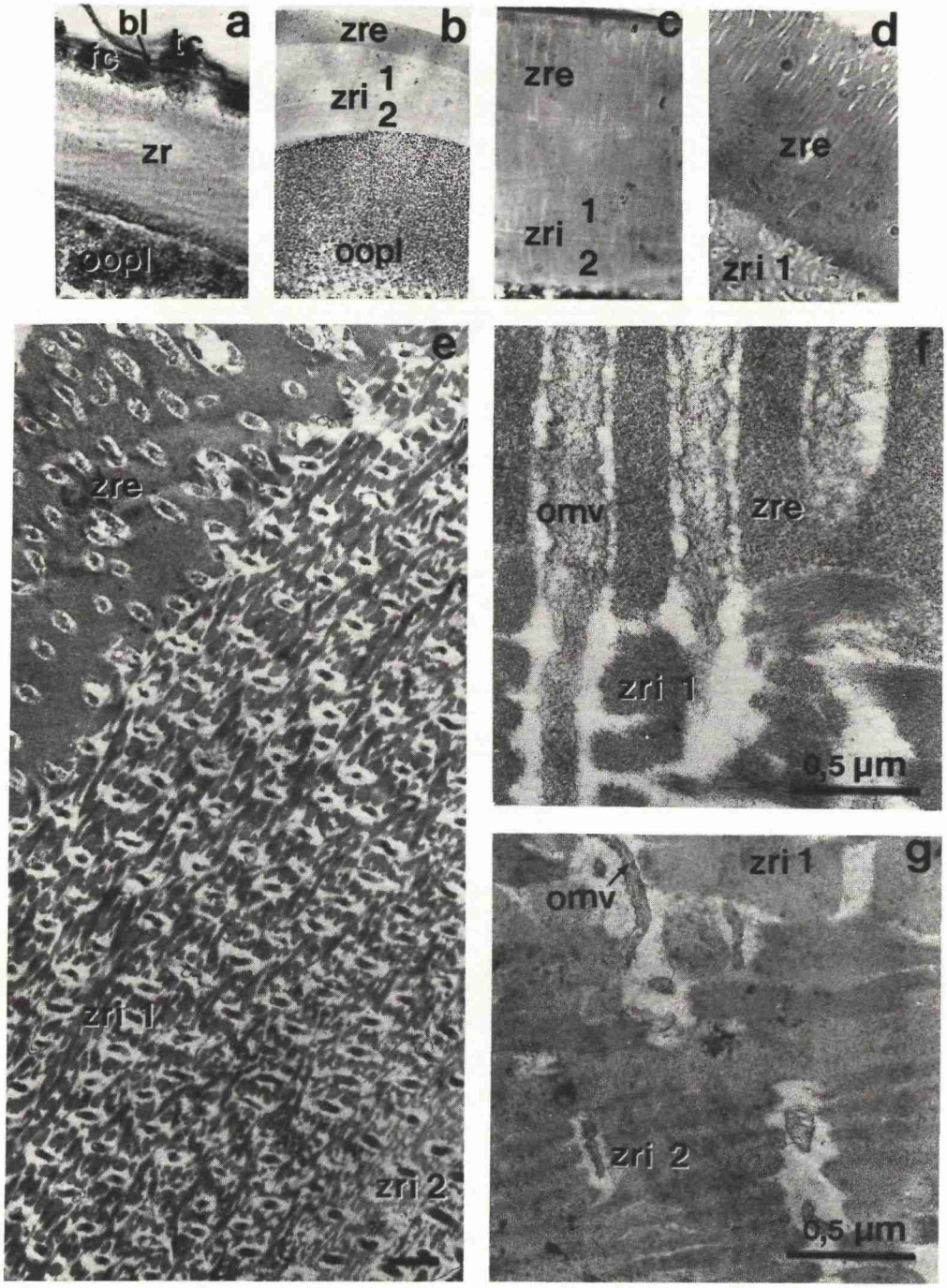


Figure 5- Stage 4 follicle. a,b,c,d- photonic micrographies of zona radiata development; e,f,g- zona radiata electron micrographs. (bl: basement lamina, fc: follicular cell, omv: oocyte microvillosities, oopl: ooplasm, zr: zona radiata, zre: zona radiata externa, zri: zona radiata interna).

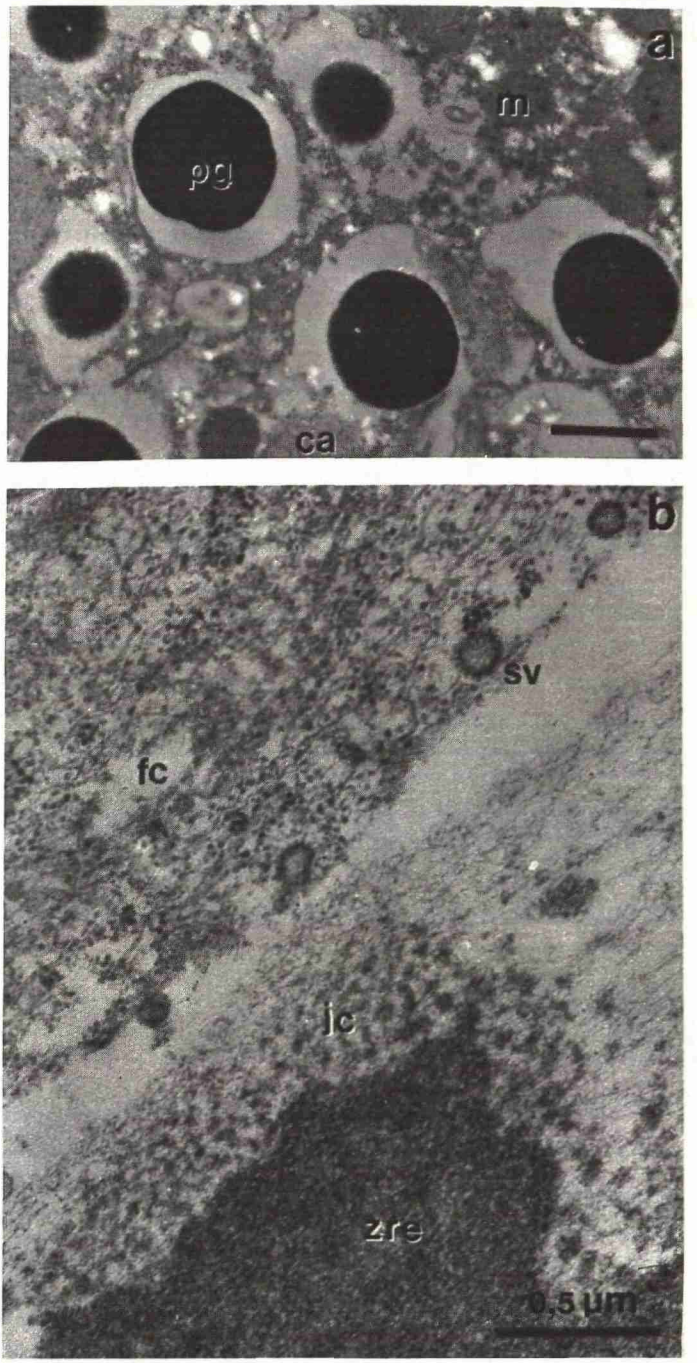


Figure 6- Electron micrographies of: a- pigment granules in the peripheral ooplasm; b- synthesis of the jelly-coat by follicular cells. (ca: cortical alveolus, fc: follicular cell, jc: jelly-coat, m: mitochondria, pg: pigment granule, sv: synthetic vesicle, zre: zona radiata externa).

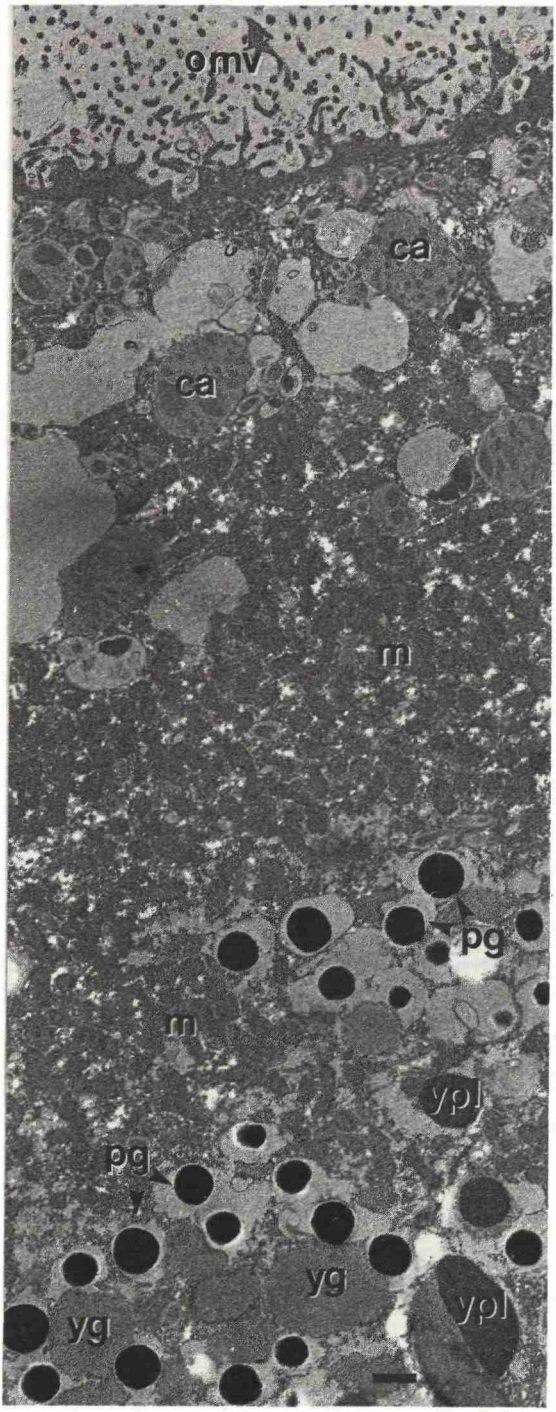


Figure 7- Peripheral cytoplasm after ovulation. (ca: cortical alveolus, m: mitichondria, omv: ovule microvillosities, pg: pigment granule, yg: yolk granule, ypl: yolk platelet).

Acipenser baeri synthetic and endocytotic features

Type 1 and type 2 vitellogenesis

Type 1 vitellogenesis corresponds to the formation in the oocyte of cortical alveoli and lipid globules. It is usually called endogenous vitellogenesis. Type 2 vitellogenesis coresponds to the concomitant incorporation of vitellogenin, a hepatically-secreted phosphoglycoprotein which is the plasma yolk precursor (review of De Vlaming et al., 1980; Wallace et al., 1983). In type 2 vitellogenesis endocytotic events greatly predominate compared with synthetic events. Previtellogenesis corresponds to the type 1 vitellogenesis (Jalabert, Le Bail and Le Menn, unpublished data).

In *Acipenser baeri* lipid globules are identified by histochemical staining from the oogonial stage onwards (Pelissero et al., 1985). This has not been observed in other species (Wallace and Selman, 1981), even in fresh water species where lipid synthesis occurs very long time before type 2 vitellogenesis (Droller and Roth, 1966, for *Anguilla anguilla* ; Ulrich, 1969, for *Brachydanio rerio* or Busson-Mabillot, 1969, for *Lebistes reticulatus*). We must recall however that this species is reared in France under specific temperature and feeding conditions, and no firm conclusion can be reached until gametogenesis in wild population living in natural conditions in the USSR has been studied.

Cortical alveoli synthesis

In the oocyte at the end of Stage 2, at the beginning of oocyte microvillosities protrusion, we observed a very considerable Golgian synthetic activity in the medium and peripheral cytoplasm with numerous Golgi complexes. In the vicinity of each Golgi complex, several spherical agregates of Golgian vesicles led to the formation of small vesicle by deposition of the Golgian secretions in central smooth endoplasmic reticulum vesicles. These vesicles were filled with granular material resembling material contained by cortical alveoli in the next stage. We supose that these features are the first visualisations of synthesis of cortical alveoli. Further investigations are necessary to confirm this hypothesis, for exemple by identifying the chemical nature of their content by chemical reactions on ultra-thin sections.

Specific structural features

Basement lamina

This extra-cellular matrix was located between the follicular epithelium and the thecal cells. The basement lamina is generally secreted by epithelium (Hay, 1981). In this case, a follicular source seemed to be indicated by observations of exocytotic features on the plasmic follicular membrane related to the matrix. Its secretion occured until the follicle reached Stage 3 where the thickness could be 10 µm. During the rapid growth phase the streching of the follicle caused the thickness to decrease to 10 nm making it invisible in light microscopy.

In *Huso huso, Acipenser gueldenstaedti, Acipenser ruthenus* and *Acipenser stellatus* (Caloianu-Yordachel, 1971; Kornienko, 1975) this matrix has been identified, as in *Acipenser baeri*, as polysaccharidic. In this species electron micrographies showed a simple organisation of small circular electron-clear conglomerates in an amorphous matrix, unlike the usual three-layered structure of a true basement lamina (Vrackq, 1974) with a lamina lucida between two lamina densa. The literature would lead us to expect a biochemical composition of glycosaminoglycans, elastin and collagen and the function of a semi-permeable filter (Hay, 1981). This function will be interesting to investigate as regards the pathway of plasma vitellogenin from thecal capillaries to the oocyte (Selman and Wallace, 1982; Abraham et al., 1984).

Extra-oocyte matrix

This matrix deposited between the oolemma and the follicular epithelium is particularly emphazized in *Acipenser baeri.*

Few authors mention its presence in other fish : its electron-clear fibrillar material mentioned without specification by Caporiccio and Connes (1977) has been interpreted as a cell-coat by Busson-Mabillot (1973), as the first deposited zone of the primary envelope (Anderson, 1967; Wourms, 1976), as a follicular matrix by Abraham et al. (1984) and as an extra-oocyte matrix by Le Menn (1984) and Nunez Rodriguez (1985). In all the fish that we have studied, ultrasructural features gave no indication about its cellular origin.

This matrix was deposited as soon as the follicular epithelium separated from oolemma at the end of Stage 1 follicle. It is in this environment that the zona radiata is constituted. The chemical composition of this matrix is not defined but it probably contains proteoglycans (Hascall and Hascall, 1981) well known for their ability to capture and release free ions. This structure could influence the pH in the vinicity of the oocyte and act on the incorporation of exogenous vitellogenin (VTG). Indeed this molecule reaches the oolemma by passing through this matrix along the oocyte microvillosities. It has been demonstrated in birds that the binding of the VTG to its receptors is pH dependent with the best binding for acid pH values (Woods and Roth, 1980; Yusko et al., 1981). Then such a structure may interfere in the process of internalisation by providing an adapted microenvironment.

Zona radiata

This glycoproteic matrix is built up between the oocyte and the follicular epithelium apparently synthetized by the oocyte. The first part deposited is then in an outer position compared with the later parts deposited.

At the end of vitellogenesis, at Stage 4, just before processes of maturation, the zona radiata is formed by three layers : in order of deposition we observed the zona radiata externa, the zona radiata interna 1 and the zona radita interna 2. During the formation of this matrix we observed numerous dense cored Golgian vesicles associated with Golgi complexes located in the peripheral ooplasm just under the oolemma. These features seem to confirm as in other fish species the oocyte origin of this matrix (Tesoriero, 1977; Le Menn, 1984; Nunez Rodriguez, 1985). These three layers have been observed in other sturgeon species (Magnin, 1967; Ginzburg, 1968; Caloianu-Yordachel, 1971; Markov, 1975; Cherr and Clark, 1982), but the interpretations of Soviet authors are not in agreement with ours. They consider the zona radiata externa as the outer layer which allows the eggs to stick on the substrate after laying, and they call it the "jelly-coat". In fact at the end of vitellogenesis during Stage 5 this new spongy layer is deposited on the zona radiata externa (Pelissero et al., 1985) causing the fixation of the egg when it is layed in water. This layer is described by Cherr and Clark (1982, 1984) in *Acipenser transmontanus*. Such specific secretions involved for the fixation of eggs have been described in teleosts : *Cichlasoma nigrofaciata* (Busson-Mabillot, 1977), *Cynolebias melanotoenia* et *Cynolebias ladigesi* (Wourms and Sheldon, 1976), *Pomatochistus minutus* (Riehl, 1978), *Gobius niger* (Le Menn, 1984). In all these species, as in *Acipenser baeri*, ultrastructural features indicated that the sticking material were secreted by the follicular cells.

Until the end of vitellogenesis, only one microvillosity is observed in each channel of the zona radiata. Follicular cells never put out microvillosities towards the oocyte. From the beginning of Stage 3 onwards, the follicular epithelium is completely separated from the surface of the oocyte as soon as the junctions with oolemma disapeared. We have never observed on this species junctions between apical part of oocyte microvillosities and apical plasmic membrane of the follicular epithelium as shown in *Plecoglossus altivelis* by Toshimori and Yasuzuni (1979).

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HISTOLOGICAL STUDY OF THE OVARIAN DEVELOPMENT IN WILD WHITE STURGEON, *Acipenser transmontanus*

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RESUME

Des sections d'ovaire de 20 esturgeons blancs capturés dans la baie de San Francisco ont été colorées par l'acide périodique Schiff et imprégnation argentique. Les diamètres des ovocytes et la densité des cellules de la granulosa ont été mesurés par analyse digitale d'image. Les concentrations du plasma en protéine, phosphore protéique alcalin labile et en oestrogène ont été déterminées sur le même poisson. Les données révèlent 5 groupes avec différents stades ovariens.

Le groupe 1 montre des conditions réfractaires, avec une valeur modale du diamètre des ovocytes de 0,2 mm, une granulosa indifférentiée et pas de chorion. Le groupe 2 a des ovocytes plus développés (0,4 - 0,6 mm) avec un mince chorion et une prolifération des cellules de la granulosa. Les groupes 3, 4 et 5 ont des follicules vitellogéniques dont les valeurs modales du diamètre s'échelonnent de 0,8 à 3,2 mm. La plus grande différence de diamètre des ovocytes est entre le groupe 4 (vitellogenèse précoce) et le groupe 5 (ovaires mûrs). Les taux de métabolites plasmatiques et d'oestrogènes sont élevés dans les groupes 3 et 4 comparativement à ceux des 3 autres groupes. Ces données nous permettent de tirer la conclusion préliminaire suivante, la vitellogénèse de l'esturgeon blanc sauvage est bisannuelle.

Mots clés : *Acipenser transmontanus*, ovogenèse, microscopie photonique, plasma.

ABSTRACT

The ovarian sections of twenty white sturgeon caught in San Francisco Bay were stained with periodic Acid-Schiff and Retic (silver impregnation) stains. Oocyte diameters and granulosa cell densities were measured by digital image analysis. Plasma concentrations of protein, alkali-labile protein phosphorus and estrogen were assayed in the same fish. Data revealed 5 groups with different ovarian stages. Group 1 exhibited refractory conditions, with a modal oocyte diameter of 0.2 mm, an undifferentiated granulosa layer and no chorion. Group 2 had more advanced oocytes (0.4 - 0.6 mm) with a thin chorion and proliferating granulosa cells. Groups 3, 4 and 5 had vitellogenic follicles with modal diameters ranging from 0.8 mm to 3.2 mm. The largest difference in oocyte diameter was between groups 4 (early vitellogenic) and 5 (ripe ovaries). Plasma metabolites and estrogen were elevated in groups 3 and 4, compared to groups 1, 2 and 5. This data allows us to draw preliminary conclusions that vitellogenesis in wild sturgeon is biennal and regulated by seasonal zeitgebers.

Key-words : *Acipenser transmontanus*, oogenesis, light microscopy, plasma.

INTRODUCTION

Sturgeons differ from most teleosts by their late puberty and apparent longer-than-annual ovarian cycles. Understanding the ovarian cycle of sturgeon is important for the regulation of fisheries and the management of cultured stocks. The adult white sturgeon of San Francisco Bay, sampled during the winter season, exhibited three distinct ovarian stages, previtellogenic, early vitellogenic and ripe ovaries ; the latter contributing only 16 % of the stock (Chapman et al., 1987). Our objective was to characterize these stages using histochemical and histometric techniques.

MATERIAL AND METHODS

We sampled (ovarian biopsy and blood plasma) 398 adult females (fork length 127-184 cm) in San Francisco Bay during the winter season (January and February). Paraffin sections of ovaries (fixed in buffered formalin) were stained with hematoxylin and eosin. Plasma was assayed for total protein (Biorad), alkalil-labile protein phosphorus (ALPP) and estrogen (Moberg et al. ; Kroll and Doroshov, this volume).

Hematoxylin and eosin slides from each fish were scored for structural differentiation of the ovarian follicle (Chapman et al., 1987). Five distinct groups were present : (1) refractory stage : oocytes with strongly basophillic cytoplasm and no differentiated chorion ; (2) previtellogenic : oocytes with slightly basophillic cytoplasm and a thin layer of chorion ; (3) vitellogenic I : oocytes with eosinophillic cytoplasm, small yolk platelets, and a two-layered chorion ; (4) vitellogenic II : oocytes with a three-layered chorion but without pigment ; (5) ripe : pigmented and polarized eggs. These five groups characterize the ovarian conditions in winter-sampled stock, and our description is not intended to classify sturgeon oogenesis.

Four blocks were randomly choosen for each group, and new paraffin sections (4 microns) were stained by periodic acid-Schiff (PAS) with an alcian blue background, and by the silver impregnation stain (Retic), using modifications of the staining procedures described in Sheehan and Hrapchak (1980). The former stain differentiates the carbohydrate-containing materials (basal lamina, chorion, platelets and glycogen granules) by a bright magenta color ; the latter stains granulosa cells to a dark color facilitating their counting. Slides were examined with high resolution light microscopy. The diameters of 50-70 oocytes, randomly selected in the optical field, were measured using a digital image analyzer (Microplan II, Nikon). Granulosa cells in the cross-sectional follicular perimeter were counted in the same fashion. Descriptive statistics for normal distribution (mean ± sem), one way randomized anova and a Student-Neuman-Keul test ($p \leq 0.05$) were applied for data processing.

RESULTS

The most advanced ovarian follicles characteristic of each group are shown in figure 1 (A-F). In group 1 a single PAS-positive extracellular matrix, basal lamina, was present and only very few undifferentiated granulosa could be seen (Figure 1a). The ooplasm was homogenous and contained only one type of inclusions, large non-staining cortical vesicles reminiscent of the primordial cortical alveoli in other teleosts.

A thin, PAS-positive single-layered chorion was apparent in group 2 (Figure 1b), along with more numerous squamous granulosa cells (Figure 1c). No structured yolk platelets could be detected ; however, the ooplasm contained numerous small vesicles and an apparent rough endoplasmic reticulum.

The follicles in group 3 possessed a thicker, two-layered chorion, numerous granulosa cells that were more cuboidal in configuration, and small yolk platelets, most abundant in the cortical layer of the egg (Figure 1d).

In group 4, the third, outer layer of the chorion was differentiated ; granulosa cells were cuboidal ; and the ooplasm was filled with medium-size platelet yolk (Figure 1e).

In group 5 the chorion dramatically increased in thickness (all three layers were PAS-positive) ; the granulosa cells became squamous ; and the ooplasm contained yolk platelets, melanin and glycogen granules in the cortical layer (the glycogen was differentiated by using control slides with diastase digestion) (Figure 1f).

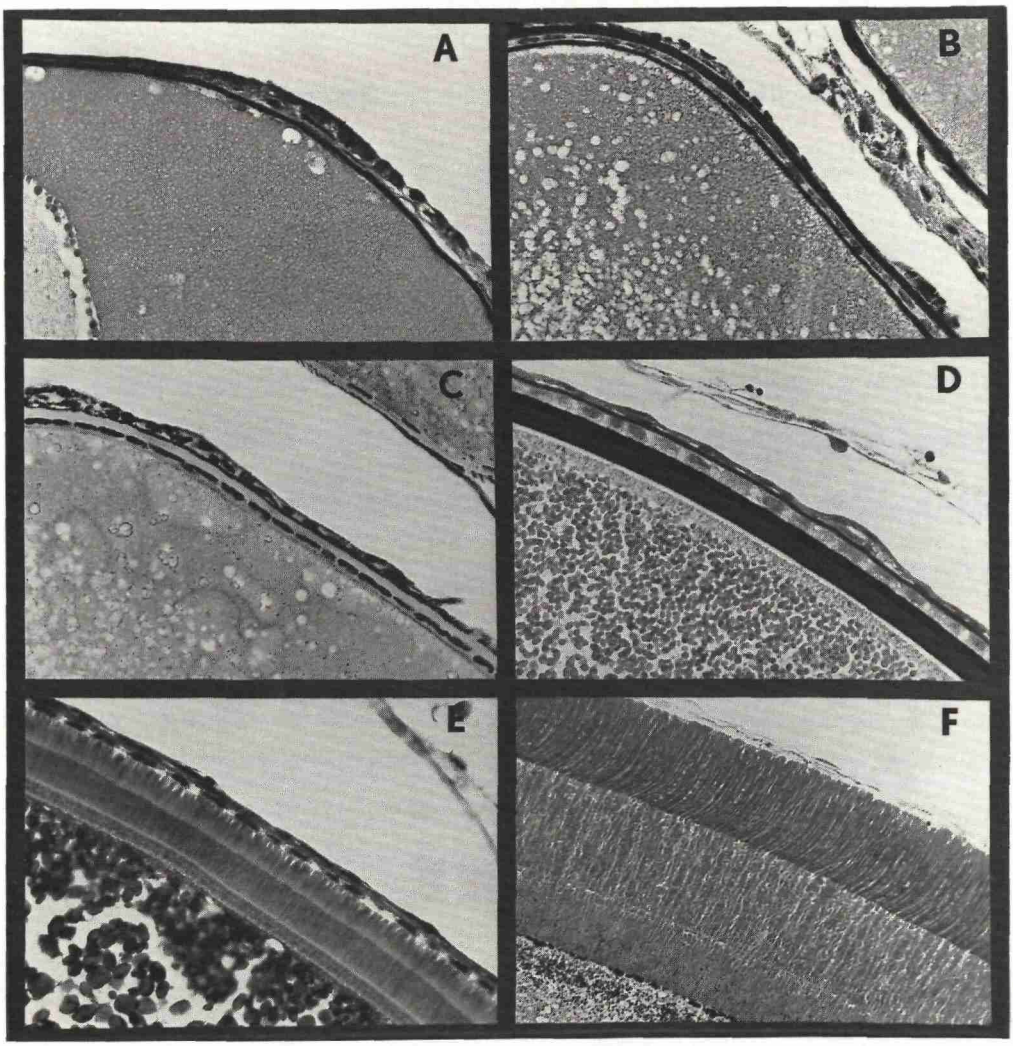


Figure 1 - A) Group 1 fish with basal lamina and undifferentiated granulosa cells (PAS). B) Group 2 fish with a single layer chorion (PAS) and C) proliferating granulosa cells (Retic). D) Group 3 follicles with a two-layered chorion and yolk platelets (PAS). E) Group 4 follicles have a three-layer chorion, cuboidal granulosa cells and medium size yolk platelets (Retic). F) Group 5 fish with a thickened three-layer chorion (PAS).
Original magnification for all photographs is the same, 157.5x.

The oocyte diameter histograms suggest unimodal distributions for groups 1 and 2, and bimodal distributions for groups 3, 4 and 5 due to the apparent recruitment of a single clutch into vitellogenic growth (Figure 2).

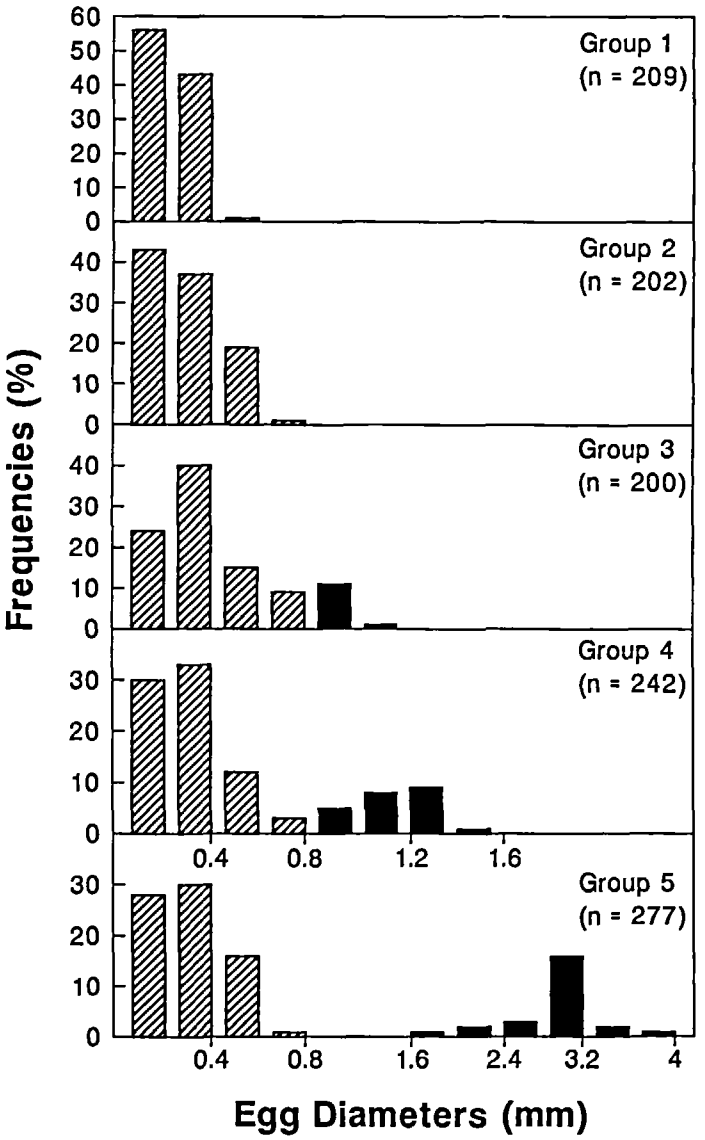


Figure 2 - Histograms of ovarian follicle size distributions in five groups of sturgeon with different ovarian stages. Shaded areas indicate oocytes with yolk platelets. Data are pooled observations for four females in each group.

The positions of a second peak in the histograms differed significantly between groups 4 and 5, with an absence of oocytes with diameters of 0.8 - 1.6 mm in the ovaries

of group 5 fish. Histograms discriminate the fish that recently initiated vitellogenesis (groups 3 and 4) and those approaching spawning (group 5).

The relationship between granulosa cell densities and oocyte size is shown as a scatter plot (Figure 3). A major increase in granulosa cell density was observed in oocytes with a diameter of 0.2 - 0.6 mm corresponding to groups 2 and 3. Cell number per 100 microns of egg perimeter increased significantly from 3.7 ± 0.1 (diameter 0.2 mm, n=37) to 8.9 ± 0.1 (diameter 0.6 - 1.4 mm, n=55), with the total number of cells in the cross-sectional perimeters increasing from 16 ± 1 to 253 ± 7. Proliferation of granulosa cells was concomittant with the early differentiation of the chorion and the appearance of yolk platelets in the cortical ooplasm. It was also apparent that granulosa cell density did not increase from group 3 to group 5 stage fish.

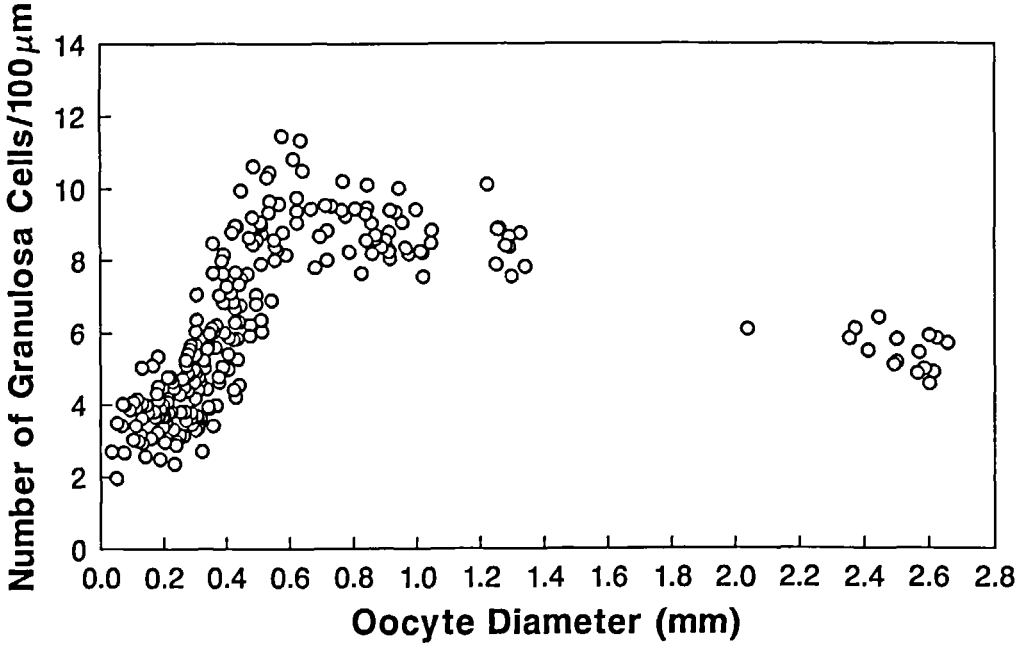


Figure 3 - Scatter plot demonstrating changes in density of granulosa cells per 100 microns of the inner follicular layer. Data are pooled observations for all sampled fish. Measurements were carried out for different size of eggs in each group of fish.

Concentrations of plasma metabolites are shown in figure 4. Total plasma protein was significantly lower in groups 1, 2 and 5 (23.2 ± 0.9 mg/ml, pooled), compared to groups 3 and 4 (30.7 ± 1.3 mg/ml, pooled). Group 4 had a significantly higher ALPP concentration (54.8 ± 8.3 ug/ml), compared to all others. Although mean ALPP for groups 1 and 2 were very low and mean ALPP in groups 3 and 5 were clearly elevated, none of these stages differed significantly due to high individual variations in groups 3 and 5. Estrogen concentrations were below assay detection in groups 1 and 2, insignificantly elevated in group 3 (0.57 ± 0.23 ng/ml), and significantly higher in group 4 (5.36 ± 0.36 ng/ml). Only one fish in group 5 was assayed for estrogen (0.69 ng/ml).

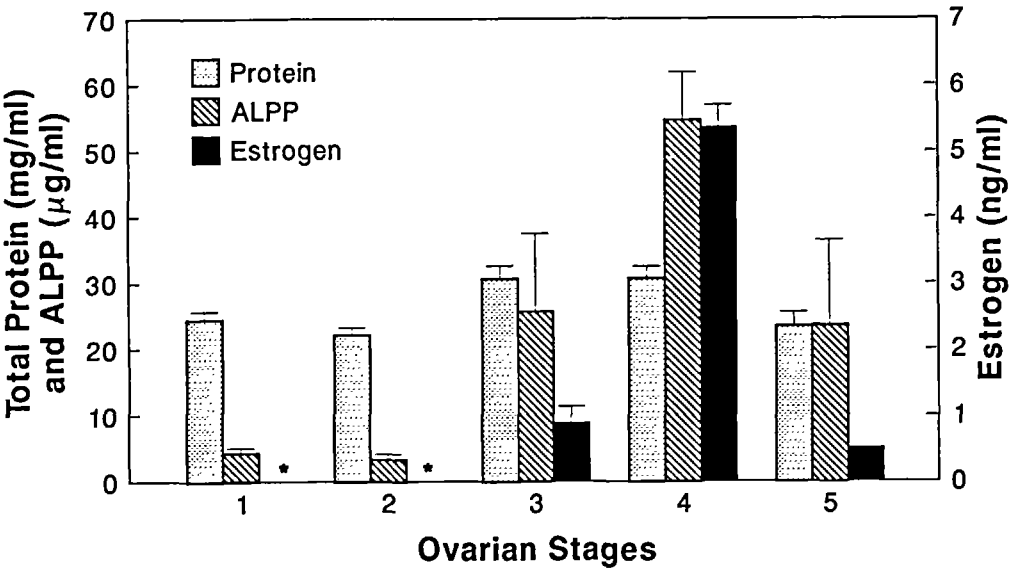


Figure 4 - Plasma concentrations of protein, alkali-labile protein phosphorus and estradiol in five different ovarian stages. Data are means and standard errors (n=4). Asteriks inidcate estrogen values below the detection limit (200 pg/ml).

DISCUSSION

Differences in ovarian follicle morphology between groups 1, 2 and 3 indicate that initiation of vitellogenesis in white sturgeon occurs when the oocytes grow to 0.6 - 0.8 mm in diameter.

Evidently, the mitotic differentiation of the granulosa layer and secretion of the egg chorion preceed or accompany the early phase of vitellogenesis. Plasma profiles of protein, ALPP, and estrogen are similar to other fish during the physiological regulation of vitellogenesis (Mommsen and Walsh, 1988). Direct evidence of estrogen-induced vitellogenesis was obtained by Moberg et al. (this volume) and substantiated by the isolation of vitellogenin mRNA from the liver of estrogenized white sturgeon (Bidwell, 1989). Rather surprising was an apparent lack of vitellogenin and estrogen in group 2 females that exhibited follicular differentiation, since Le Menn and Pelissero (this volume) found vitellogenin uptake by the oocyte in approximately similar ovarian stage of captive Siberian sturgeon. The possibility exists that we could not detect vitellogenin secretion due to the low resolution of our assays.

Spawning of white sturgeon in the rivers of San Francisco Bay estuary occurs in March-April (Kohlhorst, 1976). Group 5 adult females in the Bay, during January and February, were in prespawning conditions (Lutes et al., 1987). The presence of fish in early vitellogenic conditions (groups 2, 3 and 4) and the absence of those with mid-vitellogenic ovaries (females with oocytes 0.8 - 1.6 mm diameter) indicates that sturgeon vitellogenesis is approximately biennal and controlled by seasonal zeitgebers. The onset of vitellogenesis may occur in the fall or winter, with mid-vitellogenic stages occuring in the spring and summer. Vitellogenesis would then be completed in the fall, winter and spring of the next calendar year. A predominantly biennal ovarian cycle in domestic sturgeon stocks was reported by Burtzev (1969), Williot et al. (this volume) and were

recently observed in the first maturing domestic white sturgeon females (Joel Van Eenennaam, personnal observations). Although the vitellogenic cycle of wild white sturgeon may be biennal, the actual interval between the first and second spawning may be longer due to the unknown duration of the refractory ovarian stage (group 1). This may explain the low proportion of ripe females in white sturgeon stock (Chapman et al., 1987) and the extremely slow recovery of sturgeon from overfishing (Semakula and Larkin, 1968).

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PECULIARITIES OF INTRAPOPULATIONAL DIFFERENTIATION OF STURGEON (*Acipenser gueldenstaedti* Brandt) UNDER PRESENT-DAY CONDITIONS

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RESUME

Cette étude se rapporte à l'analyse des différences entre les populations de l'esturgeon de la Volga, *Acipenser gueldenstaedti*. On démontre l'existence de 2 formes de migrants, ceux de printemps et ceux d'hiver. Les animaux appartenant à chacune de ces formes peuvent être distingués par l'évolution de leur gamétogenèse et l'état de leurs réserves énergétiques. Les particularités des mécanismes hormonaux de régulation de reproduction sont établies. Les conditions naturelles de migration et de reproduction ayant été bouleversées, on analyse le comportement de ces populations en réaction à ces changements ainsi que les interventions possibles.

Mots clés : *Acipenser gueldenstaedti*, population, migration, régulation hormonale, différenciation

ABSTRACT

Data are given on the intrapopulational differentiation of sturgeon (*Acipenser gueldenstaedti* Brandt) on the Volga river. It has been shown that the population contains winter- and spring- annual forms which migrate into the river with different state of gonads and being provided with energetic resources in various degree. The parculiarities of hormonal regulation of gonad functions have been made clear. Under discussion are the possibilities of formation of varied intrapopulational differentiation in sturgeons. Consideration is given to the manifestations of the population responding to the changed conditions of migration and reproduction. Possible ways are discussed of a directional formation of the population at the present period.

Key-words : *Acipenser gueldenstaedti*, population, differentiation, migration, hormonal regulation

INTRODUCTION

Differentiation in the limits of population is strongly pronounced in many species of *Acipenseridae*. Its development contributes to most complete utilisation of all possibilities offered by the range, reflecting the adaptation of the population as a whole to its conditions. The majority of sturgeons of commercial importance are migratory raising upstream for spawning.

We observed different forms (biological groups) in the populations of sturgeons ; winter and spring annual forms. Spring forms migrate from sea to river just before spawning. Winter forms stay in rivers until next spring where the reproduction occurs (Barannikova, 1957). Different parts of spawning regions are used for reproduction by sturgeons of different biological groups, the spawning temperature range being fairly stable for each species of *Acipenseridae*. Depending from the peculiarities of the rivers, different correlation of winter and spring forms is developped in different populations of sturgeons (Gerbilsky, 1967; Barannikova, 1972, 1975).

Now the conditions of migration and reproduction for sturgeons have considerably changed, as a result of various antropogenic impact (including hydroengineering construction). This paper deals with the analysis of new peculiarities of intrapopulational differentiation in sturgeons linked with hormonal regulation induced by these changes.

MATERIAL AND METHODS

The intrapopulational differentiation of Russian sturgeon (*Acipenser gueldenstaedti* Brandt) has been studied on Volga river in 1981-1988, at different seasons from March till October. The material was collected in the mouth of the river and also in the dam area near Volgograd. The state of 1878 fishes was determined.

Histological analysis of gonads and pituitary (484 fishes) has been carried out after Bouin fluid fixation and staining by different methods, mostly by Azan method.

For ultrastructural analysis of gonadotropic cells (GTC), pituitaries were fixed in 6,5% glutaraldehyde solution and post-fixed in 1% osmium tetroxide. They were embedded in Epon and stained with uranyl acetate and lead citrate. The sections were examined in JEM-7 electron microscope (Barannikova et al., 1984 ; Efimova, 1989).

The content of various hormones in the blood serum was determined by radioimmunoassey (RIA). For determining the gonadotropic hormone (GTH) of the pituitary the system for RIA on the basis of purified sturgeon GTH was used. This preparation was received from Dr G. Zenkevich (Institute of Biology of Latvijas Academy of Sciences). Labelling GTH by I^{125} was compleated using the chloramine method (Greenwood et al., 1963). Detailed description of the RIA procedure was published previously (Barannikova et al., 1984).

Concentrations of sex steroids in the blood (progesterone, P, testosterone, T) and cortisol were determined using sets for RIA (accordingly RIN-T-^{3}H, steron-T-^{3}H, steron-K-^{125}/-M).

The mean ± standard error of the mean for each group of fishes was determined.

RESULTS

Main features of differentiation in the population of the Russsian sturgeon. Characteristics of spring and winter forms migrants

The anadromous migration of winter form of sturgeon from sea to Volga begins at present in april-may, the gonads of theses fishes being far from maturity. The gonosomatic index is low (table 1). Accumulation of yolk is going on in the oocytes, the process of vitellogenesis has began during sea period. The oocytes did not nearly reach their definitive size. Spermatogonia are predominating in the testis, spermatocytes are also present.

At the beginning of anadromous migration of winter form of sturgeon the GTC of pituitary are of small size, the gonadotropic activity of the pituitary being not high (Barannikova, 1975). At this period a periodic release of GTH into blood takes place (sexual differences are absent), which provides the secretion of sexual steroids and further development of gonads. The levels of GTH, P and T are not high (table 1).

In gonads, liver and other tissues great amounts of energetic resources (fats and glycogen) are present (Barannikova, 1975). Then anadromous migration and the development of gonads till maturity are possible for these sturgeons in the absence of exogenic nutrition in the river period of life, lasting 10-12 months till spawning.

The anadromous migration of winter form of sturgeon into Volga continues during the whole summer and in autumn, the gonads gradually developing. In sturgeons migrating in autumn the weight of oocytes is much higher, than in the summer migrating sturgeons, the vitellogenesis is compleated, spermatozoa are predominating in the testis (table 1). Most of GTC of pituitary are at the first steps of their secretory cycle, dark and light cells predominating. Dark cells contain many secretory granules and some large globules, light cells contain less small granules, the process of secretion release is in beginning (Efimova, 1989). The gonadotropic activity of the pituitary is high, but, as seen from the functional state of GTC, GTH is slightly released from the gland (Barannikova et al., 1984). The level of GTH in the blood does not reliably differ from its indices in the winter form of sturgeon, migrating in spring (table 1). Such dynamics of GTH was also observed in the sturgeon earlier (Bukovskaya, 1981a).

The spring form of sturgeon migrates into Volga in early spring, the oogenesis and spermatogenesis being compleated at this period, the values of gonosomatic index are high (table 1). The GTC of pituitary are in the state of active secretion, the number of dark cells being low. Light and exhausted cells are predominating in the pituitary (Barannikova et al., 1984). There is a strong release of GTH into the blood, the levels of sexual steroids also being increased (table 1). The levels of these hormones are much higher at the beginning of the river period of anadromous migration of spring form of sturgeon in comparison with the winter form (at the same period in the same part of the river). The reproduction of the spring form of sturgeon takes place shortly after coming into river on spawning grounds in its lower part.

Table 1 - Indices of winter and spring forms of sturgeon at the beginning of the river period of anadromous migration (mouth of Volga) M±m

Index	**1** *Winter form: migration in spring, spawning next spring*	**2** *Winter form: autumn migration, spawning next spring*	**3** *Spring form: migration in spring, just before spawning*
Oocytes, weight, mg	5,8 ± 0,2 (n=352)	17,5 ± 0,3 (n=185)	23,8 ± 0,4 (n=361)
Gonadosomatic index %, female	8,3 ± 0,4 (n=350)	18,2 ± 0,5 (n=190)	23,8 ± 0,9 (n=385)
GTH ng/ml			
female	1,8 ± 0,3 (n=27)	1,0 ± 0,3 (n=15)	3,4 ± 0,4 (n=17)
male	2,0 ± 0,3 (n=4)	1,6 ± 0,2 (n=15)	4,3 ± 0,6 (n=16)
Progesterone ng/ml			
female	1,3 ± 0,3 (n=6)		5,7 ± 1,3 (n=6)
male	1,6 ± 0,2 (n=8)		9,2 ± 3,7 (n=4)
Testosterone ng/ml			
female	4,1 ± 0,4 (n=6)		14,2 ± 2,5 (n=6)
male	4,3 ± 0,4 (n=4)		13,1 ± 2,9 (n=6)

For GTH 1, 2 with 3 group $p < 0,01$
For P 1 with 3 group $p < 0,05$
For T 1 with 3 females $p < 0,01$, males $p < 0,05$

Consequently, in contrast to winter form of sturgeon, the whole process of development of sexual glands and accumulation of yolk in the oocytes up to the definitive state, in spring form of sturgeon takes place in the sea.

In spite of differences in the stage of maturity and in the gonadotropic axis state in spring and winter forms of sturgeon, some similiarities were found in the hormonal regulation of anadromous migration in these forms. Main features are the activation of the pituitary-interrenal and pituitary-thyroid axis in both spring and winter forms (Barannikova, 1975; Barannikova et al., 1978).

Reproduction of all the spawning portions of the population of Russian sturgeon of both winter and spring forms occurs on the Volga in spring at similar indices of temperatures at the same spawning grounds. The spawning grounds in the upper part of Volga are inacessible now.

On the Volga however, as on a number of other rivers, reproduction of sturgeon in summer has been established at much higher indices of spawning temperatures (Alavdina, 1954; Artyukhin, 1983). It has been shown that summer spawning is carried out by Persian sturgeon *Acipenser gueldenstaedti* persicus Borodin (Artyukhin, 1979).

Thus, due to the spawning range being used by sturgeons capable to reproduce at different temperatures and therefore at different periods of time, a maximal yield is possible from the river spawning grounds.

Manifestations of the process of sturgeon population response to changes of environmental conditions

Consequently to the construction of dam near Volgograd winter form is earliest to reach the area near the dam, duration of anadromous migration being 3-4 times less than previously. The effect of shortening of anadromous migration of winter form of sturgeon manifests itself in the fact, that part of fishes, especially males, do not completely use up the accumulated energetic resources. In the past the spent sturgeons in the lower part of Volga were quite exhausted, fat was not stored in the gonads (Barannikova, 1979). At present we observe in the same part of the river spent sturgeons with gonads containing fat resources.

Negative influence of changed conditions appears to affect the winter form of sturgeons, migrating into Volga in spring and summer, most strongly. Indeed in summer and autumn degenerative changes in the gonads, hyperfunction and exhaustion of the interrenal gland were recorded in sturgeons inhabiting near the dam (Barannikova, 1975; Barannikova et al., 1978).

After wintering, however, the cortisol level in females before spawning is not high (6,4±1,7 ng/ml, n=12) in comparison with its concentration in blood during anadromous migration in spring (40,0±10,2 ng/ml, n=17). In sturgeons in the dam area with approach of spawning the level of GTH increases as compared with autumn period (table 2). The reproduction of these fishes takes place in spring, the GTH level being the highest in the preovulatory period (Bukovskaya, 1981b; Barannikova et al., 1989).

Table 2 - Dynamics of GTH in the blood of female winter-form sturgeon in Volga near the dam in different seasons

Month *state of gonads*	*GTH ng/ml* *M±m*	
October compleated *vitellogenesis before wintering*	1,6 ± 0,2 n = 43	↕ p < 0,01
April after wintering *before spawning*	3,2 ± 0,6 n = 27	

DISCUSSION

Starting with the ideas of adaptive significance of the ecological structure of sturgeon population it is clear that deep changes in the conditions of ecosystems which, in the present day period occur in short periods of time, results in disturbances of the state of adaptation of the population.

The deepest changes concern winter form of sturgeons, which are hindered by the dam near Volgograd. The length of river migration of these fishes is much shorter now, especially for the winter form of sturgeon, which enter the river in spring and summer. This form migrated before dams were constructed more than 2000 km upstream Volga and its tributaries for hibernation and reproduction in the upper part of spawning zone (Barannikova, 1957).

An important influence on the state of the population is undoubtedly exercised by the sharply increased extent of panmixia (free crossing) which is connected with the

fact that reproduction of the whole population of the Russian sturgeon occurs during same period on limited spawning grounds of the lower parts of the river.

The given scheme of differentiation in the limits of population of Russian sturgeon of Volga with winter forms prevailing is typical for the rivers of great extent with spawning grounds located in their different parts (Barannikova, 1957 ; Kazansky, 1962). A similar character of differentiation is observed in other species - *Huso huso* L., *Acipenser baeri* Br. in some rivers (Barannikova, 1975).

On shorter rivers of mountainous type the conditions for wintering being absent, the character of intrapopulational differentiation is different ; only forms without wintering in the river are developed, with clearly pronounced periods of anadromous migration (for example, river Sefid-Rud ; Rostami, 1961 ; Artyukhin, 1983).

At present 80% of natural spawning grounds in the Volga basin cannot be used for reproduction by sturgeons, the conditions of migration and spawning being changed (Barannikova et al., 1983).

In spite of it the differentiation in the limits of sturgeon population on spring and winter forms continue to exist, the winter form being more numerous as before. Apparently, total reorganisation of the ecological structure of population is connected with change of many generations, this process requires a long period of time for sturgeons, maturing in late age. At present we observe only first steps of this process.

It was shown, that the migration of winter form of sturgeon begins now earlier than 30 years ago (Barannikova, 1957). The period of migration of spring form coincides more than before with that of winter form. A part of sturgeons, especially belonging to the winter form migrating into Volga in spring and summer do not give progeny in consequence of gonad function disturbances (Barannikova, 1975). The winter form, migrating into Volga in late summer and autumn is not so heavily affected.

At present-day period monitoring of the state of sturgeon populations under changed ecological conditions is necessary. Significant degree of differentiation in the limits of population facilitates the formation of its most suitable ecostructure. One of the ways of solving this problem is to use an appropriate regime of fisheries.

In connection with this, during the period of migration, catching is discontinued, owing to this it is possible to obtain a maximum of large progeny from spring forms. For retaining the genetic geterogeneity of the population, however, the winter forms should also be periodically protected from catching at different periods of migration with the purpose of preserving natural reproduction of fishes from various parts of the population.

An important way of directed formation of populations is also fish-breeding. At present in the USSR biotechnology of sturgeon breeding of different species and biological groups has been developed ; fish-breeding plants produce annually over 110.10^6 juveniles letting out into natural waters for obtaining fishery reserves in the seas (together with the juveniles obtained as a result of natural reproduction).

Artificial propagation is used mainly for spring form of sturgeon, natural spawning being the second source for progeny of these fishes. The winter form of sturgeon is reproduced mainly by natural spawning.

Preferential breeding of sturgeons, belonging to biological groups most promising at the present period is possible. This makes possible formation of population with a appropriate eco-structure.

CONCLUSION

On the basis of the data obtained a conclusion can be drawn about spring forms of sturgeons reproducing in spring. These fishes are not negatively affected by the conditions of wintering near the dam. This is very promising.

Winter and spring forms of sturgeon, migrating into Volga with different state of gonads continue to exist, the winter form predominating. The hormones levels (GTH and some sex steroids) differ in spring and winter forms at the beginning of anadromous migration.

The dynamics, length and duration of migration for different forms have changed. Disturbances of gonads function and physiological state of some winter forms of sturgeon delayed near the dam were established.

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SIBERIAN STURGEON, *Acipenser baeri*, SPERMATOZOA : EFFECTS OF DILUTION, PH, OSMOTIC PRESSURE, SODIUM AND POTASSIUM IONS ON MOTILITY

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RESUME

L'influence du taux de dilution, du pH, de la pression osmotique et de la concentration des ions mononovalents (K^+ ; Na^+ ; Cl^-) sur la motilité des spermatozoïdes est étudiée chez l'esturgeon sibérien, *Acipenser baeri*. Les valeurs moyennes respectives du pH,de la pression osmotique, des concentrations de K^+ et Na^+ dans le liquide seminal sont de pH 8.1 ± 0.8 , 38 ± 3 mOsm/Kg, 2.5 ± 0.3 and 28 ± 7 mM. Le liquide seminal est en outre caractérisé par une courbe de titration correspondant à une forte capacité tampon dûe à la présence de phosphate.

La meilleure dilution dans l'eau déionisée afin d'observer la motilité est d'environ 1/100. Les spermatozoïdes sont motiles pour des solutions salines à pH physiologique (8.2) et pour des pressions osmotiques inférieures à 50 mOsm/Kg. L'inhibition totale de la motilité est observée en milieu acide ou par des pressions osmotiques élevées (supérieures à 100 mOsm/Kg ; mais dans ce dernier cas, la réinitiation de la motilité est possible après dilution dans l'eau deïonisée).

Le potassium à 0.1 mM inhibe fortement, mais de façon réversible la motilité des spermatozoïdes. Les ions sodium et chlore, dans la gamme des concentrations physiologiques (20 mM) n'ont aucun effet sur l'intensité de la motilité.

En conclusion, dans les conditions physiologiques, la concentration en K^+, et non la pression osmotique, est la cause déterminante de l'immobilité des spermatozoïdes.

Mots clés : sperme, *Acipenser baeri*, ions, pH, pression osmotique

ABSTRACT

The influence of the sperm dilution rate, pH, osmotic pressure and monovalent ion (K^+ ; Na^+ ; Cl^-) concentration on sperm motility was studied on the Siberian sturgeon, *Acipenser baeri*. Average values of pH, osmotic pressure, K^+ and Na^+ concentration in the seminal plasma were respectively pH 8.1 ± 0.8, 38 ± 3 mOsm/Kg, 2.5 ± 0.3 and 28 + 7 mM. Titration curve showed a strong buffer capacity of seminal plasma due mainly to presence of phosphate.

The best dilution rate in deionized water for observation of motility were about 1/100. Motility occurred in saline solutions of pH 8.2 and osmotic pressure lower than 50 mOsm/Kg. Total inhibition occurred in media of acidic pH and high osmotic pressures (100 mOsm/Kg and more ; but in this later case, reinitiation of motility was possible after further dilution in deionized water).

Potassium at 0.1 mM was strongly, but reversibly inhibiting sperm motility. Sodium and chloride ions in the range of seminal plasma conditions (20 mM) had no effect on the intensity of sperm motility.

It was concluded that in physiological contidions, K^+ concentration but not osmolality seemed to be the principal cause of sperm immotility.

Key-words : sperm, *Acipenser baeri*, ions, pH, osmotic pressure

INTRODUCTION

In seminal plasma of fish, spermatozoa are immotile ; initiation of motility occurs only after dilution during fertilization. Different factors are able to inhibit

spermatozoa motility : pH and potassium ion concentration in the trout, (Petit et al., 1973; Baynes et al., 1981; Billard, 1975; Baynes et al.,1981; Morisawa et al., 1983a), and osmotic pressure in the carp, (Morisawa et al., 1983b; Billard, 1987).

In sturgeon, few works on sperm and seminal plasma composition have been published (Kucherova and Gobulev, 1973; Fedorova, 1976). But neither the reason for the quiescence within the testis nor the mechanism of sperm activation following dilution were investigated.

The major goal of our study was to investigate which kind of parameter, in terms of osmotic pressure and/or main monovalent cations composition, acts on sperm motility of the chondrosteen, *Acipenser baeri* in natural conditions or in artificial media.

MATERIALS AND METHODS

Sexually mature male Siberian sturgeon, *Acipenser baeri*, weighing 4 to 7 kg, were collected from the pisciculture of Donzacq (France) from December to April. Spermiation was induced by intraperitoneal injection of carp pituitary extract (2 mg/Kg). Fish were maintained in running fresh water at 17°C for the experiment.

Semen (20 to 100 ml/male) were collected by abdominal massage 36 hours after injection. Care was taken to avoid contamination with feces and blood. Sampling could be done even 3 days after injection (Williot et al., 1982). Semen was stored dry at 4°C until use.

In order to study its physico-chemicals characteristics seminal plasma was separated from semen by centrifugation at 3000g during 10 minutes.

The pH of the seminal plasma were measured with a classical pH-meter (Ankersmit A.161). Osmotic pressure was determined by freezing point depression with a semi-micro osmometer (Knauer). Sodium and potassium concentration were estimated, after appropriate dilution in deionized water, using a flame photometer (Eppendorf) (Quinn et al., 1965; Hwang and Idler, 1969).

Phosphate concentration was assayed by the method of Fiske and Subbarow (1925).

Sperm motility was induced by dilution in different media and examined under the microscope. All the experiments were performed at room temperature. We used a motility scale modified from the method of Sanchez-Rodriguez (1977) and evaluated the total duration of motility, until forward movement stopped.

The effect of dilution on spermatozoa motility was assessed, firstly with deionized water as diluent and, secondly with an artificial insemination diluent (DIA), composed by mixture of 125 mM NaCl, 20 mM Tris-buffer, 30 mM glycine at pH 9. Diluents at different osmolalities and NaCl concentrations were also used.

The routine dilution rate for the subsequent experiments was 10^{-2}. The effect of the pH was studied with 10 mM Tris buffer in 25 mM NaCl solution. To investigate the osmotic and ionic effects on the sperm motility, the media with the appropriate chemicals (sucrose, NaCl, KCl, buffers reagent grade purchased from SIGMA) were made from a base solution successively diluted in deionized water to obtain the desired concentration.

RESULTS

Characteristics of Sturgeon sperm

1 Spermatogram

The sturgeon spermatogram varied from 0.5×10^8 to 5×10^8 spz./ml (spermatocrit 0.5 to 5% , n= 15 samples from 6 fishes).

2 Seminal plasma

Osmotic pressure and pH of sturgeon seminal plasma ranged respectively from 24 to 57 mOsm/Kg water (mean : 38 ± 3 mOsm/Kg ; n=20) and pH. 7.8 to 8.3 (mean = 8.1 ± 0.8 ; n= 20).

The Na^+, K^+ and phosphate concentrations means were respectively 28 ± 7 mM, 2.5 ± 0.3 mM and 9 ± 2mM (n= 20). The sodium/potassium ratio averages about 10.

The seminal plasma showed a strong buffer capacity (pK_a=7.1) when titrated with HCl or NaOH solutions (fig.3). The observed curve was characteristic of the presence of phosphate buffer.

Effects of dilution rate on sperm motility

Non-diluted sperm of Siberian sturgeon were usually motionless after collection. They became active when semen was diluted in deionized water at a ratio of 1/6 (fig.1). The longer duration of motility was the cause of the best activity (fig.1). Furthermore, high dilution rates (in the order of 1/100) gave better results. The increase of the spermatozoa activity then was due to both the increased intensity and duration of motility (fig.1).

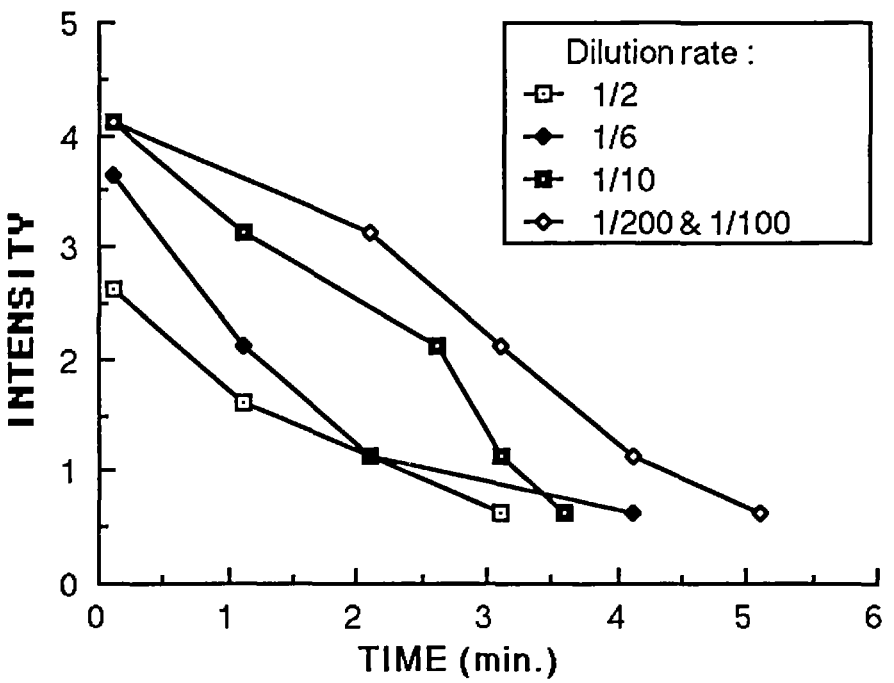


Figure 1 - Effect of dilution rate of sperm with deionized water

Sperm was also diluted in DIA at different osmotic pressures and ionic strength. Sperm remained motionless or slightly motile when semen was diluted in the more concentrated saline solution (125 mM NaCl and 310 mOsm) at a dilution ratio of 1/100. For the other saline concentrations (31.5 and 17.8 mM NaCl), as for deionized water, the best activating effect was observed with high dilution rates (ex : 1/100). Moreover the intensity of motility increased from the dilution ratio of 1/5 (Fig.2).

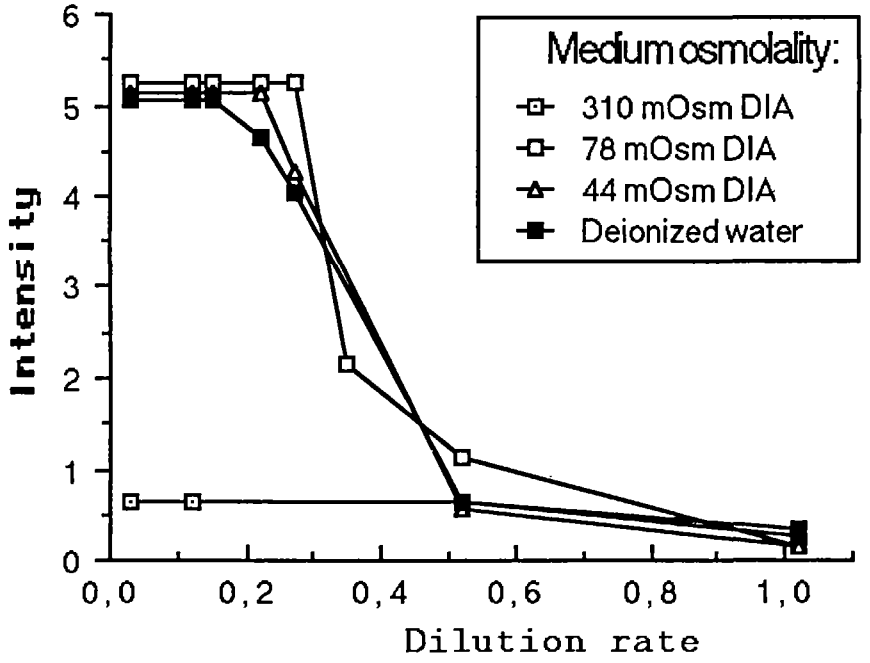


Figure 2 - Effect of dilution of sperm with DIA at various osmolalities

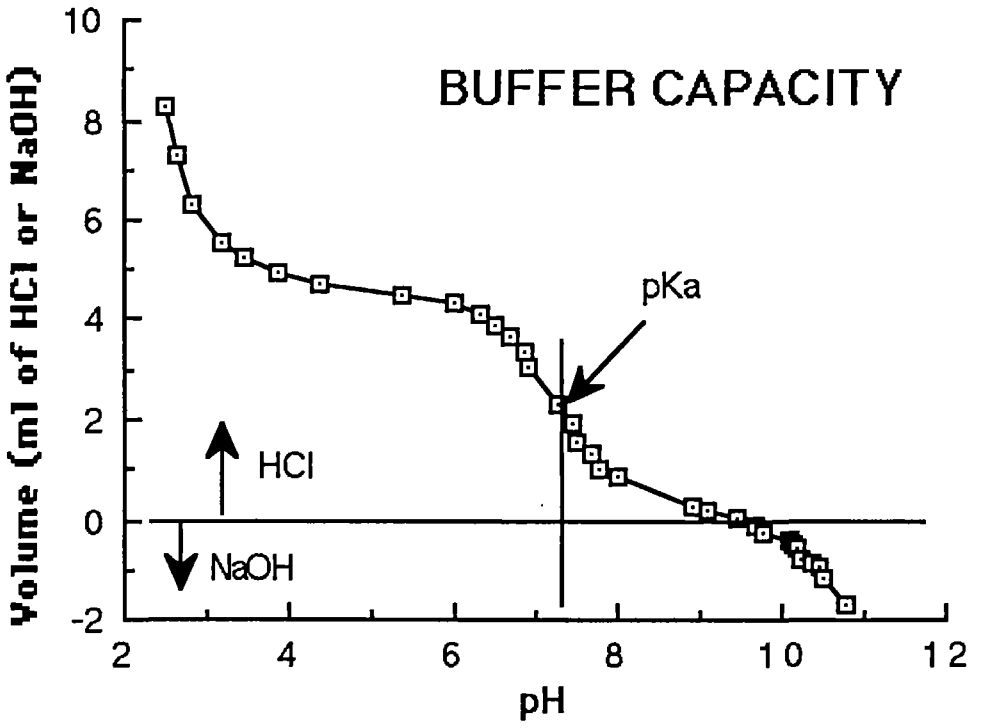


Figure 3 - Titration curve of the seminal plasma

Effect of pH

A buffer solution of 10 mM Tris in 25 mM NaCl solutions was used to test pH effect at a 1/100 dilution ratio. (table 1).

Motility was not affected by the pH until the latter was in the range of the physiological values observed in the seminal plasma(about pH 8.2). Intensity of motility was reduced when the pH was low (pH 6).

Table 1 - Effect of pH on sperm

pH	6,2	7,2	8,2	9,2
Initial intensity of motility	2	4	4+	5

Effect of osmotic pressure

It was tested with sucrose solution at a 1/100 dilution ratio (Fig.4). Sperm motility was good while the osmotic pressure of the diluent remained lower than in the seminal plasma value. Reduction in motility duration (and also intensity not shown) appeared at 55 mOsm/Kg water. Total inhibition occurred at 100 mOsm/Kg water and up to a 300 MOsm/Kg w. (test in sucrose solution).

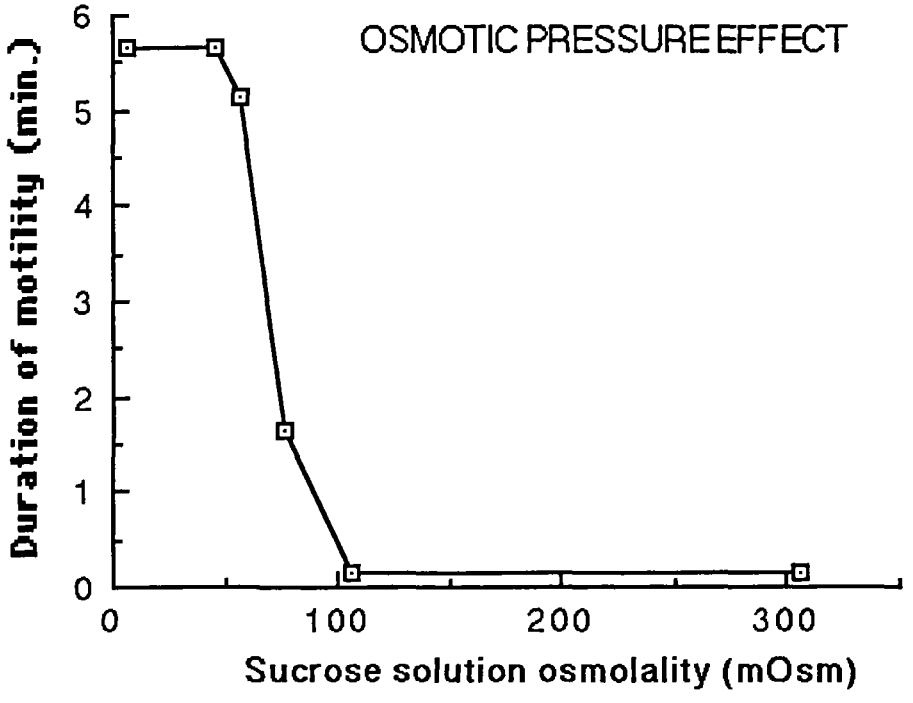


Figure 4 - Effect of osmotic pressure on the sperm motility

Nevertheless there was an apparent discrepancy between the effect of DIA saline and sucrose solutions for the same order of concentration : for instance, 78 mOsm DIA was not inhibiting. (Fig.2 and Fig.4).

This result implicated to test the eventual specific effect of the main mineral ions.

Effect of monovalent cations concentration

The effect of the two main physiological monovalent cations, sodium and potassium, was studied in the range from 0 to 20 mM (table II). Only K^+ induced a drastic inhibition of motility (from 0.1 mM). Sodium ion had no action on sperm motility.

Table 2 - Effect of monovalent cations on activation of spermatozoa in range of physiological concentrations

Ionic solutions (Chloride)	Theoritical osmolarity mOsm/kg-water	Intensity of motility
Na^+ 1 to 20 mM	2 to 40	4
K^+ 0.1 to 10 mM	0.2 to 20	0
K^+ 0.05 mM	0.1	4

Reversibility of the osmotic pressure inhibiting effect

As we saw in the previous experiments, high osmotic pressure was rapidly inhibiting : only 15 seconds were sufficient to bring motility to 0 in both the sucrose (283 mOsm) and NaCl (310 mOsm) DIA solutions. But addition of water, which provoked the decrease of the osmotic pressure, induced a reinitiation of the sperm motility. In these cases the motility intensity was indeed depending on the final osmolality : compare 47 and 32.5 mOsm. (fig.5. A and 5B).

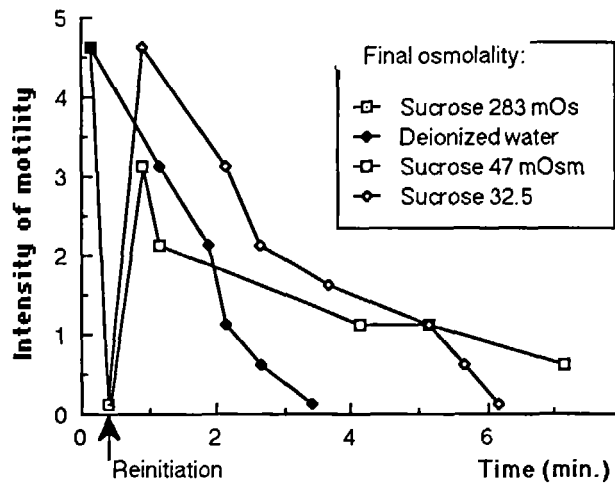


Figure 5A - Reinitiation phenomenon (↑) of sperm motility in sucrose solutions

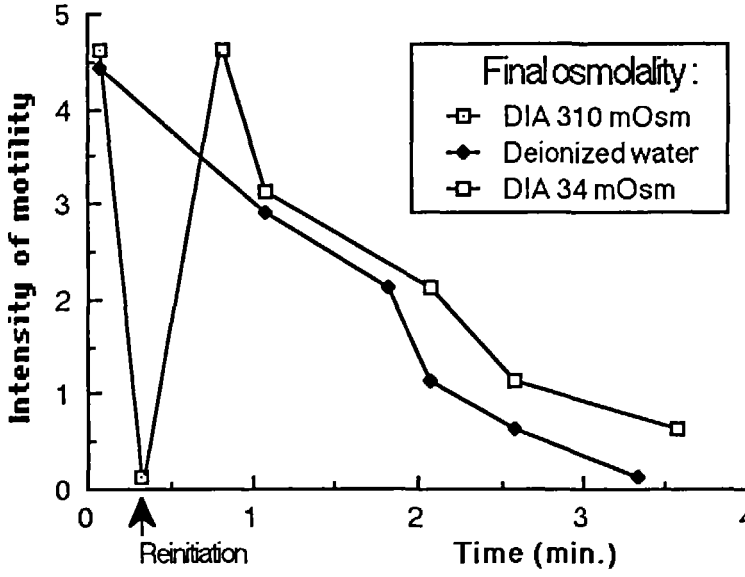


Figure 5B - Reinitiation phenomenon (↑) of sperm motility in sucrose solutions

DISCUSSION

In Siberian sturgeon, *Acipenser baeri*, it must be quoted that the osmolality (20 to 60 mOsm) of the seminal plasma strongly differs from that of freshwater or seawater teleost (range from 230 to 320 mOsm). In the laters, sodium and potassium concentration is more elevated than that observed in sturgeon seminal plasma, where the Na^+/K^+ ratio nevertheless is close to that of Salmonidae (Morisawa et al., 1983a).

The quiescence of the spermatozoa in the seminal fluid in the sturgeon may be due to the presence of inhibiting factors in the fluid or because an activating factor is absent (or both cases).

Dilution of semen is able to induce the motility of the spermatozoa, but duration and intensity of the phenomenon depend on the composition of the diluent and the rate of dilution. Different diluents were used to test some possible inhibiting factors : pH (as suggested for the trout: Baynes et al., 1981; Stoss and Holtz, 1981), osmotic pressure (as for the carp: Morisawa et al., 1983b; Billard, 1987) and potassium ion concentration (as for trout sperm: Schlenk and Kahmann, 1938; Billard, 1975; Baynes et al., 1981).

Compared to teleosts, sturgeon sperm becomes motile at low dilution ratio (below 1/6) even if sperm motility is better when dilution is more important (about 1/100). At first sight this result can be explained by the low values of inorganic cations concentrations and/or the small spermatocrit (below 5%) of the semen.

The pH of the seminal fluid is around 8.2 : Phosphate, in the HPO_4^{--}form in physiological conditions is likely responsible of the pH value. In diluents at the same pH or at a more alkaline values, there is a good activation of the sperm. The fact that dilution with deionized water, (within pH averages 6.0), does not inhibit motility, could be easily explained by the buffering capacity of 9mM Phosphate in the seminal fluid. So, as in rainbow trout (Baynes et al., 1981), pH does not prevent the sturgeon spermatozoa motility in *in vivo* conditions.

Spermatozoa of Siberian sturgeon were motile in sodium chloride (Table 2), DIA or sucrose solutions (Fig.2 and 4) isotonic to the seminal plasma (up to 60 mOsm/Kg water) evidencing that osmolality of medium is not the inhibiting factor of motility in

physiological conditions. Only higher values (70 mOsm and more with sucrose ; 310 mOsm with DIA) than in the seminal plasma showed an inhibiting effect. Nevertheless an apparent discrepancy in this osmotic preventing effect appears for the 83 mOsm DIA solution (Fig.2). It can be explained in different ways. Firstly sodium and chloride ions are not impermeant for the cell membrane as sucrose molecule is ; thus the real *in vivo* osmotic pressure is most likely smaller than the *in vitro* measured one. Secondly, it is not excluded that the electric charge of these ions can exert an activating effect on spermatozoa motility (i.e by means of change in membrane polarization) for a range of concentration (20 mM to 125 mM NaCl ; see Table 2 and Fig.2) that must be determined. For dilution with 125 mM NaCl DIA solution, osmotic effect is most likely prevailing over the postulated ionic charge effect. Investigations along these lines are now being performed in the laboratory.

The inhibition due to the hypertonic solution can be eliminated by post-dilution with deionized water of the initial diluent/sperm mixture. Immotilility of the spermatozoa is not due to their death. The function of the structures linked to the motility in the spermatozoa do not seem to be affected by the osmotic pressure action. A return to the physiological conditions, owing to the dilution, leads to a reinitiation of the sperm motility.

If, *in vivo* conditions, osmolality and NaCl do not appear as the inhibiting factors of motility, conversely only 0.1 mM KCl completely suppresses sturgeon spermatozoa motility. This effect is due to K^+ cation and not to Cl^- anion because of the lack of inhibiting effect with physiological concentration range of NaCl (1 to 20 mM). For trout sperm, inhibiting influence of K^+ concentration was first shown by Scheuring (1924) and Gaschott (1924) and later by Schlenk and Kahmann (1938), Billard (1975), Baynes et al. (1981) : weak solutions of KCl has got an inhibiting effect on trout spermatozoa motility. Seminal plasma of the sturgeon, *Acipenser baeri*, contains 2.5 mM of K^+. Thus, unlike the freshwater cyprinid fishes but as in salmonid fishes (Morisawa et al., 1983a), potassium ion is a major factor inhibiting sperm in the spermduct, and osmolality as well as chloride or sodium do not affect sperm motility in physiological conditions.

Baynes et al. (1981) and Morisawa et al. (1983a), in salmonids have found that the presence of sodium in the diluent has a mildly antagonistic effect on the potassium induced inhibition. The observed activation of motility from low dilution ratio of sperm with deionized water or diluted DIA strongly suggests such a mechanism in sturgeon. For example, dilution of sperm with water at 1/6 ratio leads to (2.5/6) 0.4 mM potassium in final mixture ; this concentration must be inhibitory. The way of antagonizing this action will be discussed in a forth-coming paper.

ACKNOWLEDGEMENTS

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ACUTE TOXICITY OF AMMONIA TO SIBERIAN STURGEON *Acipenser baeri*

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RESUME

La concentration létale à 50% (CL50) de l'ammoniaque a été déterminée chez l'esturgeon sibérien, *A.baeri*, par des tests statiques. Les 24^h NH_3 CL50 pour des poissons de 60 mg, 10 à 270 g et 450 g sont respectivement 1 mg/l, 1,7 mg/l et 2,5 mg/l. Les esturgeons sont plus résistants à l'ammoniaque que la truite (*Oncorhynchus mykiss*). D'autres investigations ont été realisé durant les tests. Les poissons montrent des signes cliniques suggérant une disfonction neurologique, les teneurs en K^+ du sang et l'hématocrite augmentent significativement en fonction de la dose d'ammoniaque mais aucune variation du taux plasmatique de Na^+ n'est apparue. Des lésions - hypertrophies, nécroses - sont visibles sur les branchies des poissons soumis à des concentrations létales. Certaines hypothèses sur la toxicité aiguë de l'ammoniaque sont discutées.

Mots clés : Ammoniaque, Toxicité, *Acipenser baeri*,

ABSTRACT

Median lethal concentrations (LC50) of ammonia were determined for siberian sturgeon (*A.baeri*) in static tests. The respective 24^h NH_3 LC50 for different weight groups - 60 mg, 10 to 270 g and 450 g - were 1 mg/l, 1,7 mg/l and 2,5 mg/l. Sturgeon are more resistant than rainbow trout (*Oncorhynchus mykiss*). Some others investigations have been made during these tests. The fish developed clinical signs suggestive of a neurogical dysfunction, the blood concentration of K^+ and the haematocrit were significantly raised in relation with ammonia, but no change appeared in Na^+ concentration. When they are exposed to lethal concentration, their gills present some lesions like hyperplasia, hypertrophy and necrosis. Some hypothesis of the acute ammonia toxicity are discussed.

Key-words : Ammonia, Toxicity, *Acipenser baeri*,

INTRODUCTION

Ammonia[1], an end products of the protein catabolism, represents 60 to 80 % of the nitrogenous excretion of fish (Smith, 1929; Forster and Goldstein, 1969; Luquet, 1972; Vellas, 1979). In intensive fish farming, excessively high doses of ammonia, issuing either from this excretion or from external pollution, can cause reductions in fish growth or even death. For these reasons, ammonia toxicity has been studied in numerous fish species (see table 3).

The complete lack of data for sturgeon on this problem led us to carry out a preliminary study of ammonia toxicity to the siberian sturgeon (*Acipenser baeri* B.) for different age groups. Thus, the intensification of sturgeon farming needs to optimize all the production's factor, especially the quality of water (Faure, 1976: Alabaster and Lloyd, 1980; Wickins, 1981; Poxton and Allouse ,1982).

In order to complete this study and illustrate the disorder caused by ammonia in fish, we observed the behavior of the animals and followed the evolution of the branchial lamella and of some blood parameters during an acute intoxication. These observations will be discussed and compared to those recorded for other species.

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[1] The term ammonia will refer to total ammonia, NH_4^+ to ammonium ion and NH_3 to non-ionic ammonia.

Fish and storage conditions

This study concerns siberian sturgeon (*Acipenser baeri* B.), obtaining from artificial reproduction and weighing between 60 mg (larvae) and 450 g (juveniles).

The larvae were randomly selected from one of the hatch tanks and transferred to one of the ten 3 liters aquariums just before the experiment (10 larvae/aquarium). The other fish were taken from the ponds where they are usually stocked and put into the experimental structure consisting of five polyester tanks (ø 1m, 200 liters) at least eight days before the beginning of the experiment. We used five to ten fish by tanks in relation with their individual weight. During this acclimatization period, the water is renewed with sufficient flow to maintain the oxygen rate above 70 % of saturation. Feeding is stopped 24 hours before the beginning of test. A comparative test was carried out on trout (*Oncorhynchus mykiss)* of 34 g coming from the same fish farm. We used the same structure and the same procedure as for sturgeon.

All the experiments were performed in the CEMAGREF experimental hatchery at Donzacq (South West of France) which has a water from a spring with very low ammonia content and a pH and temperature which are almost constant during the whole year (table 1).

Table 1 - Characteristics of water (Units: mg/l except where indicated; nd : not detectable)

Factor	Value	Factor	Value
pH	7,5 ± 0,03	Mg^{2+}	18
Temperature	16 ± 1 °C	Fe	nd
Conductivity	500 µs/cm	PO_4	nd
Oxydability	0,2 mg O_2 / L	Cl^-	21
mineral fraction	89 %	Na^+	20
MES	5	K^+	25
NO_3^-	10	Ca^{2+}	81
SO_4^{2-}	15	SiO_2	12
NH_4^+	0,017 ± 0,006	Mn^{2+}	nd
HCO_3^-	300	Phenols	0

Toxicity tests and measurement

We set out, using a serie of static 24 hours tests, to determine the lethal concentration for 50 % of the fish (24^h LC50). The experimental technique followed the AFNOR[2] NF T 90-305 (1985) ,Reish and Oshida (1987 for FAO) and EPA (1975) recommendations. At the beginning of each 24^h test, the water was no longer renewed

[2] Association Française de Normalisation.

(static test) but was aerated by compressed air to maintain the oxygen rate above 6 mg/l (> 60% of saturation). We then rapidly introduced known quantities of ammonium chloride (NH_4Cl) into four of the five tanks. We thus obtained four doses of ammonia which followed a geometrical progression and a control tank (with no ammonia). In the case of larvae, we used five to ten doses of ammonia and a control aquarium. Because of the low oxygen's consumption by the larvae, no aeration was made.

During the 24^h experiment, we noted:

- the initial and final concentration of ammonia in each tank.
- the temperature (mercury thermometer), the pH (Schott-Geräte 1/100th probe) and the oxygen content (portable YSY oxymeter) at least five times every 24 hours. The last two instruments were recalibrated before each series of measurements.
- the reactions of the fish in order to evaluate the influence and effects of ammonia to sturgeon.
- the successive deaths: we considered a fish to be dead when there was no further opercular movement.

We measured the concentration of ammonia in the water by a spectrophotometric method with blue indophenol (AFNOR NF 90-015, 1975). We doubled all measurements and readings were carried out before 12 hours. The NH_3 contents were calculated from the ammonia concentrations, the pH and the temperature using a computer program according to formulae of Emerson et al. (1975). These contents are always expressed as mg/l.

The 24^h LC50 was estimated for NH_4^+ and NH_3 from the totality of results obtained from the same weight range. We used a computer program which was realized according to the Spearman-Karber method (Hamilton et al., 1977), the AFNOR (1985) and FAO (1987) norms (method with Log/Normal transformation). In both cases, we estimated the 95% confidence interval. See more details in Salin (1987).

For two of the three series of tests on sturgeon of 270 g, we also took samples of kidney, branchial lamellae and muscle on fish just after death for a microscopic preparation (dehydratation with alcohol, inclusion, cut at 6 µm and coloration with eosin-hematoxylin). Furthermore, we took a 1 ml blood sample in heparinized tube, centrifuged (5 mins at 12.000 rotations/min) and then kept the serum at 4° C before analysis. The hematocrit was also determined after centrifugation in micro tubes. The potassium and sodium contents of the serum were measured with an Eppendorf flame photometer, the chloride content using a titration method with mercuric nitrate (Schales and Schales, 1941, slightly modified). The osmotic pressure was estimated with a freezing point osmometer and the erythrocyts' water content by the difference between the dry and fresh sample weights.

All results are presented as mean ± standard error of mean.

Statistical methods

Because of the series' low numbers we used to compare mean the non parametric U test of Mann-Whitney and for complete serie, the test T of Wilcoxon (Scherrer, 1984). For the correlations, we chose the rank coefficient of Spearman (r_s).

RESULTS

24^h LC50

The results of the whole series of tests for ammonia toxicity are resumed in table 2. We observed that the youngest fish (60 to 260 mg) are most sensitive with a 24^h LC50 of about 1 NH_3 mg/l, whereas for those weighing 450 g it is about 2.5 mg/l. It therefore appears that the sensitivity to ammonia of *A.baeri* sturgeon is age-dependent.

This phenomenon is confirmed by the study of LC50 according to the duration of exposure (figure 1) where we can also see that the variation of the LC50 according to

time is greater for the older fish than for the larvae. We observed that for the 60-65 mg and for the 270 g the data followed an exponential law:

60-65 mg : $C = 147{,}8\ e^{-0{,}043 \cdot h}$ ($r_s = -1$ n=7)

270 g : $C = 384{,}5\ e^{-0{,}0929 \cdot h}$ ($r_s = -0{,}964$ $p<0{,}005$ n=5)

with C : LC50 in mg/l ammonia
h : duration of exposure

Finally, it seems that, except for the 450g where the number of data is insufficient, the LC50 tends toward an asymptotic value of about 50 mg/l.

For the totality of the weight range used there was never any deaths for doses less than 15 mg/l NH_4^+ (0.4 mg/l NH_3), and on the other hand, there was always 100% mortality for concentrations above 215 mg/l NH_4^+ (3.8 mg/l NH_3).

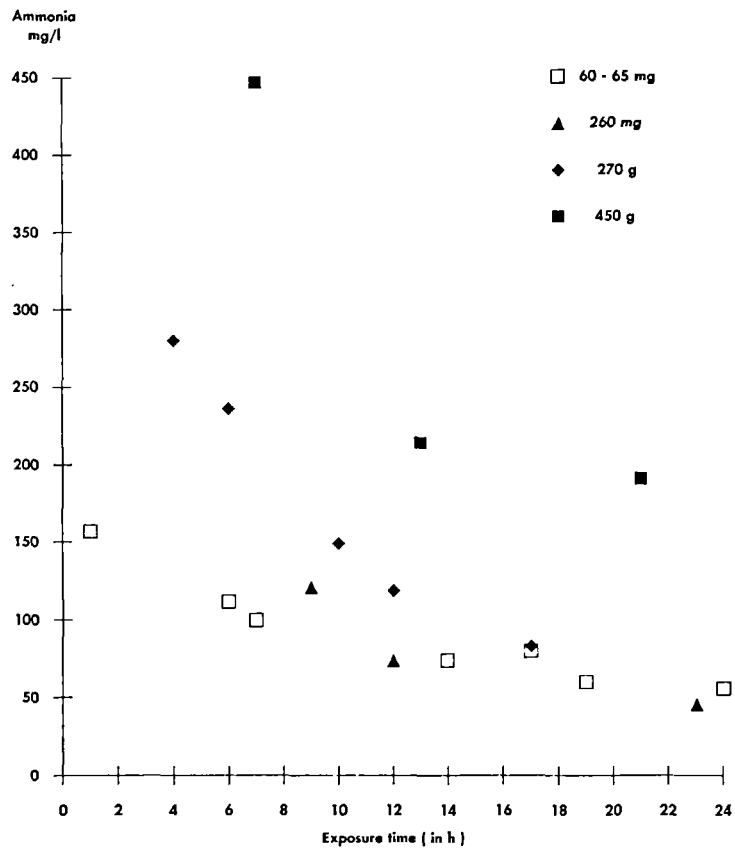


Figure 1- LC50 as a fonction of exposure time and individual weight.

As we observed an increase in pH during each test (table 2) causing a significant modification in the percentage of ammonia in the form of NH_3, we used the weighted-mean of pH during the 24^h and consequently the average percentage of un-ionized ammonia.

Table 2 - Experimental conditions and results of the toxicity tests.

Weight	Experimental conditions					24^h LC50 [3]			
	n [1]	Range		pH [2]	Temperature [2] °C	NH_4^+ mg/l		NH_3 mg/l	
		NH_4^+ mg/l	NH_3 mg/l			Spearman Karber	AFNOR FAO	Spearman Karber	AFNOR FAO
Acipenser baeri									
60 mg [1988]	8	14-112	0.40-1.99	7.7-8.1	16.5-17	57.18 (52.4-62.4)	46.96 (22.6-97.4)	1.18 (1.1-1.3)	1.05 (0.6-1.9)
65 mg [1989]	10	19-61	0.43-1.2	7.6-8.1	16.5-17	42.20 (39.8-44.7)	43.24 n.c.[4]	0.87 (0.8-0.9)	0.81 (0.7-2.4)
260 mg	4	16-75	0.48-1.00	7.7-8.1	17-18	35.12 (29.3-42.1)	34.94 (11.4-107.4)	0.82 (0.7-0.9)	0.83 (0.3-2.6)
10 g	4	40-250	0.6-2.5	7.5-7.7	18-21	147.58 (113.5-191.9)	112.54 n.c.	1.76 (1.4-2.2)	1.41 n.c.
270 g	9	59-149	1.12-3.55	7.6-8.1	17-20	79.79 (76.8-82.9)	80.59 n.c.	1.65 (1.5-1.7)	1.71 n.c.
450 g	8	89-215	1.74-3.77	7.4-8.2	14-16	166.28 (150.2-184.0)	175.44 n.c.	2.40 (2.3-2.5)	2.21 (0.5-9.4)
Oncorhynchus mykiss									
34 g	7	14-40	0.4-1.42	7.6-8.2	16-17	29.02 (26.9-31.3)	27.24 n.c.	0.75 (0.71-0.84)	0.81 (0.2-2.6)

[1] Number of concentrations use for calculations.
[2] For pH and temperature, values from begining to end of the test.
[3] For LC50, result and 95% confidence interval.
[4] n.c.: non calculable.

In order to test the influence of these variations on the calculation of the 24^h LC50, a complementary experiment was carried out using demineralized water (no change in pH) and larvae of 65 mg. The LC50 in demineralized water for these *A.baeri* is 60 mg/l NH_4^+ and 1.01 mg/l NH_3 (instead of 42 and 0.87 respectively for Donzacq water). Given the precision of the methods used, these results are sufficiently close (p=0.05, T test) to accept that variation in pH and the way of calculating the NH_3 form do not produce excessively large errors in 24^h LC50 values.

The comparative test with fingerling trouts of 34 g in the same experimental conditions (tanks, quality of water, measuring and calculating techniques) allowed us to calculate a 24^h LC50 of 29 mg/l NH_4^+ and 0.75 mg/l NH_3. We therefore established that sturgeon of comparable weight (10-270 g) are almost twice as resistant to ammonia as trout.

Behavior of fish

The observations of *A.baeri* during the toxicity experiments allowed us to define the following clinical signs:

- a period when the fish remain calmly at the bottom of the tanks and increase ventilation.

- then, jumps, more or less complete tetany with spasmodic and anarchic movements.
- loss of equilibrium, swimming on back side.
- finally, a very violent tetany followed by the death.

We also noticed the presence of an increased amount of mucus in fish submitted to sub-lethal doses of ammonia as compared with the control cases (in particular on the gills).

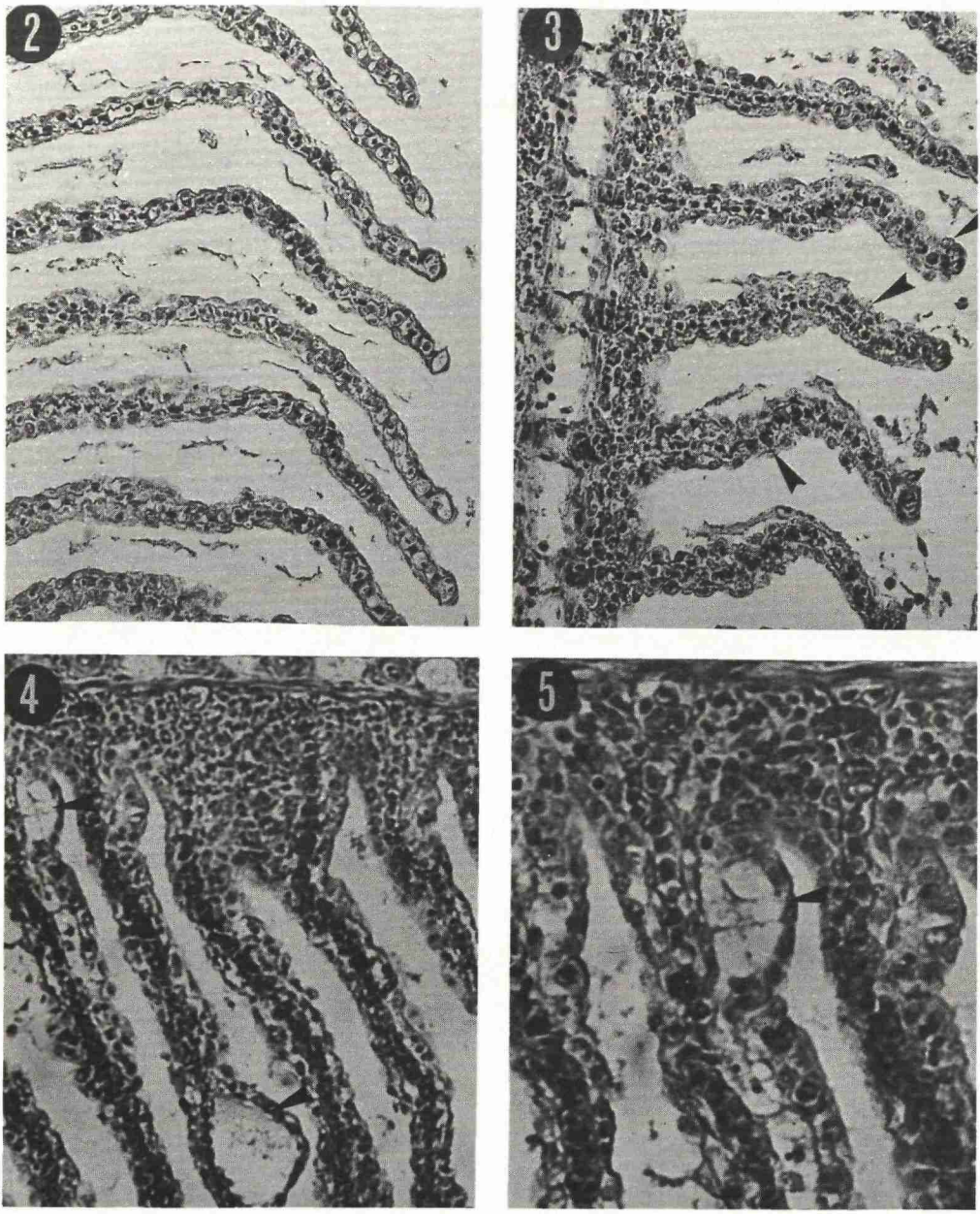
These reactions vary with time according to the dose of ammonia, but we observed also that, for the same dose, fish have different individual reactions. All the intoxicated fish that did not die after the 24^h of the experiment recovered their equilibrium if they were returned to clear water (without ammonia). They began to eat again and behave normally after 24^h in fresh water.

Microscopic observations

The microscopic fixations of the gills of fish weighting 270 g and exposed to more than 60 mg/l of ammonia (about 30% of mortalities) reveal a modification of the epithelium of the secondary lamellae with sometimes the presence of oedeme (fig 2 and 3). The volume of epithelial cells is multiplied by 2 or 3, the base of filament is slightly turgescent and the blood space is dilated. In sturgeon exposed to 80 mg/l of ammonia - 1.6 mg/l NH_3 - which causes 100% mortality, the preceding phenomena are even more accentuated. In addition to the increase in cellular volume, we observed necrosis and detachment of the epithelium cells. In certain cases, first signs of hyperplasia appeared (fig 4 and 5). For the kidney and the muscle cuts, we noticed no difference between the control fish and those exposed to lethal doses of ammonia.

Hematological analyses

According to blood analyses, the hematocrit appears in direct proportion to the dose of ammonia, the sodium and chloride contents were stable but we recorded an increase in the potassium concentration (fig 6a, 6b, 6c). We could note that the potassium levels were about 3.7 meq/l when the mortality rate is under 10%, then they increase to 6.5 meq/l when the ammonia contents causes more than 50% mortality. The water content of the erythrocyts is not modified, but the osmotic pressure of the serum of fish exposed to lethal doses (289 ± 10 mosm, n=5) is significantly higher than that of the control fish (257 ± 7, n=9) [p=0.028; U test]. This last result is however partial and should be confirmed.



Figures 2 - 5: Modifications of the gills epithelium during the 24^{h} exposure to ammonia.
(2) Control (x 50).
(3) Exposition to 60 mg/l ammonia: oedemes and dilations of the blood space (x 50).
(4) Exposition to 80 mg/l ammonia: detachments of the epithelium cells and hyperplasia (x 75).
(5) Detail from figure 4 (x 100).

Fig 6a :

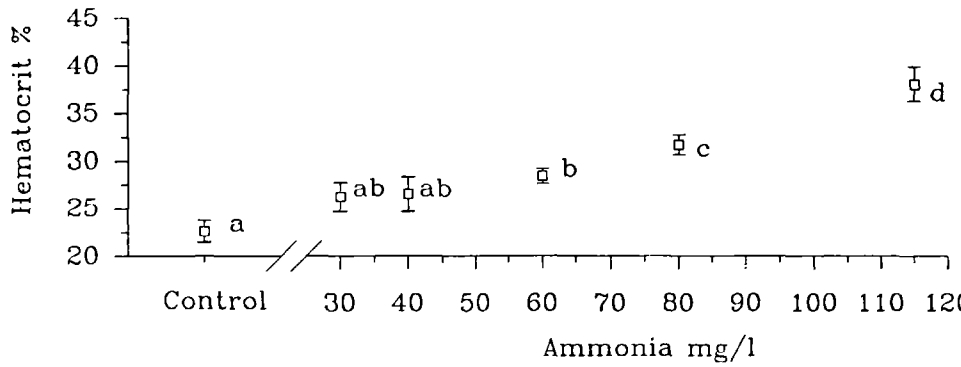


Fig 6b :

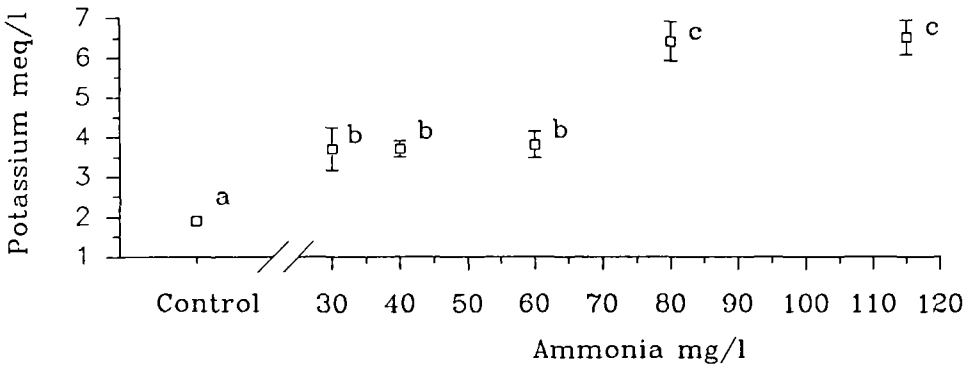


Fig 6c :

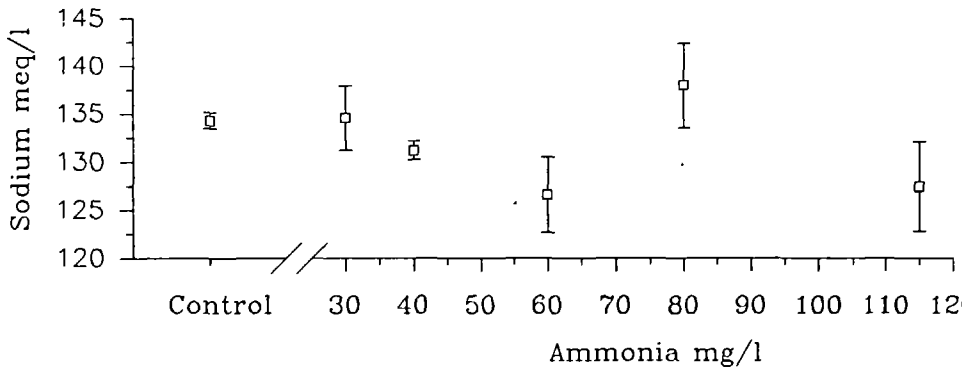


Figure 6 : Evolution of some blood parameters as a fonction of ammonia concentration.
Mean ± s.e.m. (control n = 9, others n = 5).
Data followed by a same superscript letter are not significantly different (Test U, 5 % level).

24^h LC50

The results of tests indicate that the 24^h LC50 of sturgeon (*Acipenser baeri* B.) is situated in the range of values found in other species. They are less sensitive than the trout (*Oncorhynchus mykiss*) - their 24^h LC50 commonly determined is around 0.5 mg/l NH_3 - but more sensitive than the catfish (*Ictalurus punctatus*) - 24^h LC50≈3 mg/l NH_3 - (table 3). The tolerance level of sturgeon to ammonia is about the same as that of the carp (*Cyprinus carpio*) (Dabrowska and Sikora, 1986; Hasan and Macintosh, 1986). However, it is sometimes difficult to make direct comparison, because the methods of calculating the LC50 are not the same and can, in some cases, lead to uncertain results. Thus, the method using a Log-Normal transformation only produced reliable results if one can really calculate a significant linear regression. This is why, in a certain number of our experiments we were not able to calculate the confidence interval because of the lack of a good correspondence between the experimental data and the theoretical line ($p > 0.05$).

The difference in sensitivity to ammonia according to age (*A.baeri* larvae are more sensitive than older fish) is a result already observed in other species. Thus, the eggs are very resistant - no effect for concentrations over 3.5 mg/l NH_3 (*Oncorhynchus mykiss*: Rice and Stockes, 1975; *Scianeops ocelatus*: Holt and Arnold, 1983; *Cynoscion nebulosus*: Daniels et al., 1987) - larvae become more sensitive during the reabsorption of the vitelline reserve, then the fry and adults are once again more resistant (*Ictalurus punctatus*: Colt and Tchobanoglous, 1976-1978; *Oncorhynchus mykiss*: Burkhalter and Kaya, 1977; Rice and Bailey, 1980; Thurston and Russo, 1983).

It seems necessary to recall that the LC50 values also depend on the quality of the water used during the tests and on the execution of the latter. Thus, during our experiments, the pH - and therefore the percentage of the NH_3 form - developed exponentially and stabilize 6 to 7 hours after the beginning of the tests. This phenomenon is probably due to a modification in the calco-carbonic equilibrium because of the aeration (Bairds et al., 1979). However, with larvae of 65 mg, the 24^h LC50 value obtained when using demineralized water - where the pH remained stable - is not significantly different from that using water from the pisciculture ($p=0.05$; T test).

The 24^h LC50 values seem to depend on the ion calcium content of the water. Thus, Tomasso et al. (1980) also recorded an influence of the hardness of the water on the toxicity of ammonia to the catfish and supposed this to be due to a competition between calcium and ammonia ions. This hypothesis is confirmed by the 24^h LC50 in trout, which is usually about 0.5 mg/l NH_3 (Ball, 1967; Rice and Stokes, 1975; Hillaby and Randall, 1979; Haywood, 1983; Thurston and Russo, 1983; Meade, 1985), whereas we found with the hard water of Donzacq a LC50 of 0.75 mg/l NH_3. We can suppose that the 24^h LC50 for water with lower calcium level would be slightly different.

We can nevertheless conclude that *A.baeri* sturgeon are relatively less sensitive to ammonia than most of the other species (especially the salmonids).

Influence of ammonia on the gills structure

Microscopic observations show a significant modification in the structure of the branchial lamellae for ammonia doses of over 60 mg/l NH_4^+ (for doses causing 30 to 100 % mortality in 24^h). On the other and, modifications are insignificant, even more non existent, for lower doses. These phenomena are also recorded for carp, *Cyprinus carpio* (24^h at 30 mg/l NH_4^+; Yang and Chun, 1986) and for trout, *Oncorhynchus mykiss* (6h at 34 mg/l NH_4^+; Smart, 1976). These alterations are classically described in longer experiments on sub-lethal concentrations with occasionally some tissue proliferation, fusion of branchial lamellae and increased in sensitivity to bacteria (Smith and Piper, 1975; Smart, 1976; Robinette, 1976; Thurston et al., 1984; Lang et al., 1987).

Some of these authors also believe that this modification of the gills, and consequently of respiratory capacity, is one of the causes of the decrease in the growth rate in fish exposed to sub-lethal doses of ammonia for several weeks.

Table 3 - Bibliographic's studie of toxicity tests.

Species	Weight	Type of tests	Results (NH_3 mg/l)	References
Oncorhynchus mykiss		24h LC50	0,61	Herbert and Shurben (1963)*
	40 g	24h LC50	0,41	Ball (1967)
	eggs - 50 d	24h LC50	> 3,5	Rice and Stokes (1975)
	80 days	24h LC50	0,068	"
	adults	24h LC50	0,097	"
	1 g	96h LC50	0,6 - 0,8	Thurston et al. (1978)
	3,5 g	96h LC50	0,5 - 0,6	"
	60 mg	96h LC50	0,45	Thurston and Russo (1983)
	250 mg	96h LC50	0,44	"
	10 g	96h LC50	0,62	"
	250 g	96h LC50	0,32	"
	500 g	96h LC50	0,28	"
	21 g	96h LC50	0,5	Thurston et al. (1981)
	250 g	96h LC50	0,29	"
	2600 g	96h LC50	0,16	"
	22 g	96h LC50	0,4 - 0,6	Arthur et al. (1987)
	larvae	96h LC50	0,25	Bukhalter and Kaya (1977)
Oncorhynchus kisutch	6 g	14h to 96h LC50	0,45	Buckley (1978)
Cynoscion nebulosus	larvae	24h LC50	0,28	Daniels et al. (1987)
	400 mg	24h LC50	1,38	"
	8,8 g	24h LC50	1,98	"
Pimephales promelas	200 - 300 mg	96h LC50	2,5	Thurston et al. (1983)
	1 - 3 g	96h LC50	1,4	"
Ictalurus punctatus		24h LC50	1,4 - 1,8	Tommasso et al. (1980)**
		24h LC50	2,92	Sparks (1963) *
	16g	24h LC50	0,75 - 2,1	Sheehan and Lewis (1986)
	1 g	96h LC50	3,1	Colt and Tchobanoglous (1978)
		96h LC50	3,2	Ruffier et al. (1981)
	3 - 7 g	96h LC50	1 - 1,3	Arthur et al. (1987)
		96h LC50	3	Roseboom and Richey (1977)**
	3 - 4 g	96h LC50 (30 °C)	3,8	Colt and Tchobanoglous (1976)
	"	96h LC50 (22 °C)	2,4	"
Perca fluviatilis	14 g	24h LC50	0,29	Ball (1967)
Rutilus rutilus	9 g	24h LC50	0,62	"
Abramis brama	16 g	24h LC50	0,58	"
Cyprinus Carpio	300 mg	24h LC50	1,8 - 2,1	Hasan and Macintosh (1986)
	"	48h LC50	1,8 - 1,9	"
	"	96h LC50	1,7 - 1,8	"
	30 g	48h LC50	1,6 - 2	Dabrowska and Sikora (1986)
Acipenser baeri	60 mg - 450 g	24h LC50	1,5 - 2,5	This study (1989)

* : in Ruffier et al. (1981)
** : in Haywood (1983)

However, it does not appear that these alterations are sufficiently significant to provoke death in the case of acute toxicity (Smart, 1976).

Influence of ammonia on nervous system

Our observations of the symptoms of ammonia intoxication - loss of equilibrium, spasmodic and erratic movements, hyperventilation - are described by all researchers working on acute toxicity tests (Smart, 1976,1978; Colt and Tchobanoglous, 1976; Thurston et al., 1978,1981,1983; Buckley, 1978; Hillaby and Randall, 1979; Ruffier et al., 1981; Haywood, 1983). These phenomena, which can be characterized as neurological disorders, are extremely rapid if the ammonia doses are high. Thus, in one of the tests, the concentration obtained being ten times higher than that desired (500 to 1000 mg/l NH_4^+), the sturgeon showed these clinical signs in less than 30 minutes with very violent reactions and deaths began after an hour of exposure. In the same way, Hillaby and Randal (1979) recorded that these signs begin in trout five minutes after an injection of 300 µmol of NH_4Cl / 100 g of fish.

Several hypotheses have been put forwards to explain these symptom:

- either an interaction of ammonia on the neuromuscular centers, in particular those of the respiratory and cardiac muscles (Schwartz 1958 in Mutch and Bannister, 1983; Smart, 1976; Bubien and Meade, 1986). This phenomenon would explain the hyper ventilation, the convulsions and the erratic movements noticed during all our experiments.
- or a modification of biochemical reactions of central nervous system, in particular a decrease in ATP levels and an increase in those of glutamin in direct proportion to the concentration of ammonia (Walker and Schenker, 1970; Levi et al. 1974; Schaffi, 1980). This drop in available energy (ATP) would cause a dysfunction of the brain. This hypothesis had already been suggested by work carried out on mammals presenting hepatic coma.

These two phenomena of effects of ammonia, both on the neuromuscular centers and on the brain's energy reserves are probably the main causes of death by acute ammonia intoxication.

Influence of ammonia on blood parameters

The hematocrit and ions levels of the control fish (270 g, n=9) are very close to the results found in previous studies on *A.baeri* (Williot, not published; Ait-fdil, 1986; Bennouna, 1986) or on other species of sturgeon (Magnin, 1962; Potts and Rudy, 1972; Natochin et al., 1975; Hunn and Christenson, 1977; Lavrova et al., 1984).

We notice in figure 6a an increase in the hematocrit in direct proportion to the ammonia concentration. This is not caused by an increase in volume of the erythrocyts, for their water content is not significantly modified. This phenomenon is not always observed in other species. Indeed, as concerns the blood parameters of fish exposed to sublethal doses of ammonia, there is no modification in the number of erythrocyts in carp (Dabrowska and Wlasow, 1986), nor in salmonids (Smart, 1978; Buckley et al., 1979). And the other hand, Korting (1969 in Sousa and Meade, 1977) reports increase in the hematocrit of carp exposed to high doses of ammonia and suppose that this is due to a transfer of plasma towards the tissues. This hypothesis could explain both the evolution of hematocrit and the significant increase in osmotic pressure in *A.baeri* exposed to lethal doses of ammonia.

Finally, the stress caused by ammonia intoxication and the setting up of the experiment could be sufficient to explain the modification of hematocrit by a significant splenic clearance (Soivo and Oikari, 1976 on *Esox lucius*). This would not however explain why this increase in hematocrit should be proportional to ammonia dose.

Amongst the blood parameters studied, only potassium undergoes a significant modification in the presence of ammonia (U test, $p<0.05$). Moreover, we noticed that this increase seems to appear at two stages, one for doses not causing more than 10% mortality and the other for concentrations over the 24^h LC50 (fig 6b). It therefore seems

there is a two steps phenomenon. The system regulate itself until the dose of ammonia saturates the process, causing then a strong increase in the rate of potassium in the blood.

We know that in experiments on ammonia toxicity, there is an accumulation of ammonia in the blood (Fromm and Gillette, 1968; Haywood, 1983; Thurston et al., 1984). Furthermore, for 20 years it has been clearly established that ammonia ion (NH_4^+) can take the place of potassium in active transports like Na-K-ATPases (Maetz and Garcia-Romeu, 1964; Maetz, 1973; Busacker and Chavin, 1981; Claiborne et al., 1982; Borgati et al., 1985; Garvin et al., 1985; Balm et al., 1988). Thus, Ait-fdil (1986) shows in *A.baeri* the existence of Na-K-ATPases in the gills, erythrocyts and kidney, with a greater activity in the presence of ammonia ions as compared to potassium. The substitution of K^+ by NH_4^+ would allow normal exchanges to be maintained for sodium between the blood and the tissues (fig 6c). On the other hand, when the competition between ammonia and potassium is too strong, we can imagine that it causes an increase in the blood's potassium content. This hypothesis, already put forward by Bubien and Meade (1986) for *Oncorhynchus mykiss*, needs to be verified by measuring the ammonia content in the blood, and also by measuring the totality of ions in different tissues, for example in erythrocyts.

CONCLUSION

The initial aim of this work was to situate the sensitivity of the siberian sturgeon (*Acipenser baeri* B.) to ammonia in comparison with other species. This sensitivity is currently determined by normalized tests of short duration, even if their practical application is difficult. In this framework, we showed that *A.baeri* are more resistant than *Oncorhynchus mykiss*. If we compare the results of this study with those obtained by other researchers on different species, we can class the sensitivity to ammonia of the fish as follow:

Oncorhynchus mykiss > *A.baeri* ≃ *Cyprinus carpio* > *Ictalurus punctatus*

By complementary observations, we tried to characterize the effect of high doses of ammonia on sturgeon. On the one hand, we observed tetany and alteration of the branchial tissues, on the other, we showed modifications in hematological characteristics. However, the interpretation of a part of these observations remains difficult and specific studies are necessary.

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ULTRASTRUCTURE D'UN CHEMORECEPTEUR BRANCHIAL CHEZ UN POISSON CHONDROSTEEN : *Acipenser baeri*

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RESUME

L'étude ultrastructurale des bourgeons du goût de l'esturgeon (*Acipenser baeri*) montre qu'ils sont localisés sur la surface pharyngienne de l'arc branchial. Ils s'étendent de la basale à la surface externe de l'épithelium et sont constitués de trois types de cellules : 1) des cellules présentant de nombreux tubules associés à des mitochondries ; 2) des cellules dont le cytoplasme contient des vésicules denses et des paquets de microfilaments. Le pôle apical des types 1 et 2 comporte des microvillosités courtes ; 3) des cellules basales qui contiennent parfois de très petites vésicules à coeur dense. Un grand nombre de fibres amyéliniques afférentes sont concentrées à ce niveau et viennent s'intercaler entre les cellules sensorielles. Ces fibres contiennent des mitochondries, des vésicules de type synaptique et parfois quelques vésicules à coeur dense. Elles se rassemblent pour constituer un nerf dans le conjonctif sous-jacent. Bien que l'esturgeon soit un poisson "primitif", ces organes sensoriels ont une structure très proche de celle des bourgeons gustatifs des autres vertébrés.

Mots clés : branchies, bourgeons gustatifs, ultrastructure, *Acipenser baeri*.

ABSTRACT

The taste buds of the sturgeon *Acipenser baeri* have been located on the pharyngeal surfaces of the gill arches. They spread from the basal to the external limit of the gill epithelium and consisted of three types of cells : 1) cells with numerous microtubules associated with mitochondria ; 2) cells contained many dense vesicles and packs of microfilaments. The apical tip of these two types of cells was characterized by small microvilli ; 3) basal cells contained small electron dense vesicles. A lot afferent myelin-free nervous fibers were concentrated at this level and were closed together with sensory cells. These fibers contained numerous mitochondria, synaptic vesicles and some electron dense vesicles. They joined to each other and made up a nerve in the underlying conjonctive tissue. Despite differences in gill organization between the Chondrostean and the Teleosts species, the taste buds of the sturgeon themselves were ultrastructurally and closely resembled those described in other fishes and other vertebrates.

Key-words : gills, taste buds, ultrastructure, *Acipenser baeri*.

POPULATIONS SAUVAGES

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STURGEONS IN HUNGARY, PAST AND PRESENT SITUATION

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RESUME

Cette note fait le point de la situation des esturgeons naturellement présents dans la partie hongroise de fleuves internationaux, ainsi que des essais d'élevage menés sur *Acipenser ruthenus* et sur 2 de ses hybrides avec d'autres espèces. Trois espèces, *Huso huso*, *Acipenser gueldenstaedti* et *Acipenser nudiventris* sont protégées depuis 1989. Les derniers spécimens capturés l'ont été respectivement en 1987, 1980 et 1989. On peut considérer que *Huso huso* et *Acipenser stellatus* ont disparu des eaux hongroises. *Acipenser ruthenus* est pêché depuis longtemps à des fins commerciales et sportives. L'ensemble des captures annuelles représente environ 22,5 tonnes en 1988. C'est l'espèce la plus prometteuse pour l'avenir. Il semblerait que ce résultat soit en partie dû aux efforts de repeuplement entrepris depuis de nombreuses années. On décrit l'évolution de la stratégie de reproduction artificielle. Les essais d'élevage du bester ont dû être abandonnés à cause des difficultés d'approvisionnement. Les tentatives plus récentes, couronnées de succès, de produire des hybrides entre *Acipenser ruthenus* et *Acipenser baeri* sont très encourageantes.

Mots clés : *Acipenseridae*, espèce protégée, captures, élevage, hybrides.

ABSTRACT

This paper in one hand reviews the situation of sturgeons naturally present in Hungarian part of international rivers and in other hand presents several attempts in farming *Acipenser ruthenus* and 2 of its hybrids with other sturgeon. Three species, *Huso huso*, *Acipenser gueldenstaedti* and *Acipenser nudiventris* are under protection since 1989. The last individuals have been respectively catched in 1987, 1980,and 1989. We look upon *Acipenser stellatus* and *Huso huso* are vanished of Hungarian waters. *Acipenser ruthenus* is catched since a long time for commercial and recreational purposes. Altogether, 22,5 tons have been landed in 1988. This the most promising species in the future. This result could be partly due to restocking activities undertaken during several years. The evolution of artificial breeding methods is described. The tries in farming bester have been stopped because of supplying difficulties. More recent and successfull attempts in producing hybrids between *Acipenser ruthenus* and *Acipenser baeri* are very promising.

Key-words : *Acipenseridae*, catches, protected species, farming, hybrids.

INTRODUCTION

Sturgeons have been regarded as species of primary importance in the history of Hungarian fisheries. Fishing communities of great traditions have been developed in the vicinity of spawning grounds of these species along the Danube and on the banks of the Tisza River. In the last decades, most of the fisheries research have been concentrated on Balaton Lake and on pond farming problems, as a consequence our knowledge of sturgeons ecology is rather poor.

This paper deals with the evolution of sturgeons in Hungarian parts of international rivers, Danube, Tisza and Drava. In this way the situation of *Huso huso*, *Acipenser gueldenstaedti*, *Acipenser nudiventris*, *Acipenser stellatus* and *Acipenser ruthenus* is briefly discribed. More recently, some farming trials have been developed on *Acipenser ruthenus* and their hybrids with *Huso huso* and *Acipenser baeri*.

Great sturgeon - Huso huso L.

In the Middle Ages great sturgeon was one of the most important subject of Hungarian fisheries. Especially upstream migration of this species in the Danube determined livelihood of fishing communities. In the XVIII century sudden decrease of migrating populations took place and by the second half of the XIX century great sturgeon became an occasional haul of fishermen. Since 1857 practically every specimen taken in Hungary have been registered (Herman, 1887; Khin, 1957).

A series of great sturgeon captures had been interrupted after 1963 and, especially following daming the lower Danube in the Iron Gate in 1971, it was generally accepted that this species would never return to Hungarian waters.

It was a great surprise that a great male sturgeon (TL - 300 cm, W - 181 kg) was catched by a group of fishermen from Danube at Paks(Fig. 1) (1526-1528 river km) on the 16 th of May 1987. Because of this specimen , great sturgeon has been under protection since 1989. For all that the species should be regarded as practically extinct in Hungary.

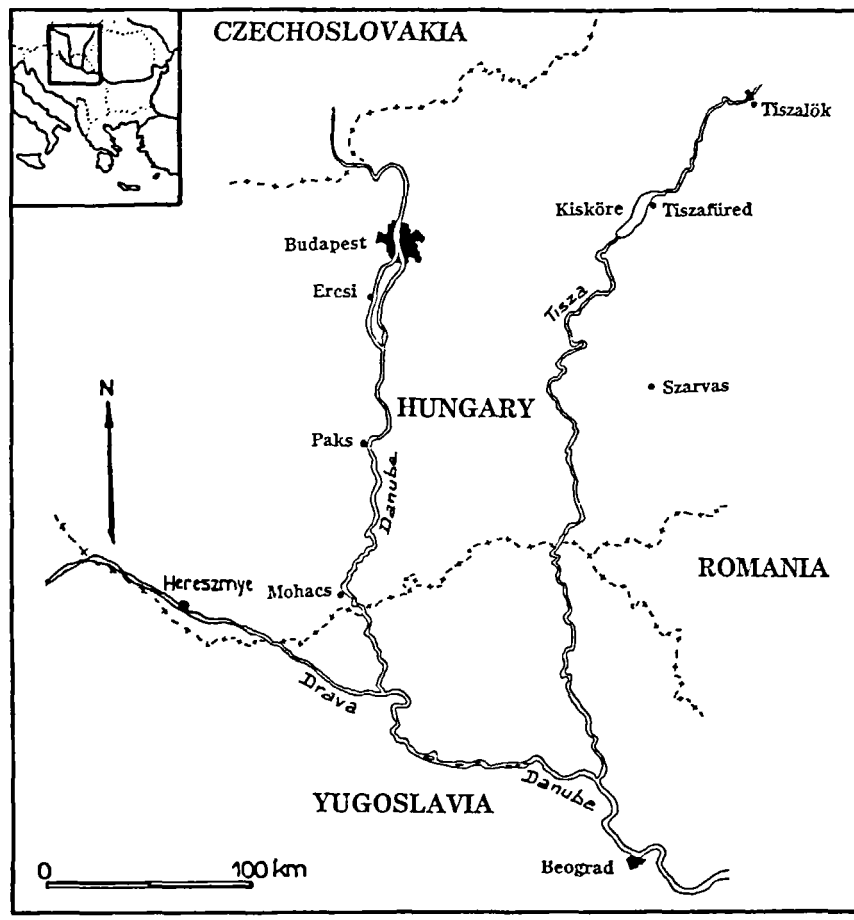


Figure 1 - Hungarian parts of rivers where sturgeons are present

Russian sturgeon - Acipenser gueldenstaedti colchicus Marti

Fishery importance of this species was always inferior to that of great sturgeon. From the end of the XIX century only sporadic catches were registered, however on the basis of these data its permanent occurence in Hungarian waters can be stated. In 1970 a juvenile specimen (LT ~40 cm) was caught from the Danube at Paks (Fig. 1) (Penzes, 1970) while the next specimen (TL - ~ 100 cm) was reported from middle Tisza at Tiszafüred in 1980 (Harka, 1980). Anglers talk also on other specimens but these catches can not be verified.

Taking into consideration these data together with the fact that russian sturgeon is still reported in Yugoslovian commercial catches above the Iron Gate, existence of non-anadromous population in the Danube system seems likely. The species has been under protection since 1989.

Ship sturgeon - Acipenser nudiventris Lovetzky.

On the basis of historical records it seems impossible to evaluate the role of this species in Hungarian fisheries. Larger specimens were reported in general in the past without distinction from russian sturgeon while smaller ones have been confused with sterlet even by professional fishermen (lack of vernacular names for this species in Hungary !). The largest specimen known in Hungary was taken from the Danube at Ercsi (Fig. 1) in 1932 (TL - 170 cm, W - 32 kg). Vasarhelyi (1957) reported numerous juvenile specimens from the upper Tisza after construction of the Tiszalök dam (Fig.1). At the same time he stressed that adults were rare.

In August 1975 a specimen of ≈ 70 cm TL was caught by an angler from middle Tisza at Kiskőre (seen by the author of this review on good quality photos sufficient to control determination). In 1979 two dead specimens were observed at Tiszalök following a catastrophic fish-kill but their identification was not unambigous. The latest definitely determined specimen was caught from the Drava River at Heresznye (185-186 river km/ on 25th of August 1989. It was a male of 147 cm TL and 20.5 kg W). The species has been under protection since 1989.

Sterlet - Acipenser ruthenus L.

Sterlet is the only common sturgeon species in Hungary. It plays an important role in both commercial and sport fisheries. On the basis of commercial statistics decrease of stocks was reported in the 1950s and 1960s, explained mainly by increasing level of water pollution (Tóth, 1960; Jaczo, 1974). Improvement of commercial catches in the Danube was observed in the middle 1970s (Tóth, 1978, 1979). The figure 2 gives evolution of both commercial and sport catches during the last three decades. These data have not been analysed in details but some tendencies are rather visible. Unfortunately data on fishing effort are not available but in comparison of sport and commercial fisheries results it should be taken into consideration that number of licenced anglers rised 7-fold (!) while the number of professional fishermen decreased by about 50 %.

Improvement of stocks can be explained by three main factors :

- improvement of water quality (especially in the Danube section above Budapest),
- regular enhancement of stocks with fry reared in hatcheries,
- positive effect of dam constructions in the Yugoslavian sections of the Danube and Tisza.

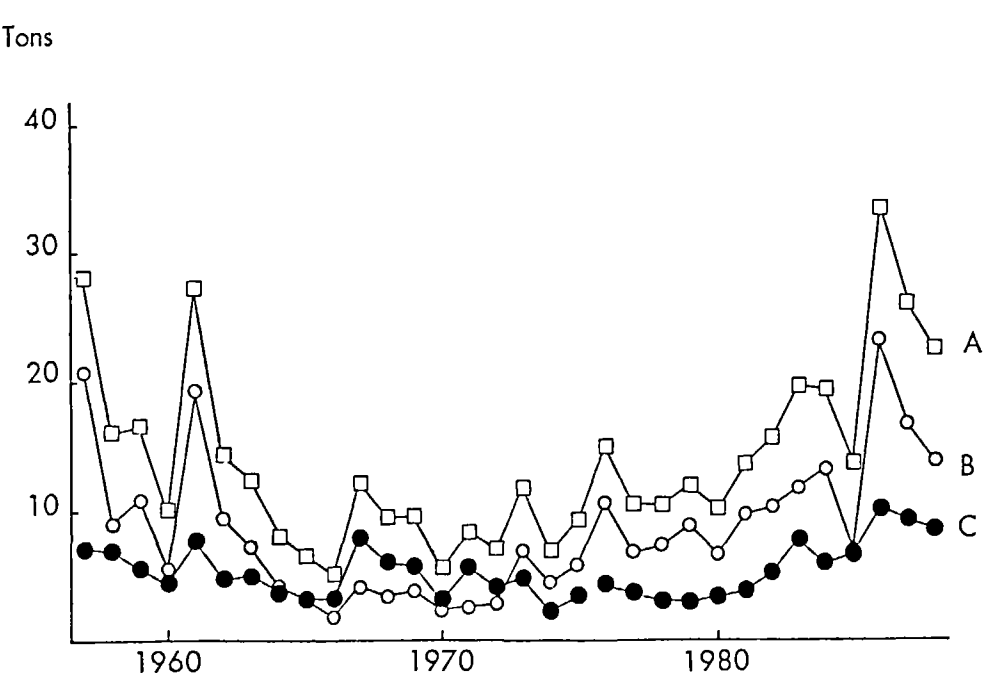


Figure 2 - Acipenser ruthenus catches in Hungary (1957-1988).
A = total ; B = commercial ; C = sport.

Sequency of these factors can be discussed only theoretically as research data are not available. In the author's opinion dramatic character of changes indicates dominant role of construction works.

In spite of fluctuations in commercial catches, present situation of sterlet stocks in larger Hungarian rivers can be regarded as very promising.

Stellate sturgeon - Acipenser stellatus Pallas

Earlier records on the occurence of stellate sturgeon are known practically from the entire Hungarian sections of Danube and Tisza as well as from their main tributaries, but even in historical times it played a minor role in fisheries. The last specimen (TL - 100 cm) was taken from Danube at Mohacs(Fig.1) in 1965. The species is regarded as extint in Hungary.

FARMING TRIALS

Sterlet - Acipenser ruthenus L.

As a consequence of unstable and decreasing sterlet catches from Hungarian rivers, artificial propagation of this species became a highlighted problem as early as in the 1950s (Lanyi, 1951; Jaczo, 1953). The use of common carp (*Cyprinus carpio* L.) pituitaries has had satisfying effect only on males (Jaczo, 1969). As *Acipenser* pituitaries are practically unobtainable in countries where sterlet is the only common

representative of the genus, a non-induced method of artificial propagation has been developed.

According to this, both sexes are collected on natural spawning grounds in early May. Eggs are obtained by opening abdominal cavity, then fertilized artificially. Following the hatchery procedures, larvae are reared in throughs or basins. (Tamás, 1975; Horváth et al., 1982) and young sterlet fry are released in natural waters.

Although application of this method has contributed to restoration of natural stocks in Hungarian rivers, it has several restrictions from the point of view of further development and new ways of fish seed production should be searched. In this respect experiments with synthetic LH-RH hormone (Horváth et al., 1986) are hopeful.

Raising one-year old or even larger sterlets in controlled environment proved to be uneconomic and results of such experiments haven't been published at all.

Bester - Huso huso x Acipenser ruthenus

In the 1970s trials for culture of bester were carried out in institutions and commercial pond farms with larvae imported from the USSR. In fry rearing similar methods were used as in the case of sterlet (Tamás, 1975). Raising of one-summer old fish in both ponds and net cages was successfully solved by the Warm-Water Hatchery (Tehag) at Százhalombatta (Tölg et al., 1978). Müller (1978) then Müller et al. (1980) reported experimental data from a net cage unit operated in the oxbowl-lake near Fish-Culture Research Institute at Szarvas. In the first season fish of 583 g average final weight were reared with a yield of 16.26 kg/m^3. Market-size (average weight : 1369 g) was achieved at the end of the second season. Duration of seasons were 153 and 150 days respectively, interrupted by a wintering period. Süto et al. (1977) reared market-size besters of 800 g in the State Farm of Tata.

Farming trials came to a stop because of difficulties in supply of stocking material from import sources.

Siberian sturgeon - Acipenser baeri stenorhynchus Nikolsky

Acclimatization of Siberian sturgeon was started by the Fish-Culture Research Institute at Szarvas in 1982. Breeding population has been raised successfully under intensive conditions. Different rearing experiments are being carried out with pure line Siberian sturgeon and - Siberian sturgeon x sterlet - hybrid groups, (Ronyai et al., 1991 a, 1991 b).

CONCLUSION

Efficiency of legal measures in protection of sturgeons is rather doubtful. Baning of catches excludes fishery organizations from participation in active protection of the species in question. In case of sterlet correct determination of closed seasons and size-limits seems to be sufficient. Protection of ship sturgeon and Russian sturgeon should be further maintained, as our knowledge on the ecology of these fishes is very poor.

Artificial enhancement of sterlet stocks should be continued but at the same time efficiency of stocking programmes should be investigated.

A special research programme should be started on ecology of ship sturgeon and Russian sturgeon to verify the theoretical concept of non-anadromous populations in the Danube system. In view to further improvement of water quality even a regular stocking programme of these two species seems to be less fantastic than hitherto. Such a venture can be realized only in close cooperation of interested Czechoslovakian, Hungarian and Yugoslavian fisheries organizations.

Experiments with Siberian sturgeon and its hybrids should be continued in order to find high-value fish with short production cycle to farmed in high-tec units.

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EVOLUTION DES CAPTURES DE TROIS ESPECES D'ESTURGEONS *Huso huso, Acipenser gueldenstaedti,* et *Acipenser stellatus* DANS LA PARTIE YOUGOSLAVE DU DANUBE

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RESUME

L'exploitation des esturgeons dans la partie yougoslave du Danube est une activité traditionnelle. On présente l'évolution des captures de 3 espèces, *Huso huso, Acipenser gueldenstaedti* et *Acipenser stellatus* avant et après la construction des 2 barrages de Djerdap en 1969 et en 1983. Les captures moyennes annuelles ne sont plus que 6,5 tonnes ces dernières années contre 11,5 avant la construction du premier barrage et 20 tonnes durant la période séparant la construction des 2 barrages. *Huso huso* et *Acipenser gueldenstaedti* représentent la quasi totalité des captures tout au long de la période, par contre la part relative de *Huso huso* a augmenté ces dernières années. Malgré la forte réduction du domaine exploitable et la faible augmentation de l'estimation de l'effort de pêche, les rendements, exprimés en tonnage moyen annuel par kilomètre de fleuve, ont considérablement augmenté. Les barrages en empêchant les migrations, concentrent les poissons qui sont ainsi plus facilement capturables.

Mots clés : *Acipenseridae*, statistiques de capture, barrage.

ABSTRACT

The sturgeon exploitation in Yugoslavian part of Danube is a traditional one. This paper presents the evolution of catches of the three following species: *Huso huso, Acipenser gueldenstaedti* and *Acipenser stellatus* before and after building the 2 Djerdap dams in 1969 and 1983. Actually the mean yearly catches are only 6,5 tons instead of 11,5 tons before the first damming and 20 tons during the period contained between the two dammings. Most of the catches are due to *Huso huso* and *Acipenser gueldenstaedti*; the relative part of the former increased the last years. In spite of the reduction of the exploited area and of the slight increase of the estimated catching effort, the yields (mean yearly weight per kilometer of river) have considerably increased. Dams, by stopping migrations concentrate the fish which become more easily catchable.

Key-words : *Acipenseridae*, statistics of catches, dam.

INTRODUCTION

Une partie du cours du Danube marque la frontière entre la Yougoslavie et la Roumanie. Sur une portion de celui-ci, les yougoslaves pêchent traditionnellement des esturgeons, et particulièrement trois grands migrateurs : *Huso huso, Acipenser gueldenstaedti* et *Acipenser stellatus*. Depuis quelques années, l'hydrologie de cette portion du Danube a été fortement modifiée par la construction de deux barrages hydroélectriques éloignés de quelques 60 kilomètres.

L'objet de cette courte note est de discuter l'évolution des captures des 3 espèces sus-citées avant et après la construction de ces 2 barrages.

MATERIEL ET METHODES

L'ampleur des zones de pêche a constamment diminué suite à la construction des barrages de Djerdap 1 en 1969 puis de Djerdap 2 en 1983 ; ils sont aussi appelés barrages des Portes de Fer (fig. 1). Les longueurs de fleuve exploitables sont indiquées dans le tableau 1. Elles correspondent aux longueurs de fleuve en amont du point kilométrique 845, c'est à dire à sa distance depuis l'embouchure.

Tableau 1 - Evolution des longueurs de fleuve exploitable par les pêcheurs yougoslaves

Période	Longueur (km)
< 1969	99
1970 - 1983	81
1984 - 1987	18

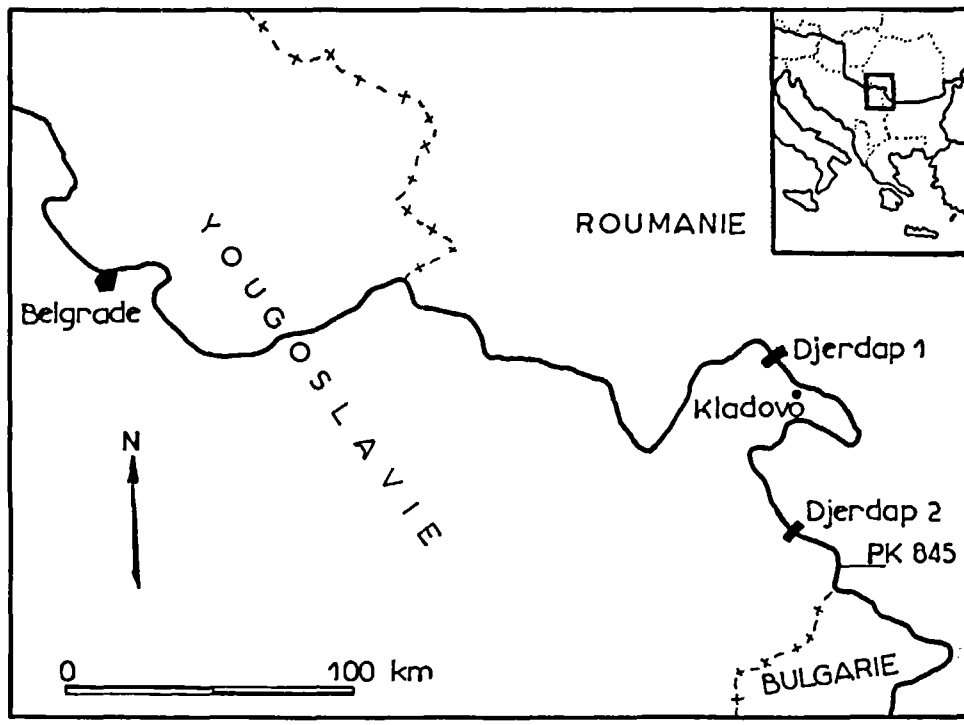


Figure 1- Le Danube Yougoslave à proximité des Portes de Fer.

Techniques de pêches

Traditionnellement, 2 types d'engin étaient utilisés : le filet et la ligne d'hameçons. Dans le second cas, pour que les hameçons soient pêchant, ils sont maintenus verticalement à proximité du fond grâce à un petit flotteur attaché à l'hameçon. Le fonctionnement des barrages entraîne de telles fluctuations de niveau d'eau et de vitesse de courant qu'il n'est plus guère possible d'utiliser les filets.

Recueil des données

Grâce aux déclarations des pêcheurs commerciaux, on peut connaître les quantités capturées par espèce tout au long de l'année ainsi que le nombre d'engins utilisés et ceci pour chaque lieu de débarquement. Les quantités représentent le poids total des animaux, gonades comprises. Toutes ces données sont disponibles au Ribarsko Gazdinstvo "Djerdap" pour les années 1958 à 1987 incluses.

RESULTATS

Quantités débarquées

Pour chacune des trois espèces, le tableau 2 indique les captures annuelles et leur importance relative. Ces pêches sont saisonnières.

Tableau 2 - Captures annuelles (tonnes) de trois principales espèces d'esturgeons, *Huso huso*, *Acipenser gueldenstaedti* et *Acipenser stellatus*.

Année	ESPECES					
	Huso huso		*A. gueldenstaedti*		*A. stellatus*	
	Poids	%	Poids	%	Poids	%
1958	5,4	68,2	2,3	28,6	0,2	3,2
1959	8,8	83,5	1,5	15,0	0,1	1,5
1960	3,4	49,1	3,3	47,2	0,2	3,7
1961	6,3	64,6	3,2	32,7	0,2	2,7
1962	3,8	35,3	6,6	60,3	0,5	4,4
1963	3,4	43,1	4,1	52,9	0,3	4,0
1964	2,7	39,5	3,7	54,5	0,4	6,0
1965	8,3	61,7	4,7	34,7	0,5	3,6
1966	6,7	54,4	5,1	41,6	0,5	4,0
1967	4,3	55,5	3,3	41,5	0,2	3,0
1968	7,6	59,7	4,9	38,2	0,3	2,1
1969	14,7	55,2	11,0	41,4	0,9	3,4
1970	9,8	52,0	8,7	46,2	0,3	1,8
1971	7,6	37,7	11,9	58,7	0,7	3,6
1972	11,2	53,4	9,2	44,1	0,5	2,5
1973	10,9	50,2	10,3	47,6	0,5	2,2
1974	9,7	46,8	10,5	50,2	0,6	3,0
1975	20,7	53,6	16,9	43,9	0,9	2,5
1976	9,7	58,2	6,6	39,8	0,3	2,0
1977	8,8	42,3	11,1	53,5	0,8	4,2
1978	8,9	46,4	9,8	51,1	0,5	2,5
1979	8,4	53,5	6,9	44,7	0,4	2,4
1980	9,4	51,9	8,4	45,9	0,4	2,2
1981	6,4	46,1	7,1	51,0	0,4	2,9
1982	15,2	76,9	4,2	21,4	0,3	1,7
1983	14,9	83,4	2,7	15,3	0,2	1,3
1984	5,3	63,7	2,8	34,1	0,2	2,2
1985	3,9	70,4	1,5	27,8	0,1	1,8
1986	3,9	74,9	1,2	22,8	0,1	2,3
1987	4,5	66,7	2,0	30,3	0,2	3,0

Sur une période de 14 ans, on constate une concentration des captures durant les 3 derniers mois de l'année (Tableau 3).

Comparaison des captures par période

Afin de pouvoir comparer entre elles les captures selon les 3 périodes (Tableau 1), on a calculé les quantités moyennes annuelles qui ont été ensuite rapportées au kilomètre de fleuve exploitable (Tableau 4).

Tableau 3 - Evolution des captures durant les 3 derniers mois de l'année (% du total annuel en poids)

Année	*Huso huso*	*A. gueldenstaedti*
1974	20,7	11,9
1975	24,4	28,3
1976	26,9	44,9
1977	24,1	37,9
1978	33,0	35,2
1979	12,8	32,3
1980	15,4	20,0
1981	18,7	13,8
1982	20,6	43,4
1983	19,7	31,0
1984	25,3	39,3
1985	61,6	62,8
1986	62,4	61,9
1987	59,2	58,8
Moyenne	30,3	37,2

Tableau 4 - Comparaison des captures moyennes annuelles

Périodes	*Huso huso*			*A. gueldenstaedti*			*A. stellatus*		
	Poids (kg)	%	Poids kg/km	Poids (kg)	%	Poids kg/km	Poids (kg)	%	Poids kg/km
1958-1969	6306	56	63,7	4488	40	45,4	374	4	3,8
1970-1983	10847	54	134	8899	44	109,9	494	2	6,1
1984-1987	4427	68	245,8	1913	30	106,2	148	2	8,3

Tableau 5 - Evolution du nombre d'engins de pêche

	1958-1969	1970-1983	1984-1987
Nombre de lieu de débarquement	15	9	2
Nombre de lignes d'hameçons	176	154	56
Nombre moyen de lignes par lieu de débarquement	11,7	17,1	28
Nombre moyen de lignes /km de fleuve exploitable	1,8	1,9	3,1

Evolution du nombre d'engins de pêche

Ce nombre a diminué avec la zone exploitable (Tableau 5). En supposant que leur utilisation soit restée constante durant les 3 périodes, on peut avoir une image de l'effort de pêche en rapportant leur nombre à la longueur de fleuve exploitable (Tableau 5).

DISCUSSION

Tout au long de cette période de trente ans, *Huso huso* et *Acipenser gueldenstaedti* constituent l'essentiel des captures, l'importance relative de *Acipenser stellatus* (voisine de 3 % des captures totales) restant à peu près constante (Tableau 1).

En l'absence de tout barrage, les captures moyennes annuelles sont d'environ 11 tonnes. La construction de Djerdap 1, tout en réduisant la zone exploitable, entraîne presque un doublement des captures annuelles moyennes (Tableau 4) à effort de pêche identique (Tableau 5). Ceci est la conséquence de l'obstacle aux migrations dues au barrage entraînant une plus grande concentration des poissons en aval ce qui les a rendus plus facilement capturables.

Cette situation s'est encore aggravée avec le second barrage. En effet, par rapport à la première période, l'estimation de l'effort de pêche a augmenté de moins de 50 % (Tableau 5), la zone exploitable a elle été diminuée par 5 et en dépit de cela les captures moyennes n'ont été que divisé par 2. L'ensemble de ces remarques sont traduites par la forte augmentation du rendement des captures au kilomètre de fleuve (Tableau 4).

On constate une augmentation de la part relative de *Huso huso* dans les captures (Tableau 4) et un décalage des captures en fin d'année (Tableau 3).

CONCLUSION

Dans le cadre de cette note, les fluctuations des captures d'esturgeons dans la partie yougoslave du Danube ont été attribuées pour l'essentiel à la construction des 2 barrages de Djerdap. Ceci suppose que les populations auxquelles ces poissons appartiennent n'aient subies aucune modification, ce qui est vraissemblablement irréaliste car elles sont également exploitées dans toute la partie aval du fleuve. Ainsi une analyse globale est-elle nécessaire sur la base d'une coopération internationale. C'est à cette seule condition qu'une dégradation accentuée des populations d'esturgeons du Danube pourra être évitée ; faute de quoi on peut craindre le pire jusqu'à la disparition de certaines espèces comme ce fut le cas de *Acipenser nudiventris* dont le dernier exemplaire mentionné date de 1954.

P. Williot, Ed. ACIPENSER, CEMAGREF Publ., 1991,

STATUT DES DIFFERENTES ESPECES D'ESTURGEONS DANS LE DANUBE ROUMAIN : PROBLEMES LIES A LEUR MAINTENANCE

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RESUME

On constate une baisse rapide des captures d'esturgeons dans le Danube, de presque 1 000 tonnes par an capturées en Roumanie durant les années 30, on ne pêche plus que quelques dizaines de tonnes par an dans tous les pays du Danube inférieur. Parmi les six espèces d'esturgeons signalées dans le Danube, *Acipenser sturio* et *A. nudiventris* sont devenus si rares que leur présence est maintenant douteuse. Les représentants de *Huso huso*, *A. gueldenstaeddti*, *A. stellatus* et *A. ruthenus* sont non seulement plus rares, mais aussi plus jeunes, comparativement aux décennies passées.

les causes de la baisse de captures d'esturgeons sont multiples :- la réduction de l'espace vital par les barrages, qui ont réduit aussi le nombre de frayères pour les esturgeons ;- les travaux pour la navigation et spécialement les dragueuses ont une influence défavorable sur les frayères restantes ;- l'intensification de la pêche aux filets à aloses utilisés au printemps dans le fleuve, contribue au prélèvement d'esturgeons immatures, tandis que celui des madragues pour les poissons de petite taille du littoral a une influence négative sur les jeunes esturgeons ;- l'introduction générale des chaluts à sprats dans la partie nord-ouest de la Mer Noire semble avoir une influence similaire et les chaluts de fond détruisent la faune benthique. L'eutrophisation de la partie peu profonde du nord-ouest de la Mer Noire, zone de nurserie pour les esturgeons, contribue à la réduction de la teneur en oxygène et à la formation d'hydrogène sulfuré près du fond.

En plus des mesures pour la maintenance d'esturgeons dans le Danube, la Roumanie est en train de construire dans le Delta du Danube une pisciculture à esturgeons.

Mots clés : *Acipenseridae*, statistique de capture, Danube

ABSTRACT

Sturgeon catch have decrease in the Danube, from almost 1 000 metric tons yearly landed only in Romania in the 1930s, all the riparian countries catches are reduced now, in the lower Danube, to some tens metric tons yearly. Out of the six sturgeon species described in the lower Danube, *Acipenser sturio* and *A. nudiventris* became so rare, that their presence is now doubtful. *Huso huso*, *A. gueldenstaedti*, *A. stellatus* and *A. ruthenus* are not only more rare but also younger than in the past decenniums.

There are many reasons of sturgeons catches decrease:- the reduction of vital space by the dams, that caused also the diminution of the number of migratory spawning places ;- the navigation works and especially the dredging ones have a detrimental influence on the remaining spawning places ;- the intensification of the use of Black Sea shad entangling nets contribute during spring to the catch of sturgeons, while the seaside small fish pounds nets, catch also joung sturgeons ;- the general introduction of sprat trawls in the north-west Black Sea seems to have the same influence and the botom-trawl are detrimental also to the benthos. The eutrophication of the north-west shallow Black Sea waters, a sturgeon feeding area, contribute to the reduction of the oxygen content and to the formation of hydrogen sulfide.

Besides other measures to the Danube sturgeons maintenance, Romania is preparing the construction of one sturgeon farm.

Key-words : *Acipenseridae*, statistics of catches, Danube

INTRODUCTION

La baisse rapide des captures d'esturgeons dans le Danube conduit à présenter cette note qui tente de faire la synthèse sur la biologie et l'état des stocks de ces poissons en Roumanie.

Données historiques

Le Danube est un des plus importants fleuves à esturgeons. La vieille colonie d'Histria (fig. 1), située près du delta du Danube, exporta durant près d'un millènaire des esturgeons sous forme de poisson salé vers le sud de l'Europe (Giurescu, 1964). Au moyen-Age le port de Kilia, au nord du delta, exportait des esturgeons vers la Pologne, tandis que ceux pêchés entre Braila et Calarasi étaient vendus en Transsylvanie (Manolescu, 1955).

Les esturgeons dépassaient les portes de Fer (fig. 1). L'huiron, *Huso huso*, atteignait la Bavière (Schrank, 1692). Au début du siècle, les captures en Roumanie dépassaient 1 000 tonnes par an.

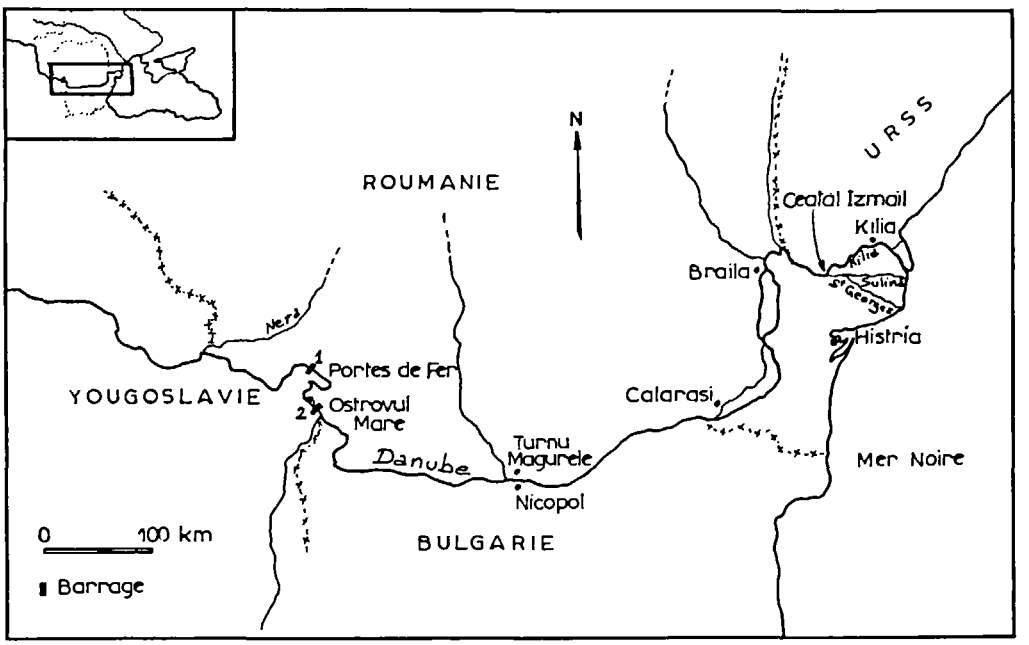


Figure 1- Cours du Danube depuis son embouchure jusqu'aux Portes de Fer.

Le Danube roumain

Le Danube est le deuxième fleuve d'Europe : sa longueur atteint 2 857 km, son bassin 817 000 km^2, son débit moyen environ 6 500m^3/s.

En Roumanie, le Danube a un parcours de 1 075 km qui peut être divisé en 3 secteurs (fig. 1): - le secteur supérieur, d'une longueur de 203 km, depuis l'embouchure de la rivière Nera (km 1 075) jusqu'à Ostrovul Mare (km 872). ;

- le secteur moyen (792 km) depuis Ostrovul Mare jusqu'à Ceastal Izmail (km 80) où commence le delta ;
- et le delta (80 km) depuis Ceatal Izmail jusqu'à la Mer Noire (fig. 1).

Deux barrages coupent le cours du fleuve :

- le premier, appelé Porte de Fer I dans le secteur supérieur au km 942,5 ;
- le second, appelé Porte de Fer II à la limite des secteurs supérieur et moyen près d'Ostrovul Mare.

Un troisième barrage est en construction à Turnu Magurele-Nicopol (km 581). Le fleuve doit être dragué régulièrement, en particulier dans le delta, pour les besoins

de la navigation. Le bras Saint-Georges du delta, autrefois très riche en poissons, a été rectifié.

BIOLOGIE DES ESTURGEONS DU DANUBE

Ce rappel synthétique des résultats obtenus sur la biologie des esturgeons du Danube est basé sur de nombreux travaux, en particulier ceux d'Antipa (1905, 1909, 1916 et 1933), de Bacalbasa-Dobrovici (1965 et 1985), de Banarescu (1964), de Leonte (1959 et 1963), de Leonte et al. (1965) et de Manea (1966, 1977 et 1980).

Six espèces d'esturgeons ont été signalées dans le Danube. Quatre d'entre elles, *Huso huso, Acipenser gueldenstaedti, A. stellatus* et *A. sturio* sont migrateurs. *A. sturio* et *A. nudiventris* sont devenus si rares que leur présence dans le Danube inférieur est maintenant douteuse.

Huso huso (huiron, ou morun)

Huso huso hiverne en Mer Noire entre 50 et 100 m de profondeur. Il entre dans le Danube en février-mars, lorsque la température de l'eau atteint 4 à 5 °C. Les mâles pénètrent les premiers, suivis une dizaine de jours plus tard par les femelles. Une autre migration, moins bien définie, a lieu en septembre-octobre. Cette migration d'automne n'a pas été observée en 1988 dans le bras St Georges où se produisent pourtant, habituellement, les plus importants passages.

La reproduction a lieu principalement dans les premiers 400 km en avril et mai, à des intervalles pour un poisson donné de plusieurs années. La maturité sexuelle est atteinte à 12-14 ans pour les mâles et 14-16 ans pour les femelles.

En Mer Noire l'huiron adulte se nourrit surtout de poisson ; dans le Danube il ne s'alimente pas. Les jeunes de première année mangent des *Gammaridae* et des *Mysidacae* ; ceux de deuxième année des mollusques, crevettes et petits poissons.

Il atteint 142 cm pour 18 kg à 6 ans et 299 cm pour 197 kg à 33 ans.

Acipenser gueldenstaedti (esturgeon du Danube, nisetru)

L'hivernage a lieu en Mer Noire, vers 60-70 cm de profondeur, plus près de la côte lorsque l'hiver est moins dur. Certains spécialistes pensent qu'il existe une sous-espèce sédentaire dans le fleuve.

L'esturgeon du Danube pénètre en eau douce en avril-mai, quand la température atteint 8-11 °C, et redescend vers la mer à l'automne. Il existe aussi une migration anadrome d'août à octobre.

Les lieux et époques de reproduction sont semblables à ceux de *Huso huso*. La maturité sexuelle est atteinte entre 8 et 12 ans pour les mâles et 13 à 15 ans pour les femelles. Il s'écoule plusieurs années entre deux participations à la fraye. A 7 ans, l'espèce atteint 92 cm et 4,2 kg ; à 21 ans, 155 cm et 17,9 kg. Son alimentation est à base de faune benthique.

Acipenser stellatus (esturgeon étoilé, pastruga)

L'hivernage a lieu en Mer Noire vers 80-100 m. L'esturgeon étoilé entre dans le fleuve après le passage des glaces, de mars à mai, pour une température de l'eau de 7 à 11 °C. En septembre-octobre a lieu une petite migration anadrome.

La reproduction a lieu au printemps, d'avril à juin, à des intervalles de plusieurs années pour un poisson donné. La maturité sexuelle est atteinte à 5 ans pour les mâles, 7 ans pour les femelles. A 5 ans l'esturgeon étoilé a entre 65 et 82,5 cm pour des poids de 1,7 à 2,4 kg ; à 15 ans, entre 129 et 160 cm pour des poids de 10,2 à 17,5 kg.

Acipenser ruthenus (sterlet, cega)

Le sterlet hiverne dans les fosses du Danube et remonte le fleuve vers les

frayères en mars-avril. La reproduction a lieu dans les zones à fort courant, vers 8-10 m de profondeur, au cours des mois d'avril et mai. Les adultes se reproduisent à des intervalles de 1 ou 2 ans, à partir de 3-4 ans pour les mâles et 4-5 pour les femelles.

L'espèce craint les eaux troubles et peu oxygénées. Si la saturation en oxygène s'abaisse en dessous de 92 %, le sterlet s'éloigne.

Il atteint 26 cm à 1 an, 35 cm à 2 ans, 52 cm à 6 ans, et 59 cm à 11 ans (Jankovic, 1956).

Les alevins d'esturgeons

Des pêches expérimentales dans les années 50-60 ont permis de capturer en mai, juin et juillet des alevins d'huiron de 50 à 150 mm, et des alevins d'esturgeon du Danube et d'esturgeon étoilé de 50 à 110 mm, sur des fonds de 5 à 10 m. Les zones de densité maximale sont le bras St Georges dans le delta, et, dans le Danube moyen, les km 169-186, 234-404 et 772-779. Les alevins commencent à apparaître en eaux saumâtres en juillet. Récemment, des pêches au chalut de fond à mailles de 5 mm dans le delta ont permis d'estimer la densité totale d'alevins des 3 espèces citées de 30 à 40 individus à l'hectare.

L'EVOLUTION DE LA PECHE DES ESTURGEONS DANS LE DANUBE

L'évolution des prises d'esturgeons du Danube en Roumanie et dans les autres pays du Danube moyen et inférieur est indiquée dans les tableaux 1 et 2.

Les captures relativement réduites des années 1920-1926 s'expliquent par le manque de matériaux destinés à la confection des engins de pêche, conséquence de la première guerre mondiale. La production s'est élevée ensuite jusqu'à plus de 1 000 tonnes en 1939 pour tomber à une centaine de tonnes pendant la première guerre mondiale. Après une faible remontée dans les années 50, les captures ont baissé progressivement et n'atteignent plus maintenant que quelques dizaines de tonnes.

L'espèce principale est *Huso huso* qui représente souvent plus de 90 % des captures. La réduction des prises affecte toutes les espèces.

PROBLEMES D'AMENAGEMENT

La réglementation de la pêche

Il est remarquable de constater que la réduction très importante des stocks d'esturgeons du Danube s'est produite malgré la protection réglementaire très ancienne. La première loi sur la pêche a été publiée en Roumanie en 1896. Elle a été modifiée plusieurs fois, la dernière modification datant de 1974 (Legea, 1974). De nombreuses mesures protègent les esturgeons : interdiction de la pêche aux hameçons non appâtés ; longueurs minimales (de la pointe du museau à la base de la caudale en cm) imposées.

170 cm pour	*Huso huso*,
140 cm pour	*A. gueldenstaedti*,
100 cm pour	*A. stellatus*,
40 cm pour	*A. ruthenus*,

Les autres pays ont adopté à peu près les mêmes mesures.

Ces mesures sont incontestablement bonnes, mais semblent insuffisantes si on compare les longueurs minimales avec les tailles maturité sexuelle qui sont du même ordre. Cela signifie que les esturgeons n'ont guère le temps de se reproduire avant d'être soumis à la pêche. Cependant il est probable que les causes principales du déclin des stocks viennent d'ailleurs, étant donné les faibles quantités capturées.

Tableau 1 - Les captures d'esturgeons (tonne) en Roumanie (statistique de pêcheries de l'état : 1920-1978, Manea 1980 ; 1979-1986, FAO Yearbooks).

Année	*Huso huso*	*A. gueldenstaedti*	*A. stellatus*	*A. ruthenus*	Total
	1	2	3	4	5
1920	162,2	171,3	89,8	32,0	455,3
1921	220,3	179,9	77,9	98,5	576,4
1922	143,5	148,6	41,0	32,2	365,2
1923	70,2	128,8	52,5	32,3	283,8
1924	285,1	148,9	33,8	49,1	516,9
1925	224,3	192,2	33,6	28,3	478,5
1926	232,6	124,5	32,5	26,9	416,5
1927	413,9	217,0	72,2	66,5	769,6
1928	415,9	195,9	46,7	65,7	724,2
1929/30	303,5	137,7	46,9	29,6	518,0
1930/31	437,6	154,0	69,8	20,5	681,9
1931/32	575,3	190,2	125,5	25,8	916,8
1932/33	532,6	198,4	141,5	31,7	904,1
1933/34	450,3	187,9	61,0	24,2	723,4
1934/35	551,6	211,8	134,2	34,4	932,0
1935/36	438,3	175,0	86,3	22,5	722,2
1936/37	476,6	162,5	134,4	18,4	791,8
1937/38	373,0	155,7	91,4	24,2	644,3
1938/39	288,1	148,6	82,2	14,4	533,2
1939/40	920,2	126,1	73,6	24,3	1144,2
1940/41	222,9	71,8	36,2	10,7	338,8
1941/42	56,3	37,8	28,0	4,5	126,6
1942/43	70,5	29,1	24,5	6,4	130,5
1943/44	74,1	34,9	34,7	11,7	155,3
1944 (1.04 31.12)	51,2	37,0	22,1	5,0	115,2
1945	169,7	43,0	22,7	1,2	236,4
1946	162,4	32,2	11,0	1,4	207,3
1947	126,9	23,1	3,6	1,8	155,4
1948	219,2	58,4	25,9	26,0	329,6
1949	209,7	46,8	7,3	20,2	279,9
1950	276,2	34,3	12,6	70,2	393,3
1951	293,3	33,6	10,5	35,4	372,7
1952	186,8	19,6	2,7	30,5	239,5
1954	172,4	6,0	2,0	15,7	196,3
1955	287,0	9,0	4,0	7,0	307,0
1956	198,0	4,0	3,0	10,0	215,0
1957	140,0	2,0	-	7,0	149,0
1958	182,0	2,0	2,0	17,0	203,0
1959	193,0	5,0	2,0	11,0	211,0
1960	274,2	5,1	2,6	10,3	292,2
1961	267,2	6,7	2,4	8,8	285,0
1962	208,1	2,8	1,4	8,8	221,0
1963	193,0	2,9	0,9	8,4	204,2
1964	272,1	3,9	1,3	7,5	284,7
1965	277,4	3,9	1,3	6,3	288,5
1966	204,0	10,0	2,0	4,0	220,0
1967	188,0	3,0	2,0	6,0	199,0

Suite Tableau 1

	1	2	3	4	5
1968	201,0	4,0	1,0	4,0	210,0
1969	145,5	3,3	1,6	4,8	155,2
1970	143,7	4,2	1,7	4,0	153,7
1971	195,7	4,1	3,2	3,9	206,9
1972	208,0	3,4	2,7	9,1	223,2
1973	205,4	5,9	3,5	7,1	221,9
1974	160,8	5,6	2,2	5,3	173,9
1975	121,7	3,7	1,6	2,4	129,4
1976	114,7	11,7	7,5	5,8	139,6
1977	114,8	9,5	5,6	7,3	137,2
1978	73,1	9,6	5,6	5,7	93,9

Tableau 2 - Les captures d'esturgeons (tonne) dans le Danube par l' URSS, la Roumanie, la Bulgarie et la Yougoslavie de 1979 à 1986 (données de la Commission mixte des Pêches pour le Danube).

Année	*Huso huso*	*A. gueldenstaedti*	*A. stellatus*	*A. ruthenus*	Total
	1	2	3	4	5
			URSS		
1979	5,1	4,9	-	-	10,0
1980	3,8	4,2	-	-	8,0
1981	4,2	10,1	-	-	14,3
1982	5,4	10,7	-	-	16,1
1983	5,0	9,9	0,1	-	15,0
1985	2,5	5,4	0,3	-	8,2
1986	4,0	4,6	-	-	8,6
			Roumanie		
1979	1 + 2 + 3 =	64,7		2,9	67,6
1980	id.	42,0		1,0	43,0
1981	id.	69,3		2,2	71,5
1982	id.	61,1		13,4	74,5
1983	id.	18,9		3,6	22,5
1985	id.	12,6		1,9	14,5
1986	id.	15,3		2,8	18,1
			Bulgarie		
1979	-	14,2	-	5,2	19,4
1980	-	10,9	-	5,3	16,2
1981	2,8	8,6	3,2	6,0	20,6
1982	3,9	10,3	1,6	8,1	23,9
1983	5,9	3,5	0,2	4,8	14,4
1985	2,6	3,7	0,1	2,8	9,2
1986	2,1	2,6	0,3	2,0	7,0

Suite tableau 2

	1	2	3	4	5
			Yougoslavie		
1979	8,4	6,4	0,9	46,4*	62,1
1980	9,4	8,8	-	28,1	46,3
1981	6,4	7,1	-		13,6
1982	15,3	4,2	-	70,0	89,5
1983	14,9	2,7	-	63,1	80,7
1985	3,9	1,6	-	42,1	47,6
1986	3,9	1,3	<0,1	63,1	68,3

* Y comprises toutes les captures yougoslaves du secteur pannonien.

Les modifications du milieu naturel

Depuis la dernière guerre, de nombreuses transformations ont bouleversées les milieux du bassin du Danube.

Les dragueuses et autres travaux pour la navigation se sont considérablement développés. Dans le bras de Sulina (delta), la pêche des esturgeons n'est plus possible, tandis que les bras de Kilia et de St Georges ont été rectifiés. Les prélèvements de sable et gravier sont importants. Les endiguements ont réduit les zones d'inondation. Enfin les barrages ont réduit l'espace vital et fait disparaître d'importantes frayères pour les esturgeons migrateurs.
L'agriculture intensive a apporté de grandes quantités de phosphore et d'azote provoquant un excessif développement des algues et finalement une réduction de la teneur en oxygène. Les mêmes phénomènes sont constatés dans tous les autres fleuves tributaires de la Mer Noire (Bashkin, 1989), de sorte que le taux d'hydrogène sulfuré de cette mer continue à augmenter (Aizatulin et al., 1989), réduisant l'espace vital disponible pour la croissance des jeunes esturgeons. Des pluies intenses provoquent parfois des crues catastrophiques pour les poissons par les déchets d'origine agricole ou minière qu'elles entraînent dans les cours d'eau.

Comme conséquence de toutes ces pollutions, on peut signaler par exemple la disparition presque totale des larves de *Palingenia sp.*, nourriture préférée du sterlet.

CONCLUSION

Les recherches scientifiques faites jusqu'à présent n'apportent pas de réponses claires au problème de la survie des esturgeons du Danube. Il faudrait développer les études sur les principaux points suivants :

- les migrations,
- l'emplacement des frayères,
- l'influence de la pêche sur les esturgeons, en particulier l'impact des filets à aloses utilisés au printemps dans le fleuve, celui des madragues pour les petits poissons du littoral deltaïque en été, et celui des chaluts à sprats dans la partie nord-ouest de la Mer Noire.

Un problème capital et malheureusement très difficile à résoudre est celui de l'eutrophisation des fleuves tributaires de la Mer Noire, car les mesures à adopter dépassent les possibilités actuelles des pays du bassin hydrographique de la Mer Noire. Le maintien des stocks d'esturgeons dans des conditions naturelles de plus en plus

mauvaises paraît donc très compromis. C'est pourquoi la Roumanie est en train de construire dans le delta du Danube un établissement d'acipensériculture.

Enfin les recherches devraient être coordonnées par la Commission mixte pour l'application de la convention sur la pêche dans les eaux du Danube, convention signée à Bucarest en 1958.

L'Association internationale pour les recherches sur le Danube, filiale de la Société internationale de limnologie, pourrait également contribuer aux travaux.

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AMELIORATION DE LA METHODE DE DETERMINATION DE L'AGE DE L'ESTURGEON COMMUN *Acipenser sturio* ET PREMIERES APPLICATIONS.

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RESUME

Les auteurs ont mis au point une technique originale de prélèvement *in vivo* de tranches de rayon pectoral, moins traumatisante que le classique prélèvement *in toto* du rayon. Ils précisent les outils utilisables, la faisabilité ainsi que les modalités de cette opération. Parmi cinq méthodes d'observation ils ont déterminés celles qui semblent les plus efficaces pour effectuer des lectures d'âges. A titre d'application une relation taille-âge de l'*Acipenser sturio* en Gironde est calculée.

Mots clés : *Acipenser sturio*, détermination de l'âge, squelettochronologie, rayon pectoral, croissance, méthodologie.

ABSTRACT

The aim of this paper is to propose an original technic for fin ray slices removal on alive sturgeon, less traumatic than the classic *in toto* pectoral fin ray removal. The authors specify usable tools and the modalities of the operation. Among five methods they determine those which seem to be the bests for age determination. A length-age relationship is given for *Acipenser sturio* in Gironde.

Key-words : *Acipenser sturio*, age determination, skeletochronology, pectoral fin ray, growth, methodology.

INTRODUCTION

Ce travail concerne la population ouest européenne de l'esturgeon commun *Acipenser sturio* L. 1758 qui fréquente l'estuaire de la Gironde.

Après quelques années d'une intense exploitation et suite à une chute des captures cette espèce est actuellement protégée sur tout le territoire français. Sa distribution en mer couvre une large étendue (Figure 1), du golfe de Gascogne aux côtes danoises en passant par les îles britanniques, mais les effectifs sont devenus très faibles.

Afin de connaître la structure de cette population et pour suivre sa dynamique nous devons pouvoir déterminer l'âge des individus. Ce stock étant relativement fragile nous avons tenté de mettre au point une méthode qui soit aussi peu traumatisante que possible pour les animaux et qui nous permette de les relâcher dans de bonnes conditions.

C'est ainsi que nous avons choisi de travailler sur des structures superficielles (rayon pectoral), afin de minimiser les lésions dues aux prélèvements.

Les esturgeons étant dépourvus d'écailles, les biologistes utilisent depuis longtemps pour déterminer l'âge individuel de ces poissons des techniques de squelettochronologie basées sur l'observation des rayons de nageoires (Meunier, 1988).

On utilise de façon préférentielle les coupes transversales du premier rayon pectoral, à partir d'un prélèvement complet de ce rayon : (Classen, 1944; Cuerrier, 1951; Probst et Cooper, 1954; Magnin, 1959, 1962; Priegel et Wirth, 1971, 1975; Kohlhorst et al, 1980; Wilson, 1987).

Un certain nombre d'auteurs prélèvent ces rayons sur des esturgeons relâchés vivants, à des fins d'estimations de stocks par marquage et recapture : (Cuerrier et Roussow, 1951; Priegel, 1973; Cochnauer et al, 1985; Wilson, 1987) mais seul Kohlhorst

(1979), sur *Acipenser transmontanus* évalue les conséquences de ces opérations sur la survie des animaux.

Certains auteurs se sont penchés sur la méthodologie des lectures d'âge, ceci afin que les résultats de chercheurs différents puissent être comparés. Pour ce type d'estimations d'écarts, seul la méthode du "pourcentage d'accord de lecture" est généralement utilisée (Kohlhorst et al, 1980 ; Wilson, 1987).

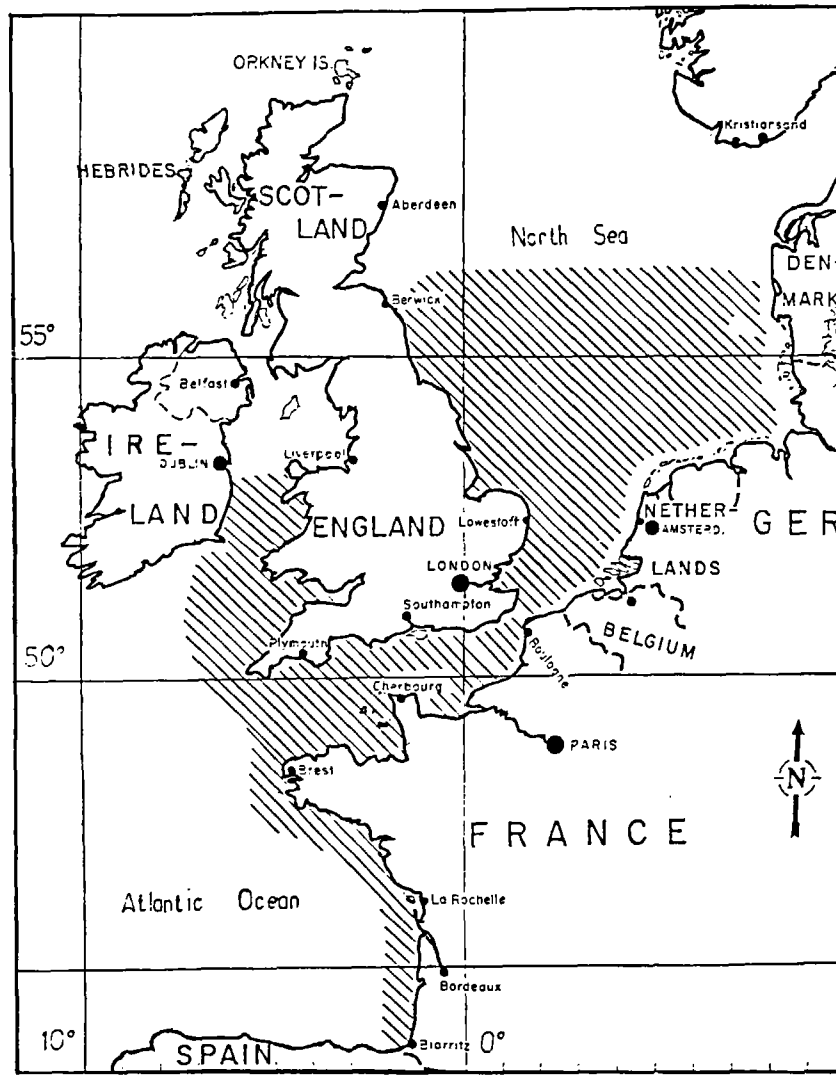


Figure 1 - Aire de répartition actuelle de l'esturgeon commun *Acipenser sturio* en Europe du nord-ouest.

Les objectifs de cette étude consistent à :

- mettre au point, une méthode de prélèvement "*in vivo*" de tranche de rayon pectoral, moins traumatisante que le classique prélèvement *in toto* du rayon ;
- préciser les outils utilisables, la faisabilité et les modalités de cette opération ;

- déterminer parmi cinq méthodes d'observations celles qui nous semblent les plus efficace et les éventuelles différences de lectures entre les méthodes ;
- et à titre d'application, calculer la relation taille-âge actuelle de la population d'*Acipenser sturio* de l'estuaire de la Gironde.

MATERIEL ET METHODES

Les esturgeons, *Acipenser sturio*, ont été capturés avec des filets tramails, dans l'estuaire de la Gironde (Figure 2), entre le 1/02/88 et le 31/08/88. Au cours de ces pêches ils ont été mesurés (Longueur totale : L) au cm près à l'aide d'un ichthyomètre. Leur poids est évalué avec une précision de ± 200 g et une marque individuelle spécialement conçue est placée à l'avant de la nageoire dorsale (Castelnaud, 1988).

Réalisation des prélèvements

Ils sont effectués sur des animaux choisis selon un échantillonnage stratifié par classes de taille (un animal prélevé pour 3 animaux capturés). Cet échantillonnage compense l'impossibilité technique de manipuler tous les esturgeons capturés dans un laps de temps raisonnable et nous permet de considérer notre échantillon comme représentatif de la population. De plus il permettra ultérieurement, par comparaison des taux de recaptures et des croissances observées entre animaux prélevés et non prélevés, d'estimer la mortalité consécutive au prélèvement (Kohlhorst, 1979), une éventuelle différence de croissance et la régénération des tissus osseux prélevés.

Plutôt que de prélever le rayon pectoral en entier (Kohlhorst, 1979 ; Kohlhorst et al, 1980 ; Wilson, 1987), nous prélevons comme Cochnauer et al (1985), une tranche, directement sur le premier rayon pectoral gauche de l'animal (le choix du rayon gauche est arbitraire) (Figure 3).

Deux outils ont été testés :
- une scie à main à denture fine (Peugeot rubis 12d) ;
- un petit disque à tronçonner, monté sur une perceuse électrique autonome, tournant à basse vitesse (150 rotations par minutes).

On effectue deux traits de scie espacés d'environ 2 mm, puis on détache la section ainsi obtenue par une incision au scalpel entre les premier et second rayons pectoraux. Pour diminuer les risques d'infection, la plaie est badigeonnée avec une solution antiseptique de vétédine (Polyvinyl-pyrrolidone iode 10%).

Dans les deux cas, le premier trait de coupe est réalisée à environ 15 mm de l'articulation de la nageoire (Kohlhorst et al, 1980). Les poissons sont ensuite relâchés.

Le traitement des prélèvements

Au laboratoire, après un rinçage à l'eau, on mesure l'épaisseur des tranches de rayons avec une précision de 10 µm, afin de pouvoir comparer la précision des deux techniques de prélèvements. Elles sont ensuite poncées manuellement avec du papier abrasif imperméable de grain 400 puis 800, tout en contrôlant fréquemment au microscope binoculaire (grossissement x20) la lisibilité et la netteté de la tranche dans une goutte d'éthanol. On arrête le ponçage lorsque la netteté est satisfaisante, c'est à dire lorsqu'on juge que le contraste permet une bonne discrimination entre zones opaques et stries hyalines. On note alors l'épaisseur correspondante.

Avant d'effectuer les lectures on laisse sécher la pièce osseuse quelques semaines. Ce séchage améliore le contraste entre les marques. Wilson (1987) procède de la même façon avec les rayons entiers.

Chaque tranche subit cinq traitements réversibles successifs, avec quatre produits différents utilisés classiquement pour éclaircir les préparations (Tableau 1). Pour l'un de ces produits (glycérol) on utilise deux protocoles différents. On réalise en premiers les traitements les plus facilement réversibles pour finir par ceux qui le sont moins. A chaque traitement correspond une observation réalisée de façon aléatoire en privilégiant l'anonymat des échantillons.

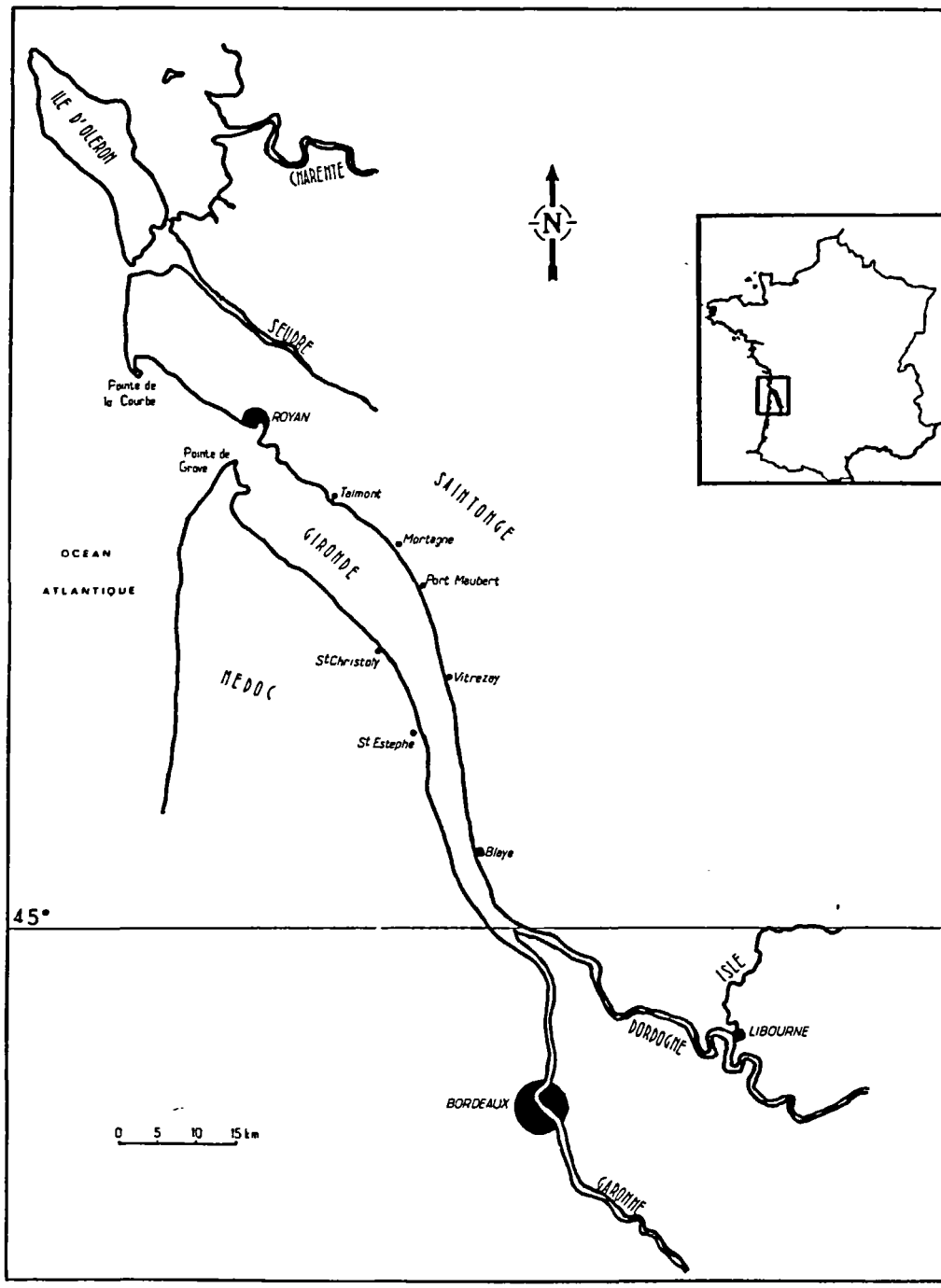


Figure 2 - L'estuaire de la Gironde, au sud-ouest de la France, constitue notre aire d'étude.

Tableau 1 - Traitements et observations réalisées sur les tranches de rayons pectoraux.

1	- bain de 5 min dans de l'éthanol absolu ; - observation dans une goutte d'éthanol absolu (éclaircissant) ; - séchage sur du papier absorbant.
2	- bain de 5 min dans du xylène pur (diméthyl benzène) ; - observation dans une goutte de xylène pur (éclaircissant) ; - séchage sur du papier absorbant.
3	- imprégnation de 15 min dans du glycérol ; - observation dans une goutte de glycérol (nappant) ; - rinçage 5 min dans de l'éthanol absolu ; - trempage 55 min dans de l'éthanol absolu ; - séchage sur du papier absorbant.
4	- imprégnation de 60 min dans du glycérol ; - observation dans une goutte de glycérol ; - rinçage 5 min dans de l'éthanol absolu ; - trempage 55 min dans de l'éthanol absolu ; - séchage sur du papier absorbant.
5	- déshydratation de 5 min dans de l'éthanol absolu ; - bain de 5 min dans du créosote de hêtre (éclaircissant) ; - observation dans une goutte de créosote de hêtre ; - rinçage 5 min dans du benzène ; - trempage 60 min dans du benzène ; - séchage sur du papier absorbant.

Les observations sont effectuées au microscope binoculaire en lumière transmise avec les grossissements x20 et x50.

Détermination de l'âge, comparaisons et reproductibilité des lectures

On observe sur les tranches polies des stries hyalines (claires en lumière transmise) alternant avec des zones opaques (sombres en lumière transmise), généralement plus larges (Figures 4 et 5).

Les stries hyalines (périodes de croissance ralentie) présentent probablement un tissus osseux plus minéralisé que les zones opaques de croissance active (Meunier, 1982). Nous avons retenu l'hypothèse selon laquelle une strie hyaline est déposée chaque hiver.

A la surface des tranches de rayons, les zones où l'on effectue les dénombrements et les principaux critères permettant le comptage des stries hyalines, ont été déterminés par des lectures comparées. Il s'agit notamment de l'espacement régulier des stries et de leur continuité.

A l'issue des cinq séries de lectures, les coupes sont classées sommairement en quatre catégories. Ainsi en fonction de la netteté des contrastes entre stries hyalines et zones opaques et de l'allure générale, on les qualifiera d'illisibles, peu nettes, nettes et très nettes. On note le nombre de stries hyalines (Sh) obtenu pour chacune d'entre-elles.

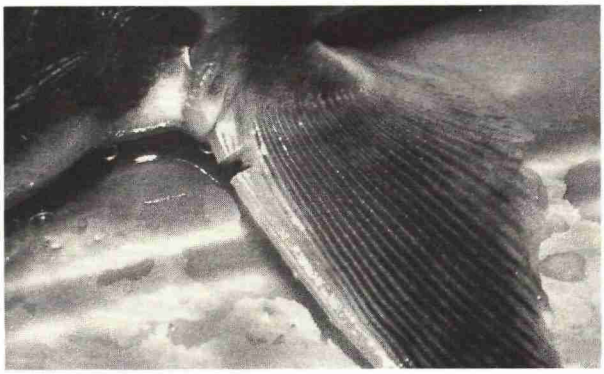
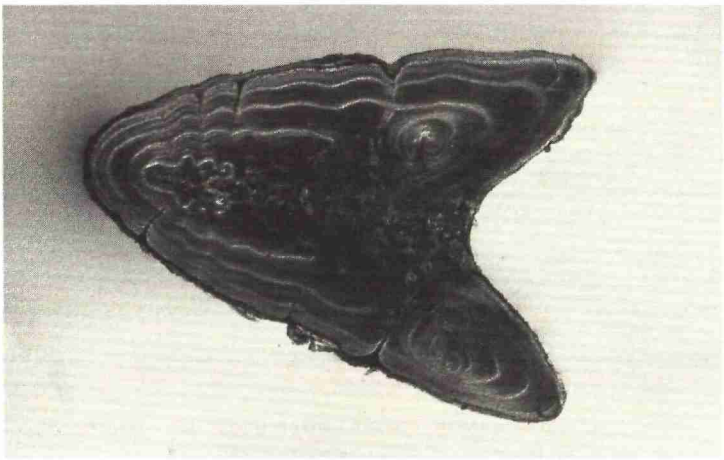


Figure 3 - Trace d'un prélèvement réalisé avec une scie à main sur un très jeune *Acipenser sturio* (Classe 1).

Figure 4 - Coupe de rayon pectoral d' A. *sturio*. 21/06/88 Estuaire de la Gironde. L 111 cm, 5 Sh.



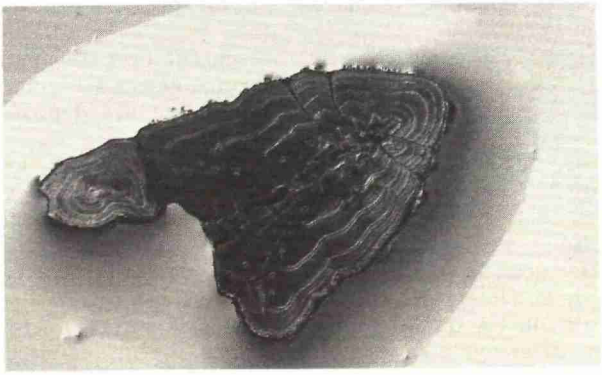


Figure 5 - Coupe de rayon pectoral d'A. *sturio*. 21/06/88. Estuaire de la Gironde. L 105 cm, 5 Sh.

On suppose que chaque Sh indique une période hivernale, puisque correspondant à un ralentissement de croissance, ce qui est plausible pour cette espèce vivant en zone tempérée, au moins pour les individus n'ayant pas encore atteint leur maturité sexuelle (des marques de fraie ont été signalées sur *Acipenser oxyrhynchus* par Smith, 1985). Ceci est corroboré par les données de croissance provenant des animaux marqués et recapturés (Castelnaud et al, 1991) et par le suivi d'une cohorte durant sa première année (Rochard, travaux en cours).
Pour la dénomination de l'âge nous avons retenu la clef suivante :

Aucune strie hyaline	Classe 0
Une strie hyaline	Classe 1.
Deux stries hyalines	Classe 2.
Trois stries hyalines	Classe 3.
etc...	

La période de reproduction d' *Acipenser sturio* se situe vraisemblablement en mai-juin (Magnin, 1962). Ainsi un animal de classe 1 capturé au printemps aura environ 1 an et pris en début d'hiver il aura environ 1 an et demi.

Pour définir l'influence des traitements sur la détermination de l'âge, les cinq séries de lectures sont comparées, grâce à un test statistique. Comme chaque tranche de rayon est observée après chacun des cinq traitements, les cinq séries de lectures qui en résultent sont dépendantes, pour comparer ces séries dont les données ne sont pas distribuées selon une loi normale, nous avons retenu le test des rangs de Friedman (Siegel et Castellan, 1956).

Selon la méthode développée par Beamish et Fournier (1981) on note pour chaque tranche (j) et après chaque traitement (i) l'âge moyen estimé (X_{ij}). A l'issue des 5 observations, on peut calculer l'âge moyen estimé (X_j) du poisson dont provient la tranche.

$$X_j = 1/5 \, \Sigma_i \, X_{ij}$$

On calcule ensuite l'erreur moyenne sur l'âge de ce $_j$ème poisson : EMA_j (APE_j de Beamish et Fournier, 1981).

$$EMA_j = 1/5 \, \Sigma_i \, |X_{ij} - X_j| / X_j$$

Puis pour l'ensemble des tranches (N) on établit le pourcentage d'erreur moyenne sur l'âge : EMA (APE de Beamish et Fournier, 1981).

$$EMA = 1/N \; \Sigma_j [\; 1/5 \, \Sigma_i (|X_{ij} - X_j| / X_j) \;] \; 100$$

Selon Beamish et Fournier (1981) et Chang (1982), cette méthode, en tenant compte de l'âge des individus, fournit une meilleure estimation de la reproductibilité des lectures d'âge que le calcul habituel du pourcentage d'accord de lecture.

Etablissement de la courbe taille-âge et de l'équation de croissance

A partir de l'âge moyen estimé on établit des classes :

$$(\text{Classe i} : \; (i-0,5) \leq X_j < (i+0,5))$$

et on détermine la longueur moyenne des animaux pour chacune de ces classes. On peut alors tracer une courbe taille-âge, et dresser un graphe de Ford-Walford pour établir l'équation de croissance de notre échantillon.

$$L_{(t+1)} = k \, L_t + Y$$

avec k coefficient de croissance de Ford.

On calcule alors les paramètres de l'équation de croissance de Von Bertalanffy (Equation 9.9 de Ricker, 1980).

$$L_t = L_\infty \, (1 - e^{-K \, (t-to)})$$

Avec K, le coefficient de croissance de Brody, L_∞, la taille théorique à laquelle l'animal a une croissance nulle et t_0, la date théorique à laquelle l'animal a une taille nulle, selon l'équation de croissance précédente.

$$K = - \text{Log } k$$
$$L_\infty = Y/(1-k)$$
$$t_0 = t + (1/K \text{ log } e) \log ((L_\infty - L_t)/L_\infty)$$

Méthode retenue pour la validation

66 prélèvements ont été fait de février à août 1988. On espère recapturer au moins 30 de ces animaux durant les campagnes 1989 et 1990.

Sur ces animaux on aura ainsi un prélèvement en t et un prélèvement en $t+x$ mois, x comprenant au minimum une période hivernale. Par la suite, en comparant les lectures on pourra déterminer ce qui constitue réellement un *annuli* pour ces animaux.

RESULTATS

Les prélèvements sur les rayons pectoraux ont été effectués sur 66 esturgeons mesurant de 77 cm à 179 cm et pesant de 2,9 kg à 34 kg, choisis selon un échantillonnage stratifié par classe de taille (1 individu prélevé pour 3 individus capturés) parmi les animaux capturés.

Sur l'ensemble des tranches de rayon, 32 (soit 48 %) ont été réalisées avec la scie à main et 34 (soit 52 %) l'ont été avec le disque à tronçonner. Le tableau 2 résume les contraintes et les caractéristiques de chaque méthode.

Tableau 2- Avantages et contraintes des deux outils utilisés pour les prélèvements.

Méthodes	scie à main	Disque à tronçonner
Besoin en personnel	1 personne	2 personnes
Risque pour le personnel	nul	non nul
Durée du prélèvement	1 à 2 min.	1 à 3 min.
Hémorragie au niveau de la coupe	pas ou peu	pas ou peu
Qualité des coupes	bonne	moyenne
Autonomie de l'appareil	illimitée	3 heures
Epaisseur détruite par deux traits de coupe	2400 µm	1600 µm
Sensibilité aux mouvements du bateau	faible	forte

Les prélèvements réalisés

a. *Caractéristiques physiques des tranches de rayon*

Avant ponçage, l'épaisseur des tranches faites à la scie à main est distribuée suivant une loi normale avec pour moyenne 2547 µm (± 327 µm à 5%). Celles faites avec le disque à tronçonner, suivent également une loi normale, mais de moyenne 2355 µm (± 355 µm).

Après ponçage, l'épaisseur des tranches se distribue selon une répartition normale de moyenne 434 µm (± 29 µm à 5%) et d'écart type 120 µm (valeurs extrêmes : Min. 170 µm, Max. 990 µm.).

b. *Qualité des tranches de rayon et des lectures*

Parmi les 330 observations réalisées (66 tranches * 5 observations), 5 cas, correspondant à 3 tranches, sont considérés comme illisibles (1.5%).

Sur l'ensemble des tranches : 13 sont très nettes (20%), 34 sont nettes (52%), 16 sont peu nettes (24%) et 3 sont illisibles pour au moins une observation (4%). Illisible signifie ici que le dénombrement des stries hyalines, donc la détermination de l'âge de l'animal, est impossible.

Selon les observations les résultats sont les suivants :

N° d'observation	Pourcentages de tranches lisibles
1	98,5%
2	100,0%
3	98,5%
4	97,0%
5	98,0%

On remarque que même dans le cas le moins satisfaisant, (observation 4, Glycérol 60 min) les résultats sont tout à fait acceptables.

c. *Lectures d'âge, comparaisons, reproductibilité*

Le tableau 3 donne la distribution des lectures d'âge, en fréquence absolue, pour chacune des séries d'observations et pour la série des "ages moyens estimés" (moyenne des lectures).

Tableau 3 - Distribution des lectures d'âges réalisées pour les cinq séries d'observations et la moyenne des lectures. (Age : en classe), (N : effectifs), (Moyenne : longueur moyenne), (Min. : longueur minimum), (Max. : longueur maximum).

	Lectures à l'éthanol				Lectures au xylène				
Age	N	Moyenne	Dév. st.	Min.	Max.	N	Moyenne	Dév. st.	Min.	Max.
3	7	83,1	5,9	77	94	9	83,9	5,3	77	94
4	9	97,3	10	85	114	5	99,6	7,3	92	111
5	34	111	5,9	100	127	37	110,4	6,6	91	127
6	5	114,8	9,7	104	125	4	114,5	15,3	100	133
7	3	122,7	14	109	137	5	122,4	10,1	109	137
8	3	141	8,5	133	150	2	145	7,1	140	150
9	0					0				
10	1	154		154	154	0				
11	0					0				
12	0					0				
13	0					0				
14	0					1	154		154	154
15	1	154		154	154	1	154		154	154
16	1	179		179	179	1	179		179	179
17	0					1	179		179	179
18	0					0				
19	1	179		179	179	0				

Tableau 3- (Suite)

		Lecture à la glycérine 15'					Lecture à la glycérine 60'			
Age	N	Moyenne	Dév. st.	Min.	Max.	N	Moyenne	Dév. st.	Min.	Max.
3	10	85	6,1	77	95	9	83,9	5,3	77	94
4	3	97,3	4,7	92	101	6	100,7	7,4	92	113
5	35	110,3	6,6	91	127	34	110,1	6,6	91	127
6	5	110,2	9,7	100	122	4	11,7	10,5	100	122
7	5	120,2	11,8	109	137	5	126,4	12,6	109	140
8	3	141	8,5	133	150	2	141,5	12	133	150
9	0					0				
10	0					0				
11	0					1	179		179	179
12	1			179	179	0				
13	1	154		154	154	1	154		154	154
14	0					0				
15	1	154		154	154	1	154		154	154
16	0					0				
17	1	179		179	179	1	179		179	179
18	0					0				
19	0					0				

		Lecture au créosote					Moyenne des lectures			
Age	N	Moyenne	Dév. st.	Min.	Max.	N	Moyenne	Dév. st.	Min.	Max.
3	9	83,9	5,3	77	94	10	85	6,1	77	95
4	3	95	4	91	99	7	103	9,7	91	114
5	36	110,5	6,6	92	127	36	110,3	6,1	100	127
6	6	112,3	12	100	133	4	121,7	2,5	119	125
7	6	125,6	11,4	109	140	4	129,7	14,1	109	140
8	1	150		150	150	1	150		150	150
9	0									
10	0									
11	1	154		154	154					
12	0					1	154		154	154
13	1	154		154	154					
14	0					2	166,5	17,7	154	179
15	1	179		179	179					
16	0					1	179		179	179
17	1	179		179	179					
18	0									
19	0									

Nous avons utilisé le test de Friedman, d'analyse de la variance sur les rangs pour des séries dépendantes (logiciel P.C.S.M. *) pour comparer les résultats des cinq séries d'observations, en prenant comme hypothèse nulle, qu'il n'y a pas de différence

* P.C.S.M., 1987. Programme conversationnel de statistiques pour les sciences et le marketing. Deltasoft. Grenoble.

significative entre elles. Cette hypothèse est confirmée (à 95%) : $p > 0,05$ ($p = 0,962$). Les cinq séries de lectures peuvent être considérées comme appartenant à la même population statistique.

En ce qui concerne la reproductibilité des lectures, dans 41 cas (62 %), il y a accord entre les cinq observations. A l'opposé, trois cas (4,5 %), présentent une EMA_j supérieure à 0,3.

L' EMA (en fait ici une mesure des écarts de lectures entre les séries d'observations) est de 6,83 %. On notera qu'une part importante de l'EMA (18 %), est imputable à deux animaux âgés de plus de 14 ans et présentant des EMA_j de 0,5 et 0,4.

Premiers éléments de la croissance

A partir du graphe de Walford présentée figure 6, on obtient pour les classes d'age 3 à 8, l'équation de Walford suivante :

$$L_{t+1} = 0.897\ L_t + 23.8$$

L'équation de croissance de Von Bertalanffy est donc :

$$L_t = 231\ (1 - e^{-0.109\ (t + 1.36)})$$

On peut alors tracer (pour les classes d'age retenues) la courbe de croissance actuelle des *Acipenser sturio* de la Gironde, figure 7.

DISCUSSION

Les tranches ont été réalisées sur des animaux issus d'un échantillonnage stratifié, sur l'ensemble des captures réalisées en zone estuarienne.

On peut donc considérer que l'échantillon retenu présente la même structure en taille et en âge, que le groupe d'esturgeons, présents et capturables, au même moment, dans l'estuaire de la Gironde. Cependant, sa composition limite nos interprétations aux animaux des classes d'age 3 à 8.

Les méthodes de coupes et la qualité des prélèvements

a. *La méthode de prélèvement retenue*

La technique la plus satisfaisante sur le terrain, est dans notre cas, le prélèvement direct avec une scie à main à denture fine. Son avantage principal tient à la sécurité du personnel quelles que soient les conditions météorologiques. Le prélèvement avec le disque à tronçonner est plus risqué, il a pour seul avantage de faire pour une même épaisseur de prélèvement une entaille systématiquement un peu plus petite, mais pas suffisamment cependant pour que l'on puisse se dispenser de la phase de ponçage.

Les coupes prélevées *in vivo* avec les outils testés ici n'auront jamais au départ la finesse et l'état de surface de coupes réalisées en laboratoire avec une tronçonneuse a disque diamanté. Pour atteindre cette qualité elles doivent être longuement poncées.

L'épaisseur des tranches de rayon au moment du prélèvement varie trop peu d'une méthode à l'autre pour que ce puisse être un critère déterminant dans le choix de l'outil et l'avantage pour l'animal (si même il existe), en terme de survie, semble faible.

Toutefois, les deux outils répondent à notre objectif principal à savoir : prélever une tranche de rayon *in vivo*, de façon moins traumatisante pour l'animal que l'ablation totale et compromettant moins ses chances de survie ultérieure. En effet Kohlhorst (1979), a montré pour *Acipenser transmontanus* que dans le cas d'une ablation totale du rayon, la mortalité pouvait atteindre 36% durant l'année suivant l'opération.

Quelque soit l'outil utilisé nous n'avons pas eu d'hémorragie, bien que nous fassions notre prélèvement à 15 mm de la base de la nageoire.

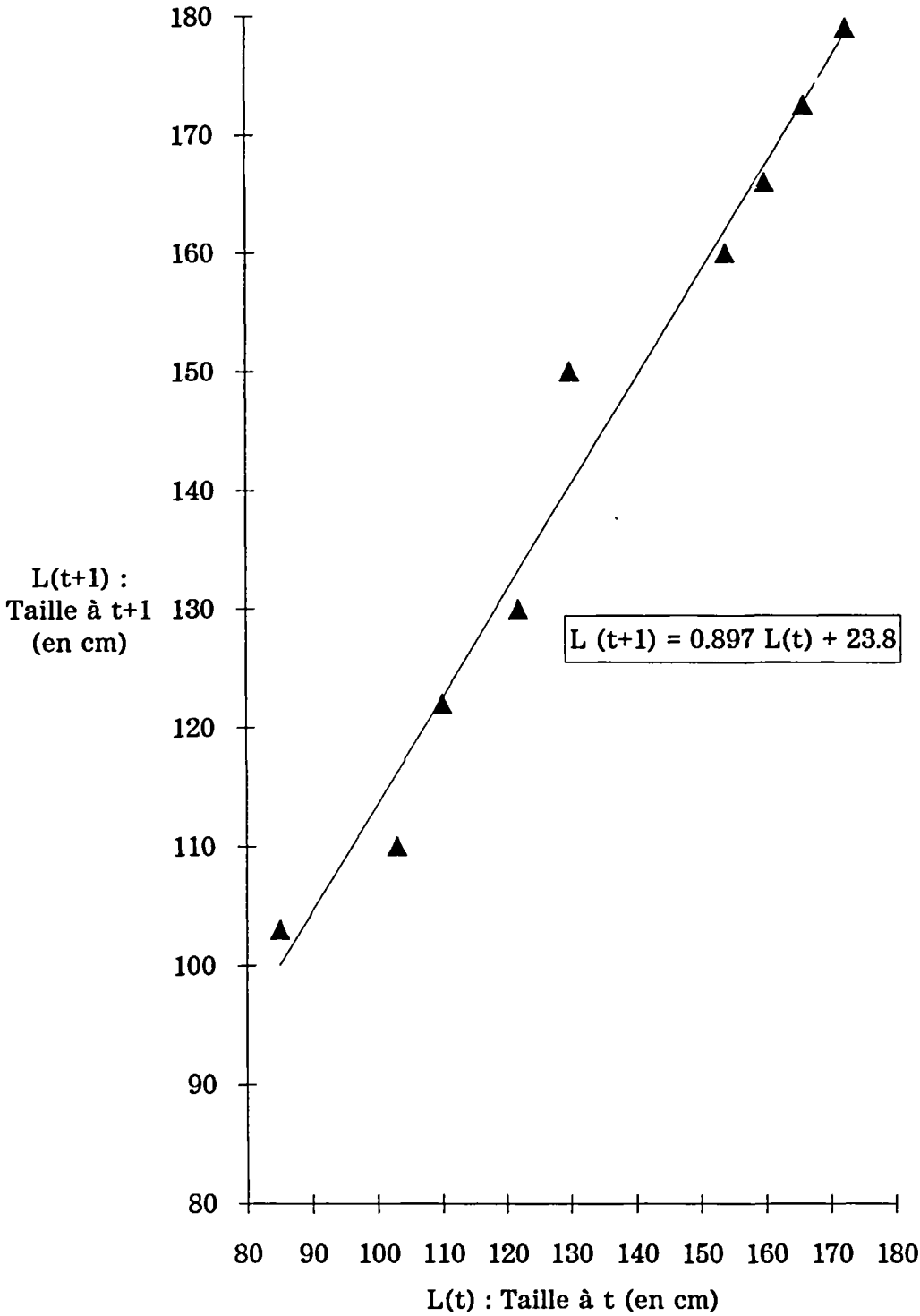


Figure 6 - Graphe de Walford. $L_{(t+1)}$ par rapport à L_t.

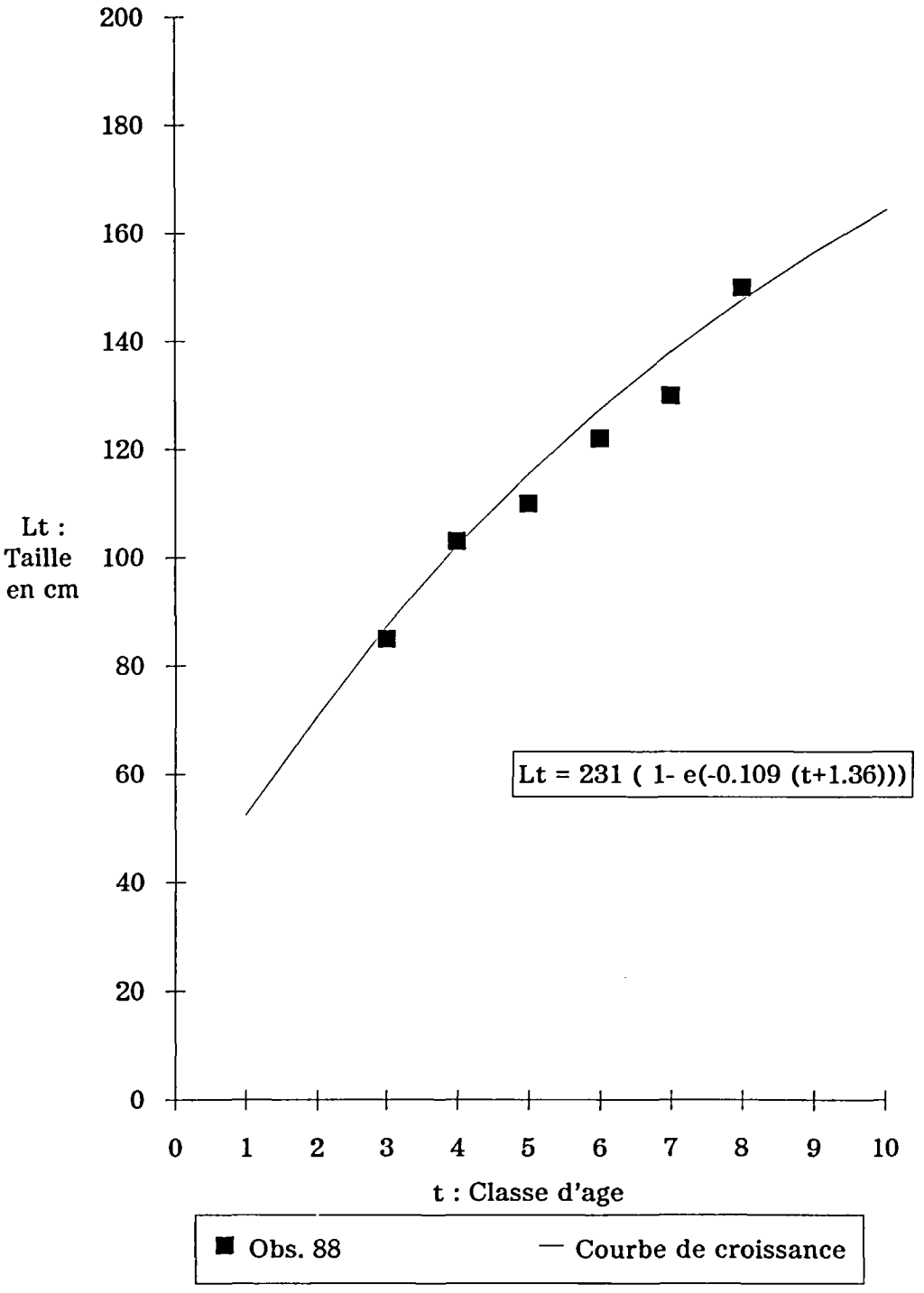


Figure 7 - Courbe de croissance Taille-Age, et observations réalisées en 1988 (Obs. 88).

Il semble que ce risque, qui existe lorsque le rayon est prélevé en entier (Threader, 1981), soit quasiment éliminé. C'est un des avantages commun aux deux outils testé dans ce travail.

Bien que nous n'ayons pas pu suivre la cicatrisation en milieu naturel, un suivi d'un an en pisciculture sur *Acipenser baeri* a montré une cicatrisation au bout de 1 à 3 mois, on peut supposer qu'elle s'opère de la même façon sur *Acipenser sturio*. Il conviendrait cependant d'effectuer un suivi de la régénération des tissus osseux prélevés.

b. *Influence de l'épaisseur sur les prélèvements*

Il faut noter que nous avons réalisé nos observations sur des coupes relativement épaisses. Sur les coupes les plus fines (épaisseur inférieure à 250 µm), le contraste entre les zones opaques et les stries hyalines est moins bon et comme le souligne Cuerrier (1951) sur un ensemble d'espèces de poissons, les pièces deviennent transparentes et moins faciles à lire ; à l'opposé, les coupes les plus épaisses (> 600 µm), trop opaques, sont difficiles à interpréter. Wilson (1987) arrive à des conclusions similaires sur *Acipenser fulvescens*.

Influence des préparations sur les comptages

Il n'y a pas de différences significatives entre les cinq séries de lectures correspondant aux cinq préparations. On notera cependant que certaines rendent les lectures plus aisées, en faisant mieux ressortir les zones opaques et les stries hyalines. Ce sont les préparations 2 (xylène) et 5 (créosote de hêtre). Le xylène (N°2) a notamment permis les comptages dans tous les cas. Pourtant le créosote et le xylène, bien qu'éclaircissants classiques en otolimétrie, sont peu cités dans les publications concernant les esturgeons, alors qu'alcool et glycérol couramment utilisés par les auteurs, se sont avérés bien moins satisfaisants dans nos travaux.

Si on considère deux préparations comme deux lecteurs différents, on peut mesurer les écarts de numération auxquels on doit s'attendre. On constate alors que ces écarts sont faibles mais qu'ils ont tendance à augmenter avec l'âge et ce, bien que les EMA_j intègrent l'âge dans leur formule. Plus les animaux sont âgés plus les lectures d'âge sont difficiles et plus ils présentent des EMA_j élevés.

Difficultés de lectures et d'interprètation

Les coupes de rayon pectoral présentent des problèmes de lecture qui leur sont propres, entre autres :

- manque de netteté à la périphérie ;
- perturbations dues à des centres d'ossifications secondaires (Magnin, 1959).

Cette seconde "anomalie", qui apparait sur des animaux assez âgés (> 10 ans) entraîne certaines difficultés de lectures.

Nous avons retenu l'hypothèse selon laquelle une Sh est déposée lors de chaque période hivernale. Cependant de nombreux auteurs (Beamish et Mac Farlane, 1983 ; Prouzet et Souplet, 1987 ; Meunier, 1988) ont montré qu'il est nécessaire de confirmer ce type d' observations par une validation directe de la méthode, actuellement en cours. On peut toutefois penser que l'acquisition de la maturité sexuelle et que les migrations de grande ampleur réalisées par certains animaux, peuvent amener à modifier sensiblement cette hypothèse.

Premiers éléments concernant la croissance

La classe 5 est prépondérante dans nôtre échantillon où les classes 0, 1 et 2 sont absentes (mal échantillonnées ou non représentés dans la zone de pêche). Au delà de la classe 8 la faible taille de l'échantillon rend les interprétations difficiles. C'est pourquoi la figure 7 (Courbe de croissance et observations réalisées en 1988) ne concerne que les classes 3 à 8 pour lesquelles l'équation de croissance a été calculée.

La partie gauche de cette courbe théorique de croissance semble être confirmée par les premiers résultats d'un suivi de cohorte (Rochard, travaux en cours).

La croissance relativement rapide des classes d'age 3 à 8 est confirmée par le suivi d'animaux marqués (Castelnaud et al, 1991).

CONCLUSION

La méthode de prélèvement

La technique de prélèvement de tranche de rayon *in vivo* que nous proposons a le grand avantage d'être nettement moins traumatisante pour les animaux que le classique prélèvement *in toto*. Elle semble pouvoir s'appliquer à d'autres espèces d'esturgeons et est réalisable dans tous les cas, même à bord de petites embarcations. Pour sa facilité nous préférons utiliser la scie à main à denture fine mais les deux outils testés répondent à notre objectif principal.

Les méthodes de lecture

Pour un même lecteur les différences de comptage d'une préparation à l'autre sont très faibles. On aura donc intérêt pour des lectures d'âge en routine à utiliser une technique simple, altérant peu les pièces osseuses. Nous recommandons les lectures en lumière transmise, avec des coupes d'environ 400 µm d'épaisseur. Le xylène s'avère facile d'emploi, mais sa forte toxicité nous fait lui préférer le créosote de hêtre.

Les techniques à la glycérine ou au créosote seront utilisées préférentiellement pour des observations plus fines (par exemple : détermination de la nature du dernier dépôt).

Enfin il faut rappeler que l'utilisation du xylène et du benzène (lors du rinçage du créosote) en raison de leur toxicité nécessite impérativement que l'on prenne les précautions d'usage.

Application à la croissance

D'après nos travaux la croissance que nous observons sur les animaux des classes 3 à 8 est relativement rapide, elle ne semble pas correspondre à l'image "classique" de l'esturgeon : animal à croissance lente. Dans les années à venir, ce paramètre essentiel de l'état d'un stock devrait être suivi de façon précise.

Depuis l'automne 1988 nous suivons une cohorte d'*Acipenser sturio*, qui devrait nous permettre, après validation de la méthode d'âgeage, pour toutes les classes d'âge, d'améliorer notre connaissance de la croissance de cette espèce et de mieux cerner la dynamique de cette population menacée.

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AGE DETERMINATION AND VALIDATION STUDIES OF WHITE STURGEON, *Acipenser transmontanus*, IN CALIFORNIA

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RESUME

Trois approches ont été utilisées pour développer à la fois la méthode la plus exacte et la plus reproductible pour déterminer l'âge de l'esturgeon blanc en Californie: 1) Analyse comparative sur six différentes structures calcifiées de poissons sauvages, pour déterminer les meilleures structures et méthodes de détermination de l'âge, 2) Vérification de l'âge selon la taille des animaux sauvages par une étude similaire sur des animaux en élevage, 3) Etude marquage-recapture d'animaux sauvages afin de déterminer les périodes et fréquences de formation des anneaux de croissance dans les structures calcifiées.

Au total, un échantillon de 230 structures calcifiées d'esturgeons (rayons de nageoires pectorales, opercules, clavicules, clithrum, écussons, et plaques nuchales), provenant de la Baie de San Francisco et du Delta, a été récolté pour des analyses comparatives. Plusieurs méthodes furent utilisées pour dégager un modèle de croissance (coupe fine, huile éclaircissante, brûlage, rayon x, et balayage au microscope électronique) et déterminer la technique la plus appropriée pour chaque structure. Bien que toutes les structures calcifiées aient possédé des informations de croissance/âge et révélé des comptages similaires de croissance pour les différentes structures calcifiées d'un même poisson, la section du rayon de la nageoire pectorale s'avère être constamment la plus facile à analyser et procure l'estimation la plus précise de l'âge. Toutes les structures, incluant le rayon pectoral, ont des limitations majeures.

Les esturgeons d'élevage, dont l'âge était connu, ont été élevés dans des conditions contrôlées des laboratoires d'aquaculture de U.C. Davis. L'oxytétracycline (OTC) a été utilisé pour marquer une partie des os de ces poissons pour déterminer la fréquence de dépôt des anneaux de croissance, et ces structures ont été analysées pour la croissance postérieure au marquage à l'OTC. Bien que les structures osseuses provenant de ces poissons aient montré une croissance, elles sont beaucoup plus difficile à analyser que celles des poissons sauvages et ne fournissent pas toujours de façon claire la trace normale du dépôt. Cela peut être dû à des facteurs tels que température, salinité et alimentation qui diffèrent considérablement des conditions naturelles.

Actuellement, nous avons plus de 419 recaptures signalées et plus de 113 structures osseuses provenant de 2187 esturgeons qui ont été mesurés, marqués, injectés avec de l'OTC et relâchés dans la Baie de San Francisco. Bien que la plupart des recaptures aient été faites dans la Baie de San Francisco et dans le système deltaïque ou leurs environs immédiats, 13 d'entre elles ont eu lieu dans les états d'Orégon et de Washington. Les parties du squelette des poissons recapturés ont fourni des informations sur la croissance et les dépôts de calcium pour la période comprise entre le marquage et la recapture. Toutes ont montré distinctement les traces de marquage à l'OTC.

De l'analyse, à plus d'un an d'intervalle, de 19 échantillons provenant de poissons sauvages, il apparaît que les anneaux de croissance sont annuels. Un plus grand nombre d'individu recapturé après une longue période de temps donnera une meilleure compréhension de ce phénomène. A partir de ces données, des courbes de croissance ont été construites selon la taille, l'âge et le sexe des animaux sauvages, et ces courbes ont été comparées à celles d'autres études. Elles ont seulement permis de vérifier partiellement les estimations d'âge, le choix de la technique de determination de l'âge la plus précise pour cette espèce nécessitera l'achèvement de nos études de validation.

Mots clés : *Acipenser transmontanus*, détermination de l'âge, squelettochronologie, méthodologie, croissance.

ABSTRACT

Three approaches were used to develop the most accurate and reproducible methods for ageing white sturgeon in California: A comparative analysis of six different calcified structures of wild fish to determine the best overall structure and method for age determination, laboratory grow-outs to verify wild-

stock determinations of age at size, and field mark-recapture studies to determine timing and frequency of growth ring formation in calcified structures.

A total of 230 samples of sturgeon calcified structures (pectoral fin rays, opercles, clavicles, cleithra, scutes, and medial nuchals) was collected from San Francisco Bay and Delta to be used for comparative analysis. Various methods were used to elucidate growth patterns (thin sectioning, oil clearing, staining, x-radiography, and scanning electron microscopy) to determine the most useful ageing technique for each structure. Although all calcified structures had growth/age information and revealed similar growth increment counts among different structures of the same fish, the pectoral fin ray sections were consistently the easiest to analyze and provided the most precise age estimates.Yet, all structures, including fin sections, had major limitations.

Laboratory grown sturgeon of known age were raised under controlled conditions at the U.C. Davis aquaculture laboratories. Oxytetracycline (OTC) was used to mark the bones on a portion of these fish to determine how often growth increments were deposited, and these structures were analyzed for growth subsequent to the OTC mark. Although the bony structures from these fish showed growth increments, they were much more difficult to analyze than wild fish structures and did not always provide apparent, timely depositional patterns. This may be due to factors such as temperature, salinity, and diet which differ significantly from wild conditions.

Currently, we have over 419 reported recaptures and over 113 recoveries of calcified structures from the 2187 sturgeon that were measured, tagged, injected with OTC and released into San Francisco Bay. Although most recaptures were taken in or about the San Francisco Bay and Delta system, 13 were recaptured from Oregon and Washington states. Skeletal components from the recaptured fish have provided information on growth and calcium deposition from the time of release to recapture. All showed distinct OTC markers.

From analysis of the 19 samples that were at large for over 1 year, it appears that the growth patterns are annual. A greater number of long-term returns will give a more complete understanding of this phenomenon. From these data, growth curves have been generated comparing size, age, and sex of field-collected specimens, and these curves have been compared to those of other studies. Although these studies have at least partially verified sturgeon age estimates, determination of the most accurate and suitable ageing technique for this species will not be available until we have completed our validation studies.

Key-words : *Acipenser transmontanus*, age determination, skeletochronology, methodology, growth.

INTRODUCTION

Over the past several years, much attention has centered on the white sturgeon (*Acipenser transmontanus*) in California, primarily due to the increasing popularity of its sportfishery and potential for cultivation. There is rising interest in the sportfishery because : 1) white sturgeon are renowned as the largest freshwater fish in North America with excellent fighting ability, 2) the quality of the flesh and roe is high, 3) the stocks of other popular gamefishes are decreasing and, 4) their life history characteristics may make them susceptible to overexploitation. Despite the popularity and economic importance of white sturgeon, information on their size, age composition, and rate of growth is not well documented. Yet, data on these features are essential for establishing adequate management of such a fishery.

Before the 1870's the white sturgeon resource in the San Francisco Bay was virtually untapped (Smith and Kato, 1979). A boom in the fishery came with a diminished east coast supply and an increased interest in sturgeon flesh and caviar (Skinner, 1962). This economically important fishery lasted less than 30 years (1875-1901). Landings from 1875 to 1892 averaged 227 tons annually, while between 1892 and 1901, they ranged from about 45 to 91 tons (Smith and Kato, 1979). In 1901, the State Legislature temporarily closed the fishery, claiming the white sturgeon to be on the verge of extinction. An unsuccessful reopening of the fishery was attempted between 1910 and 1917.

Following a 35 year prohibition of sturgeon fishing in the San Francisco Bay area, sport fishing was allowed again in 1954, and subsequently this popular fishery has increasing numbers of private and party boats fishing heavily for sturgeon. Catch rates generally declined from the late 1960's to 1970's (Kohlhorst et al., 1980),

but more recently harvests have increased dramatically in the mid-1980's. The 1954 reopening of the sport fishery also stimulated studies of sturgeon biology to provide information to manage the fishery. Pycha (1956) reported on white sturgeon age and growth estimates, size composition, migrations, and abundance. Chadwick (1959) and Miller (1972 a,b) estimated harvest rates and described migration. The food habits of young (Schreiber, 1962) and adult (McKechnie and Fenner, 1971) sturgeon were also described. Stevens and Miller (1970) and Kohlhorst (1976) estimated time and location of spawning. Kohlhorst et al. (1980) reported on sturgeon age and growth, but found considerable disagreement between individual estimates of age using transverse sections of marginal pectoral fin rays.

The ability to age fish accurately is essential to a complete understanding of the dynamics of fish populations. Recognition of annual growth patterns in bony structures requires the understanding of how these patterns are related to the annual growth cycles of fishes (Chilton and Beamish, 1982).

The most reliable means of estimating age and growth rates of fishes is by examination of calcified structures for growth patterns (Bagenal, 1974) and tag/mark-recapture studies for validation of age estimates (Beamish and McFarlane, 1983). Tagging and recapture of fish whose calcified structures are marked with oxytetracycline (OTC) offers a simple and conclusive method for verifying the timing of growth zone formation (Smith, 1984). Although OTC has been used to study human bone formation since the 1950's, its use as a marker in fishes was not suggested until 1962 (Weber and Ridgway, 1962). It has had successful and wide application in both field and laboratory studies since that time (i.e., Holden and Vince, 1973; Smith, 1984; Gruber and Stout, 1983; Cailliet et al., 1986).

The importance of age determination of sturgeons relative to fishery management is evidenced by the numerous and various methods used by different authors since to the early 1900's (Arnold, 1911; Kleer, 1916; Harkness, 1923; Hollzmayer, 1924; Bajkov, 1930; Derjavin, 1932; Greeley, 1937; Schneberger and Woodbury, 1944; Cuerrier, 1951; Sunde, 1961; Pycha, 1956; Roussow, 1957; Semakula and Larkin, 1968; Malm, 1979 and Kohlhorst et al., 1980). Various structures were used in these studies, including scutes (ganoid scales), pectoral fin rays, otoliths, opercles, and several other skeletal parts. Although the interpretation of ages based on thin sections of pectoral fin rays has been the method of choice, until recently this has not been subjected to comparative analysis of all possible structures to determine which methods are the best for any species (Brennan and Cailliet, 1989). In addition, most authors assumed that the growth increments found in skeletal structures were deposited annually, but few verified age estimates and none have attempted validation. Verification is defined as the repeatability of a numerical interpretation (analogous to precision) that may be independent of age, and validation is the determination of the true temporal meaning of a growth increment (analogous to accuracy) (Cailliet et al., 1986; Summerfelt and Hall, 1987; Brennan and Cailliet, 1989).

The purpose of this paper is to report on the three approaches we have used to verify and validate white sturgeon age and growth estimates. First, we evaluated various calcified structures of wild sturgeon to determine age and growth parameters more precisely (Brennan and Cailliet, 1989). Specific objectives of this study were : 1) to assess the usefulness of several calcified structures for determining the age of sturgeon ; 2) to develop techniques that would elucidate growth patterns in these structures making them more useful for age determination and ; 3) to compare age estimates from different structures to determine the most precise method for determining age and growth parameters. Second, we used captive, hatchery-reared sturgeon to verify wild sturgeon age estimates. Specific objectives of this study were : 1) to establish the characteristics and timing of the formation of early annuli ; 2) to assess the usefulness of OTC as an internal marker for sturgeon, and ; 3) to verify age estimates of wild fish by comparing growth patterns in calcified structures of hatchery-reared sturgeon to those of wild fish. Our third approach has been to use field-marked and recaptured sturgeon to validate age estimates. Specific objectives of this study were to : 1) develop accurate and reproducible criteria for ageing ; 2) assess field growth, and ; 3) determine periodicity of growth pattern formations in calcified structures of white sturgeon.

METHODS

Comparative analysis

White sturgeon were collected from the San Francisco Bay and Delta region of California between January 1984 and December 1985 for comparative analysis studies (Figure 1).

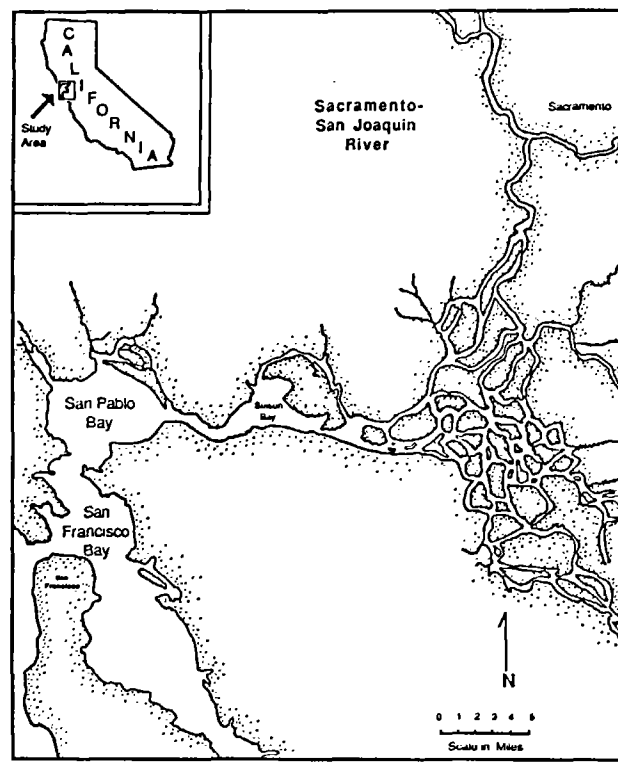


Figure 1 - Map of the study

Body weight (WT) to the nearest kilogram, total length (TL), and fork length (FL) to the nearest centimeter, and sex were recorded for most samples (Figure 2). From each fish we collected a variety of calcified structures including marginal pectoral fin rays, opercles, clavicles, cleithra, medial nuchals, and dorsal scutes, because they all had visible calcified depositional patterns (Figure 3). Pectoral fin rays were collected from 147 sturgeon to be used for age analysis. After removal of the anterior ray of the pectoral fin, the rays were dried, thin sectioned, and inspected for growth increment formation with the aid of a binocular dissecting microscope. In addition, five other structures from a subsample of 44 fish were selected for comparative age analysis from the range of size classes available. Initially, all structures from each fish were qualitatively assessed for quality (lack of chips, cracks or deformities) and readability (clarity of ring patterns). Subsequently, one of each type of structure (i.e. one scute, opercle, clavicle, cleithrum, medial nuchal) was selected from each fish for ageing.

All structures, other than pectoral fin ray sections, were viewed with the aid of a large magnifying glass using reflected light and a dark background. Occasionally, a low power binocular dissecting microscope and/or reflected light were required to aid in the assessment of illegible growth rings.

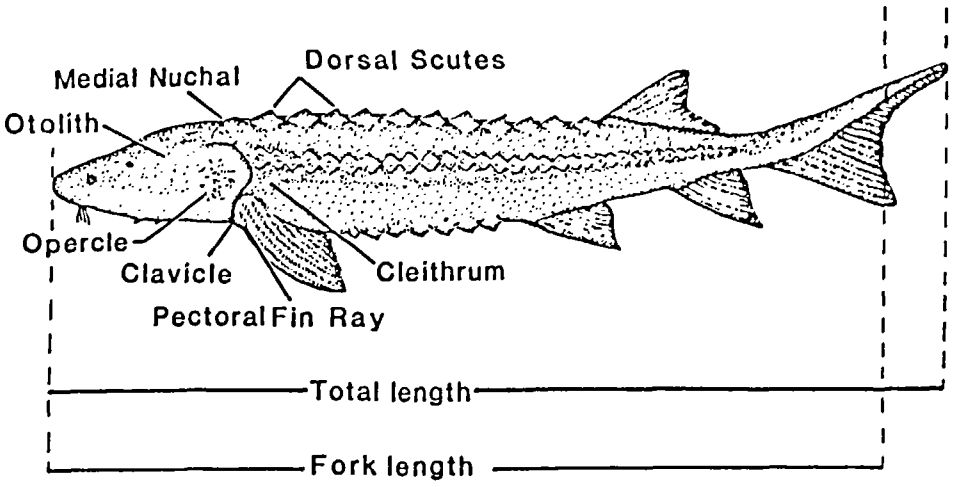


Figure 2 - White sturgeon illustration showing measurements of total length, fork length and location of calcified structures inspected for age determination.

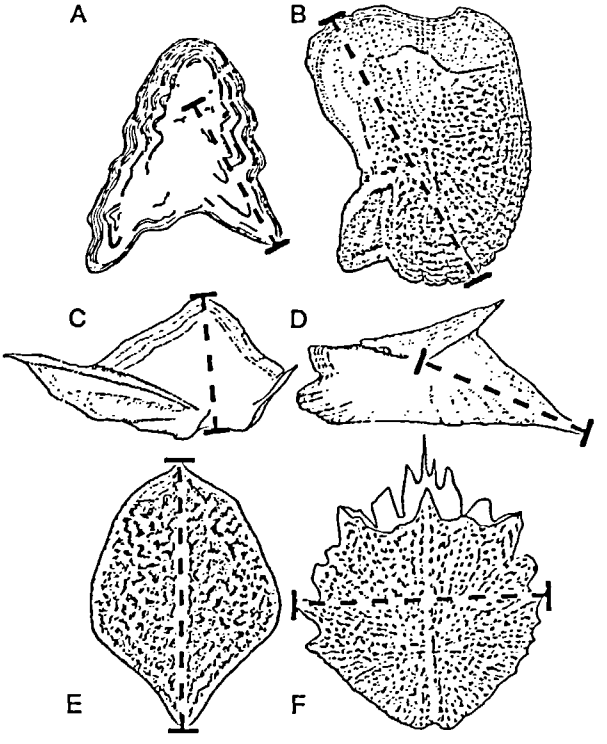


Figure 3 - Illustrations of calcified structures used for age determination. Lines mark axis of growth measured. A Pectoral fin ray section; B Opercle; C Cleithrum; D Clavicle; E Dorsal scute; F Medial nuchal (Taken from Brennan et cailliet 1989).

All structures selected for age analysis were read twice by the first author and once by at least one other reader to determine inter-reader variability (with the exception of dorsal scutes). In addition, single reader annulus counts from corresponding structures of the same fish were compared using a regression analysis in which the coefficient of determination, r^2, was used as a measure of strength of the straight-line relationship (Zar 1984). Corresponding structure annulus counts were also compared to pectoral fin section counts of the same fish as a standard to determine intra-reader variability. Each structure was then ranked, based on the cumulative percent agreement within + 3 annulus counts, to provide a relative index of interpretability. Sample preparation, elucidation techniques, and precision tests for each structure are described by Brennan (1988) and Brennan and Cailliet (1989).

Laboratory grow-outs

In 1984 and 1985, a total of 102 cultivated white sturgeon was collected from the University of California, Davis aquaculture facilities for analysis of early depositional patterns and verifying age estimates. Twenty fish from the 1983 year class were selected for grow-out studies and skeletal structures from an additional 87 cultivated sturgeon, from four year classes, were acquired from fish sacrificed for nutritional studies.

On 20 January 1984, twenty hatchery sturgeon were collected from the 1983 year class for grow-out studies; 10 fish were tagged with T-bar spaghetti tags and injected with OTC at a dosage of 25 mg/kg body weight, and 10 were tagged only. Body weight (gm), TL (cm), and FL (cm), were recorded for each fish. Fourteen fish were remeasured and retagged on 18 April 1984 due to the loss of tags, again resulting in 20 tagged fish. Oxytetracycline was injected into 10 of the 20 fish at a dosage of 25 mg/kg of body weight (Holden and Vince, 1973; Beamish et al., 1983; Smith, 1984; McFarlane and Beamish, 1987). The sturgeon were reared for up to 30 months in 4-meter, circular fiberglass tanks with a flow through water system. Water temperatures ranged from about 11°C to 20°C and fish were feed Silver Cup Trout Chow, a commercially available pelleted fish food, ad libitum from mechanical feeders. Body measurements and pectoral fin rays were collected from each fish when it was sacrificed. Fin rays were dried, thin sectioned, and inspected for growth increment formation with the aid of a binocular dissecting microscope.

Field mark-recapture

During the fall of 1984, 1985, and 1987 we participated in cooperative tagging efforts with the California Department of Fish and Game (CDFG) who were tagging sturgeon in San Pablo and Suisun Bays for estimates of abundance, harvest rate, survival rate and movements (Figure 1). Sturgeon were captured in a 20.3 cm stretched mesh trammel net, 366 m long and 6 m deep that was drifted from 0.25 to 2.25 h. Captured sturgeon were brought aboard the vessel, removed from the net, measured (TL), single tagged or double tagged, and released. Fish less than 100 cm were recorded, but not tagged.

Double-tagged fish WT was estimated from a length-weight equation (Kohlhorst et al., 1980) and they were given intramuscular injections of OTC at the same dosage as for laboratory fish. Tagging methods for the single tags used in all years are described by Kohlhorst (1979, 1980). Briefly, the plastic disc-dangler tags were attached with stainless steel wire through the anterior portion of the basal musculature of the dorsal fin. A portion of the sturgeon were double tagged and injected in 1984 and 1985 with the single tag described above and either T-bar disc or spaghetti tags. All fish were single tagged in 1987.

We depended on anglers to report the capture of tagged sturgeon and to return the calcified structures needed for age analysis. They were offered and paid a reward of up to 20 dollars for returning tags. Also, measurements of TL and WT were acquired from the angler whenever they were available. Length frequencies and the recapture rate were then determined from these data.

Pectoral fin rays recovered from all reported recaptures were cleaned, air dried, and thin-sectioned using the same methods as those for the comparative analysis and laboratory grow-out studies. Thin sections from recovered fin rays were mounted on glass slides and viewed with the aid of a binocular microscope while being illuminated with ultraviolet (UV) light (360 nm wavelength) which causes the OTC marker in the bone to fluoresce. Annuli were counted from the origin of growth to the OTC mark and from the mark to the outer edge. For counts of annuli distal to the OTC mark, a value of 0.5 growth increments was assigned where a growth zone was only partially formed on the outer edge, whereas a value of 1 was assigned to counts of complete annuli. Assuming that growth increment formation and time at liberty, in years, were equivalent, counts of growth zones outside of the OTC marker were then plotted against the number of years at liberty after marking. A linear regression analysis was performed on these data and both the significance of the regression coefficient and the slope of the relationship were analyzed (Zar, 1984). The null hypothesis for this curve was that the slope (b) was equal to one, the expected situation that should occur if one annulus (one set of translucent and opaque rings) was deposited each year, and this was tested using a t-test, as described in Zar (1984).

Criteria for defining growth patterns and ageing terminology were as follows : We use the terms ring, annulus, and band to describe the growth patterns or growth increments found in the various calcified structures. The term ring describes depositional patterns associated with either winter (translucent) or summer (opaque) growth zones (Chilton and Beamish, 1982). Visually, these rings are not simply fine lines that encircle the origin of growth, but are of variable width, conform to the shape of each structure, and radiate from a point of origin somewhat parallel to the outer edge. The term band describes the wide opaque growth zones in only three of the structures used for comparative age analysis : opercles, clavicles, and cleithra. In these structures, the wide bands were composed of opaque bone with numerous ancillary rings, or calcium layers, which were initially confused with translucent rings. With some practice, these ancillary rings could be distinguished from translucent rings. As with the other structures, each (opaque) band and (translucent) ring pair were counted as an annulus (Brennan and Cailliet, 1989). Although the time period of translucent or opaque ring deposition has not been verified, we assumed that each annulus represented a yearly event. This is the basic assumption of all ageing studies in which age validation has not yet been attempted or completed.

RESULTS

Comparative Analysis

Summary of fish sampled

Of 147 white sturgeon collected between 1984 and 1985, TL and WT data were collected from 143 and 129 fish, respectively. Overall, sizes ranged from 31 cm TL to 224 cm, with most fish being between 100-180 cm. Sexes were determined for 66 males and 54 females. Males ranged in size from 52 cm (weight undetermined) to 190.5 cm (46.72 kg), whereas females ranged in size from 103 cm (5.44 kg) to 224 cm TL (80.74 kg)

There were statistically significant linear correlations between TL and FL for males ($N = 50$; $r^2 = 0.98$), females ($N = 39$; $r^2 = 0.98$) and all samples combined (sexed and unsexed, $N = 106$; $FL = - 3.55 + 0.93x$; $r^2 = 1.00$) ($P< 0.001$ for all correlations). The relationship between TL and WT (kg) was best expressed by the equation : log WT = - 5.21 + 2.98 log TL for males ($r^2 = 0.94$; $N = 63$), log WT = - 6.04 + 3.36 log TL for females ($r^2 = 0.94$; $N = 52$), and log WT = -5.70 + 3.19 log TL for all samples combined ($r^2 = 0.94$; $N = 124$) using a geometric mean functional regression (Ricker, 1973).

Pectoral fin ray sections as ageing material

Pectoral fin rays were collected from all 147 fish and thin-sectioned. Of those, 122 were measured from the origin to the outer edge of one posterior lobe.

There was a significant positive linear relationship between TL and fin section width (r^2 = 0.81), although there were differences in the morphology of each section due to the generally inconsistent formation and width of annuli between samples. Fin section width ranged from 1.67 mm (29 cm TL) to 8.62 mm (173 cm TL).

Growth rings were present in all fin sections examined and increased in number relative to section width and fish size. Annuli were more widely spaced near the origin and usually more tightly grouped toward the outer edge, especially in larger (older) fish (Figure 4). Tight groupings of annuli occurred frequently, even in earlier years, and was a common problem in annulus delineation. Therefore, it was occasionally necessary to get a consensus of more than one reader to define annuli in some samples.

Incorporation of secondary fin rays into the posterior lobe of the first ray, a phenomenon which has previously been described for white sturgeon, also reduced the readability of fin sections through crowding of annuli (Figure 5). This could potentially lead to errors in age determination. The inspection of samples with secondary rays incorporated and unincorporated revealed annulus counts that corresponded to counts of the first ray alone. In addition, the incorporation of secondary rays did not always occur bilaterally (in both right and left fin rays), nor did it always occur in both lobes of the same first ray. Secondary fin ray incorporation occurred in 52% of all fin sections examined.

The seasonality of depositional patterns is often determined by characterizing the clarity of the edge during the course of the year. It was difficult, however, to determine whether the edge was translucent or opaque in fin ray sections of white sturgeon. This was particularly true in larger (older) fish which tended to have more narrowly spaced annuli in the posterior portion of the fin section. Of the 125 fin section samples examined, distinct trends in seasonal formation were not apparent.

The precision of age estimates based on two independent, blind counts of fin section annuli by a single reader resulted in 38% exact agreement, 74% agreement within + 1 annulus counts and 84% agreement within + 2 annulus counts (Table 1). Comparisons between the two independent, blind counts of fin section annuli by more than one reader also were high (17 - 31% exact agreement, 57 - 63% agreement within + 1 annulus counts, and 77 - 84% within + 2 annulus counts.).

Other structures

There were positive, significant linear relationships between total length and measurements of all other skeletal structures used for comparative analysis (r^2 values ranged from 0.61 to 0.94 ; n = 110-126). Concentric ring and band patterns increased in number with the size of the fish, but were more difficult to analyze than pectoral fin sections because of the morphology of the structures, crowding of annuli, or incorporation of natural oils into the bone that reduced the legibility of certain structures. Tests for inter-reader precision resulted in relatively low levels of agreement (Table 1).

Single Reader Comparison of All Structures

There were positive, significant linear relationships in all 15 comparisons of annulus counts based on different structures from the same fish (Table 2).

All regressions were highly significant. Coefficients of determination ranged from r^2 = 0.96 for opercle versus clavicle annulus counts, to r^2 = 0.81 for opercle versus dorsal scute counts, and the slopes of all regressions did not differ significantly from 1 ($P < 0.001$). Exact agreement of paired annulus counts was relatively consistent, ranging from 24% to 28% for all structures compared to paired fin section counts (46%) (Table 3). The percent agreement within + 1 annulus counts increased markedly for cleithra (45%), clavicles (36%) and opercles (36%), with a cumulative percent agreement of 70%, 63%, and 62% respectively. Agreement within + 2 annulus counts was again highest for clavicles (18%), cleithra (11%) and opercles (12%) with cumulative percent agreement figures, (81%, 81%, and 74% respectively), almost equal to that of pectoral fin section annulus counts alone (84%).

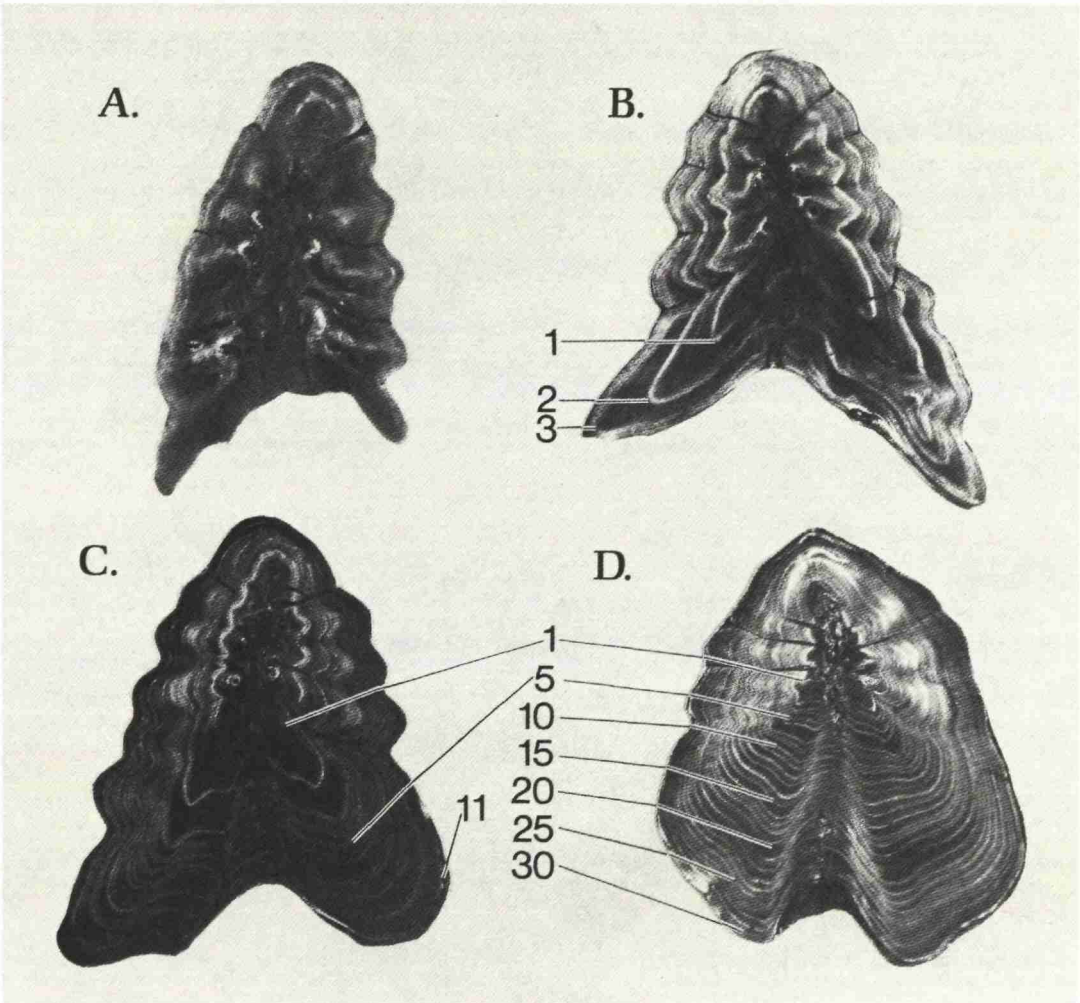


Figure 4 - Transverse section of first pectoral fin rays of white sturgeon of four ages. White rings are translucent and are assumed to be deposited during the winter growth period. Annuli (band pairs) are numbered for reference:
A- A zero year old fish (TL:31cm); B- Three year old fish (TL:60cm);
C- Eleven year old fish (TL:119cm); D- Thirty year old fish (TL:191cm)
(Taken from Brennan and Cailliet 1989).

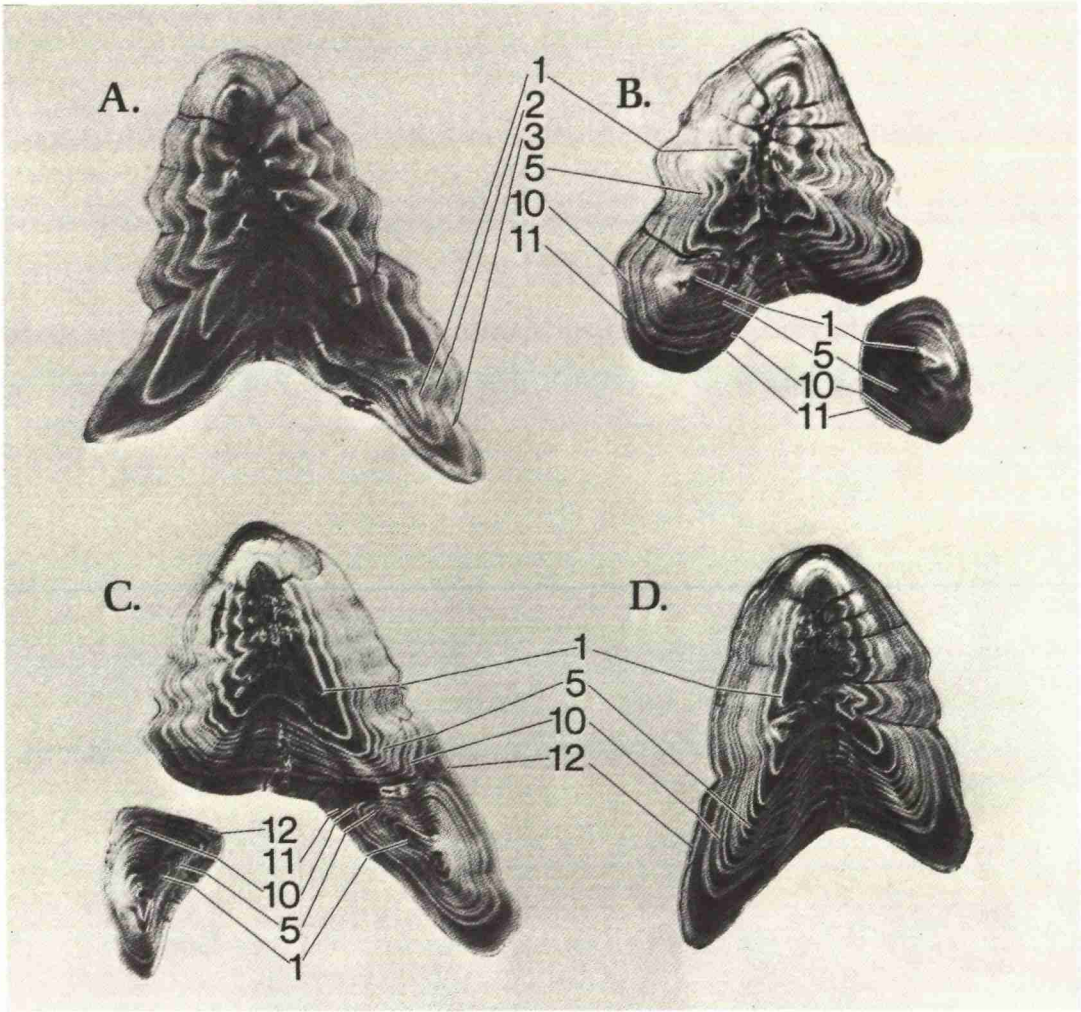


Figure 5 - Transverse section of first pectoral fin rays of white sturgeon which have incorporated secondary rays. Annuli are numbered for reference:
A- Secondary rays incorporated during second year of growth in a three year old fish(TL:60cm);
B- Secondary ray incorporated during fourth year of growth in a eleven year old fish(TL:122cm);
C- Secondary ray incorporated in the right fin ray (sample number 3122) during the eleventh year old fish (TL:127cm);
D- Left fin ray, sample number 3122. Secondary rays have not incorporated. Note that in all samples, secondary ray annulus counts, incorporated and unincorporated, agree with primary ray annulus counts. (Taken from Brennan et Cailliet 1989).

Table 1 - Summary of multiple reader count variability for five calcified structures of white sturgeon. Agreement categories were determined by comparing tow or more age estimates from the same structure (exact agreement = no discrepancies ; ± 1 = a difference of 1 annulus count, etc...). Values are cumulative percentages of agreements for the paired readings that fit each category. Reader numbers represent one of three readers making age estimates.

	Reader																
	1 versus 1					1 versus 2				1 versus 3				2 versus 3			
Agreement	exact	±1	±2	±3	n	exact	±1	±2	n	exact	±1	±2	n	exact	±1	±2	n
Fin sections	38	74	84	92	118	17	57	77	131	31	62	78	131	18	63	84	138
Medial nuchals						11	26	39	38	18	28	35	40	16	42	66	38
Clavicles										11	35	48	38				
Cleithra									14	32	43	29					
Opercles										15	30	53	39				

Table 2 - Results of regression analysis of annulus counts from the six calcified structures of white sturgeon used for age estimation. r^2 = coefficient of determination. b = regression coefficient, or slope of the best fit regression line. (Reproduced from Brennan and Cailliet, 1989).

b	r^2	Structure counts compared
1.10	0.88	Pectoral fin section versus medial nuchal
1.01	0.96	Clavicle versus operculum
1.01	0.94	Cleithrum versus operculum
0.98	0.96	Clavicle versus cleithrum
0.93	0.83	Pectoral fin section versus dorsal scute
0.90	0.94	Pectoral fin section versus operculum
0.89	0.92	Pectoral fin section versus clavicle
0.88	0.88	Pectoral fin section versus cleithrum
0.87	0.81	Dorsal scute versus operculum
0.85	0.88	Dorsal scute versus clavicle
0.84	0.90	Medial nuchal versus dorsal scute
0.80	0.85	Dorsal scute versus cleithrum
0.74	0.86	Medial nuchal versus clavicle
0.73	0.83	Medial nuchal versus cleithrum
0.72	0.85	Medial nuchal versus operculum

Each structure was ranked, based on the cumulative percent agreement within + 3 annulus counts, to provide a relative index of interpretability. Pectoral fin section annulus counts ranked highest with 92% cumulative agreement followed by cleithra (90% cumulative agreement with the standard), clavicles (88%), opercles (86%), dorsal scutes (67%), and medial nuchals (51%).

Table 3 - Percent agreement values of annulus counts of each structure versus pectoral fin ray section annulus counts used as a standard. Each comparison of annulus counts were of different structures from the same sturgeon, and were of a single reader. For fin sections, the data are two independent counts of annuli from the same structure. Percent agreement exact equals perfect agreement, ± 1 equals a difference of one annulus, etc... (Reproduced from Brennan and Cailliet, 1989).

	Structure					
Percent agreement	Fin section	Medial nuchal	Dorsal scute	Clavicle	Cleithrum	Opercle
exact	46	24	28	27	25	26
± 1	36	10	14	36	45	36
± 2	2	7	11	18	11	12
± 3	8	10	14	7	9	12
Cumulative	92	51	67	88	90	86
Rank	1	6	5	3	2	4

Laboratory grow-outs

Of the 102 hatchery-reared sturgeon collected for incremental growth analysis, TL values ranged from 290 mm to 1170 mm for the 87 untagged sturgeon and from 460 mm to 940 mm for 15 of the original 20 tagged sturgeon remaining at the end of the 20-month grow-out period (Figure 6). Ages at sacrifice ranged from 10 to 49 months (Table 4).

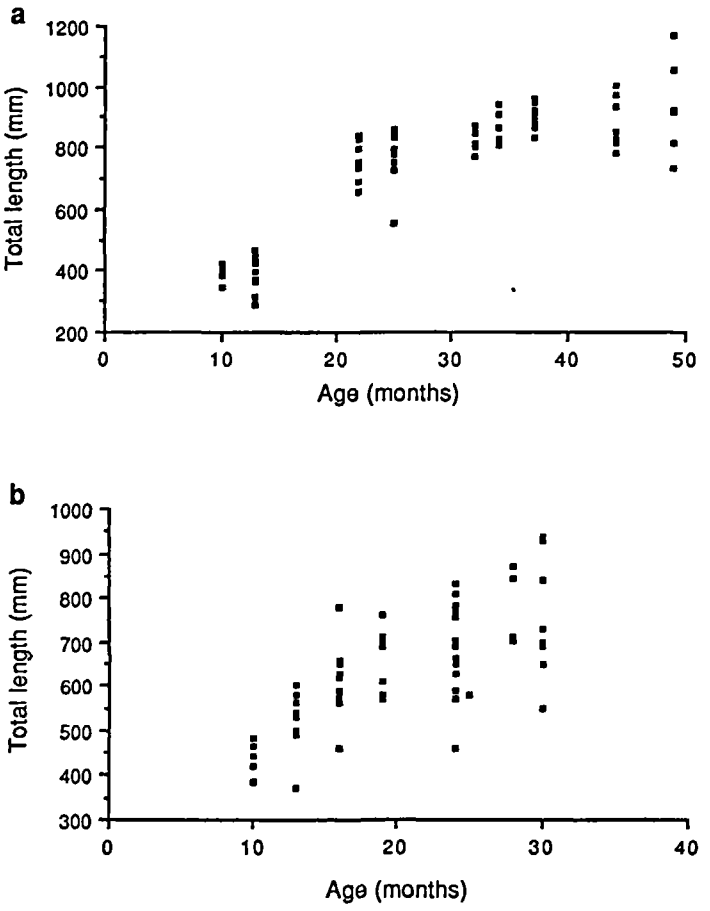


Figure 6 - Scatterplots of total lengths and and known ages of:
a- 87 hatchery-reared sturgeon from 4 year classes and b- 67 measurements of total length taken at 6 intervals, during a 20-month period, from 15 sturgeon grow-outs used for age verification studies.

Of the 102 pectoral fin rays that were collected for analysis, most were curled and deformed, about 10 % of the fin sections inspected for depositional patterns revealed annulus counts that corresponded to the known age, and distinct translucent and opaque ring patterns were not apparent as in fin ray sections of wild fish (Figure 7). The illegibility of the fin sections was due to a variety of characteristics including the translucency of sections, the distinct morphological differences of the sections, and the "marbled" appearance in these fin sections.

Table 4 - Summary of all hatchery-reared sturgeon used for growth increment analysis

Year Class	Sacrificed month/year	Number sampled	Known Age (months)
1981	11/84	8	44
19818	4/85	8	49
1982	11/84	10	32
1982	1/85	11	34
1982	4/85	10	37
1983	1/85	10*	22
1983	3/85	2*	24
1983	4/85	10*	25
1983	4/85	1*	25
1983	7/85	4*	28
1983	9/85	8*	30
1984	1/85	10	10
1984	4/85	10	13

* Tagged fish used for grow-out studies

More than 30% of the fin sections examined had a translucent appearance when illuminated with transmitted or reflected light and appeared in all fish from three of the year classes sampled (Figure 8a). This resulted in an inability to distinguish alternating, contrasting translucent and opaque growth zones. This characteristic ranged from completely translucent and totally illegible to partially translucent with some contrasting zonation, most often unlike wild fish growth patterns. Although translucency has been noted in wild sturgeon fin sections, the occurrence is rare and does not usually impair the reader from making an age estimate.

The morphology of the fin section perimeter was distinctly different from wild sturgeon samples (Figure 8b). The overall shape of the ray section appeared compressed and malformed, especially the anterior edge. In addition, fingerlike projections of calcified material that radiated out from the medial line were common in ray sections of younger fish. This resulted in the compression or interruption of translucent or opaque patterns.

The "marbled" appearance of fin sections was usually the result of the compressed shape (anterior to posterior), presence of numerous vascular channels, and often secondary ray incorporation and heavily calcified, opaque patchy zones (Figure 8c). In addition, fingerlike projections from early growth malformations were evident in many samples. The combined effect of these characteristics obscured or interrupted translucent and opaque patterns.

Of the 15 remaining hatchery sturgeon used for grow-out studies, 7 had been injected with OTC and 8 had not been injected. Microscopic inspection and UV illumination of the 7 fin ray sections of the marked fish revealed distinct OTC markers incorporated into all 7 sections. The one fish that had been injected on two occasions showed two OTC markers. Although fin sections from marked hatchery fish revealed calcium deposition distal to the OTC marker, growth patterns were unlike those of wild fish, with or without OTC markers (Figure 9).

Field mark-recapture

Of the 2187 sturgeon that were tagged, injected with OTC, and released between October, 1984 and December 1987, a total of 419 was recaptured as of mid-March 1989, and pectoral fin rays were recovered from 113 of the recaptures that were reported(Table5).

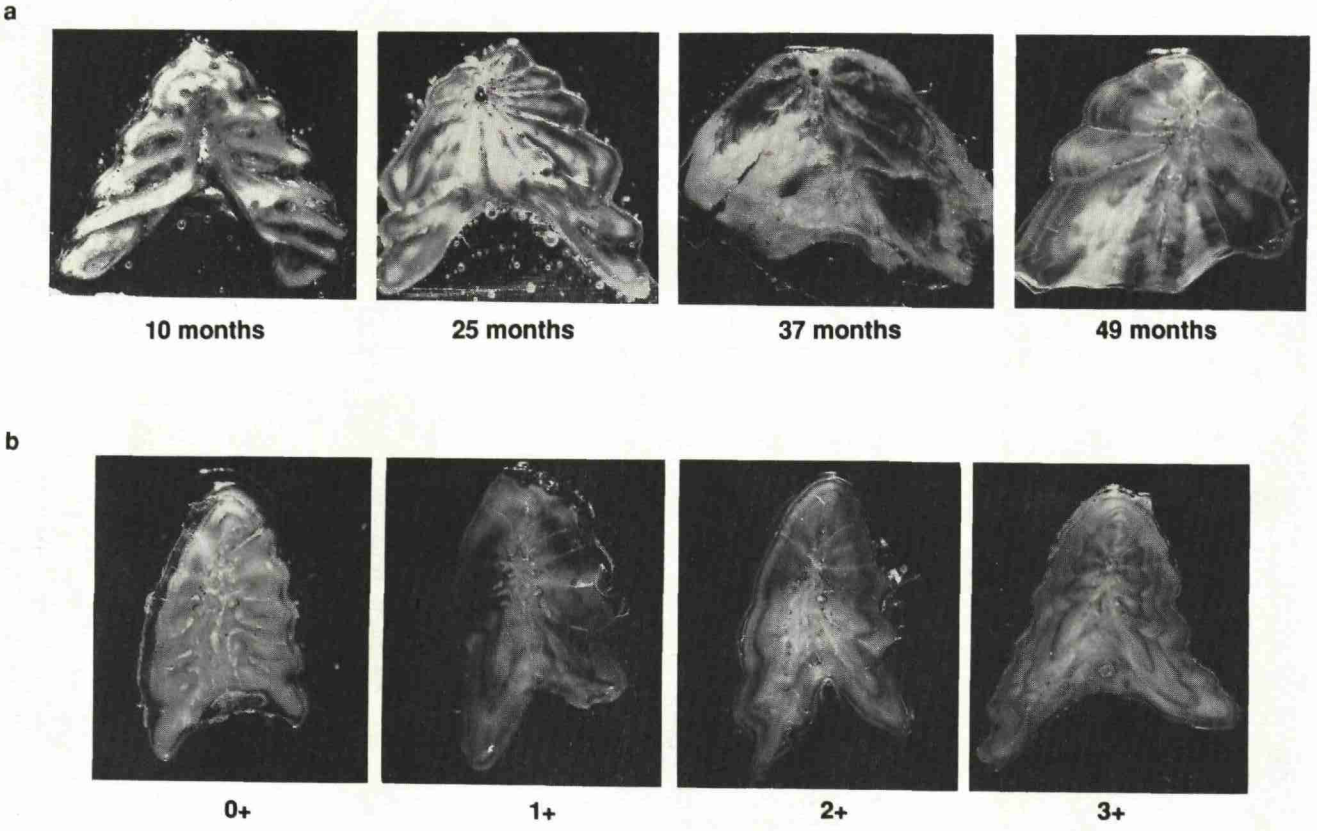


Figure 7- Photographs of white sturgeon pectoral fin ray sections from: a- Hatchery-reared sturgeon of 4 known age groups(10-49months), b- Wild sturgeon from 4 age-estimated groups.

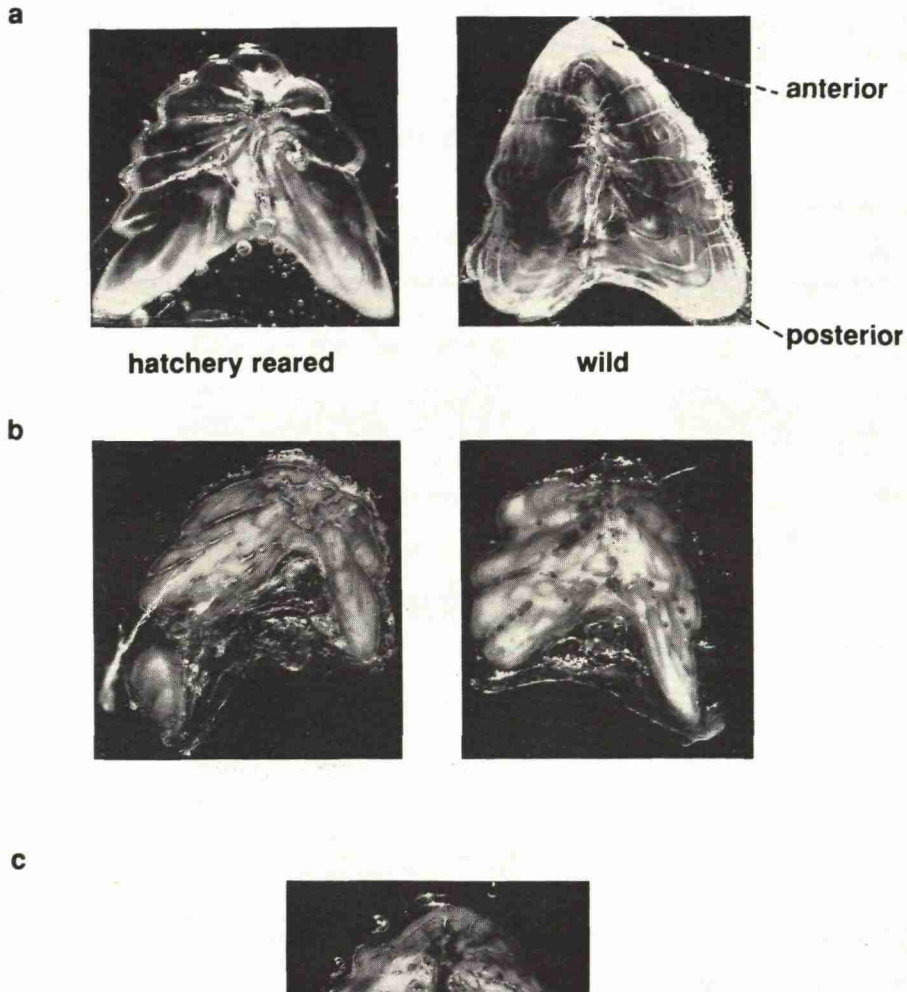


Figure 8- Photograhs of white sturgeon pectoral fin ray sections from hatchery-reared sturgeon to illustrate the illegibility due to:
a- Translucency in hatchery-reared samples versus a wild sample,
b- Morphological differences and
c- The "marbled" appearance of deposition. Characteristics are labeled for.reference.

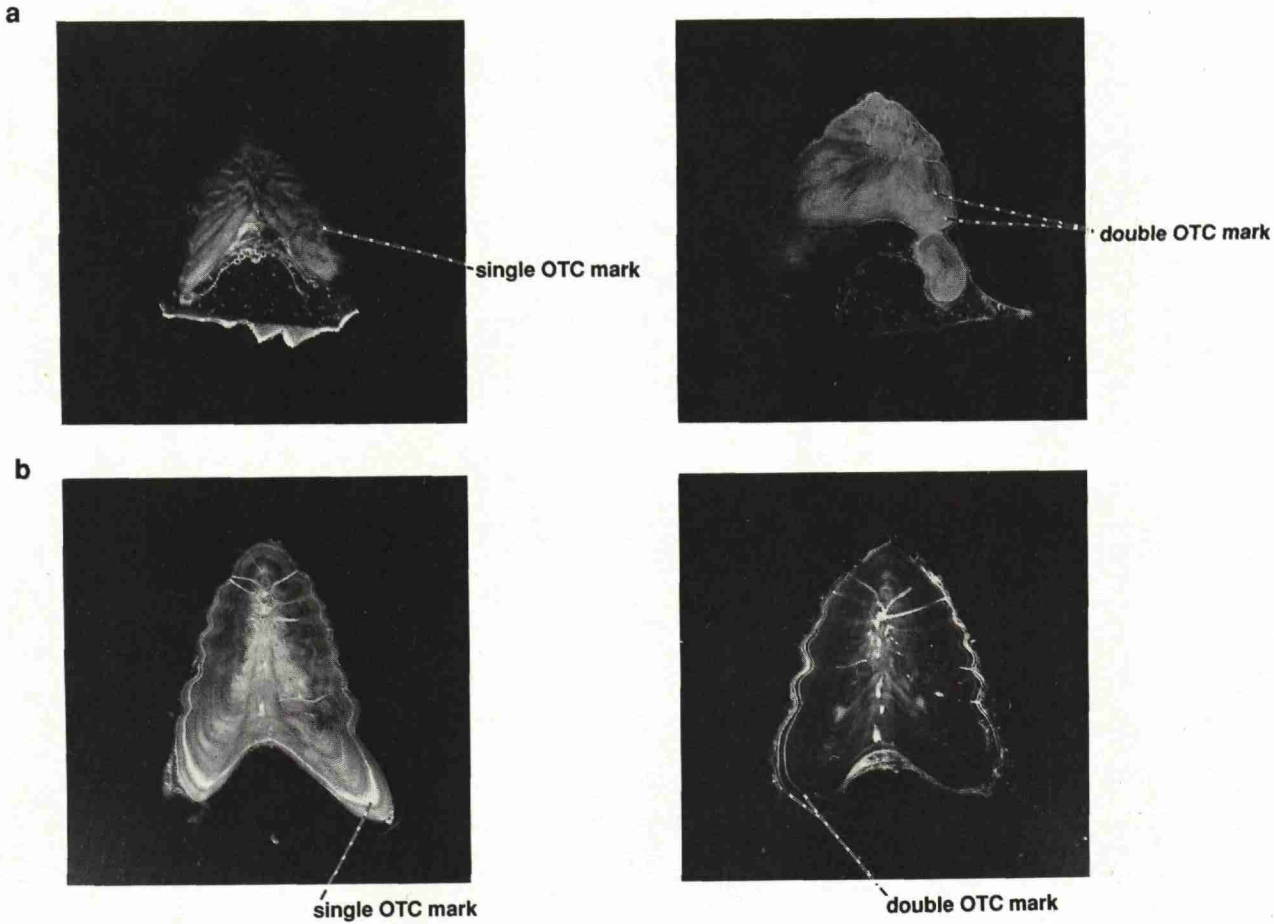


Figure 9 - Photographs of white sturgeon pectoral fin ray section illustrating incorporation of OTC in: a- Hatchery-reared sample, and b- Wild sample

Table 5 - Summary of marked and recaptured sturgeon from three tagging years .

1984							
Number tagged (no OTC)	1901			Years			
Number tagged (w/OTC)	662						
Total tagged	2563	1	2	3	4	5	Totals
Number of tags at start of year (no OTC)		1901	1765	1622	1572	1535*	1522
Number recaptures (no OTC)		136	143	50	37	13	379
Percent recapture (no OTC)		7.2	8.1	3.2	2.4	0.8	
Number of tags at start of year		662	600	542	513	496	492
Number recaptures (w/OTC)		62	58	29	17	4	170
Percent recapture (w/OTC)		9.4	9.7	5.4	3.3	0.8	
Total Recaptures		198	201	79	54	17	549
Total percent recaptures		7.7	8.5	3.6	2.6	0.8	
(OTC injected samples recovered for age analysis = 36 or 21%)							
1985							
Number tagged (no OTC)	1653						
Number tagged (w/OTC)	754						
Total Tagged	2407						
Number of tags at start of year (no OTC)		1653	1512	1443	1385*		1378
Number recaptures (no OTC)		141	69	58	7		277
Percent recapture (no OTC)		8.5	4.5	4	0.5		
Number of tags at start of year (w/OTC)		754	667	627	599		592
Number recaptures (w/OTC)		87	40	28	7		162
Percent recapture (w/OTC)		11.5	6	4.5	1.2		
Total recaptures		228	109	86	14		437
Total percent recaptures		9.5	5	4.2	0.7		
(OTC injected samples recovered for age analysis = 45 or 28%)							
1987							
Number tagged (no OTC)	211						
Number tagged (w/OTC)	754						
Total Tagged	982						
Number of tags at start of year (no OTC)		211	199*				193
Number recaptures (no OTC)		12	6				18
Percent recapture (no OTC)		5.7	3				
Number of tags at start of year (w/OTC)		771	705				684
Number recaptures (w/OTC)		66	21				87
Percent recapture (w/OTC)		8.6	3				
Total Recaptures		78	27				105
Total percent recaptures		7.9	3				

* (OTC injected samples recovered for age analysis = 32 or 41%)
Data represent only 5 months of recapture year 1988-1989

Total lengths of OTC-marked fish ranged from 100 cm to 214 cm at tagging and 96.5 cm to 188 cm at recapture (Figure 10). Time-at-liberty after tagging ranged from 9 to 1138 days.

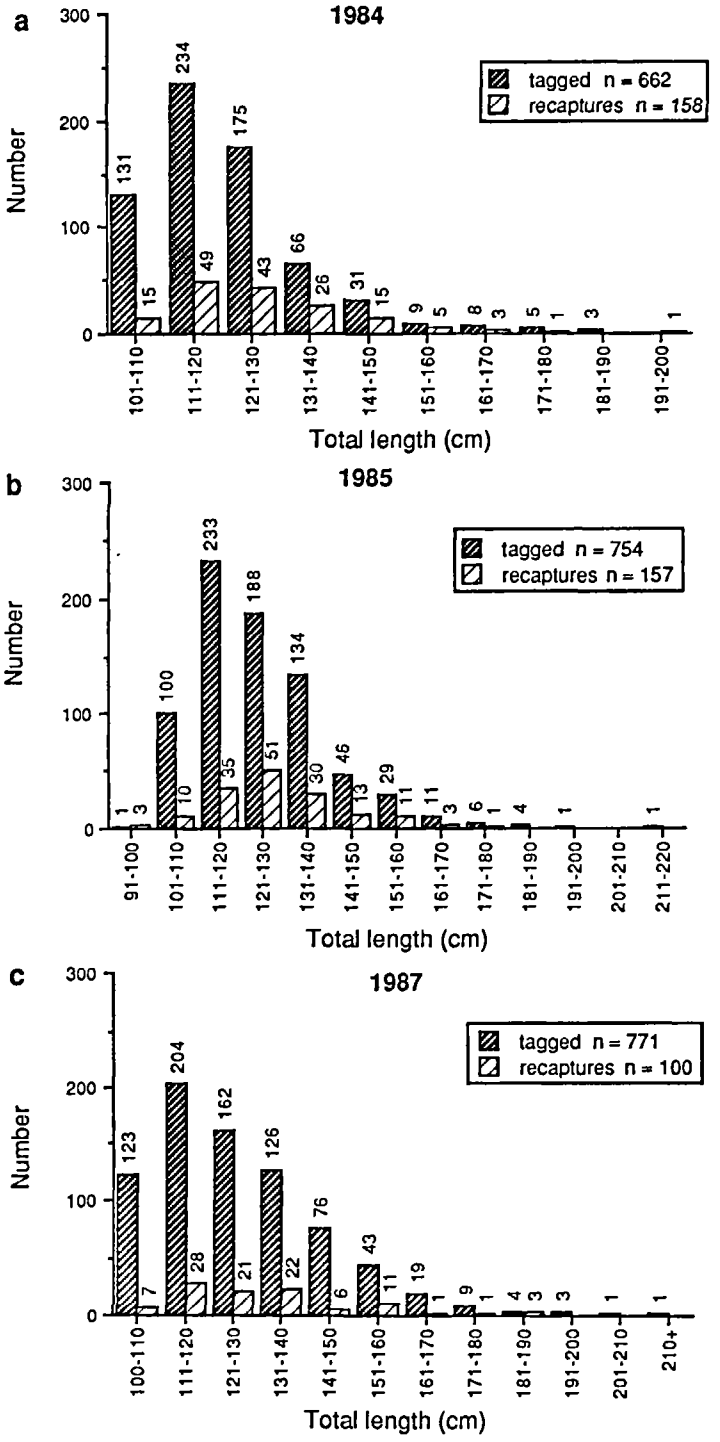


Figure 10 - Total lengths of tagged and recaptured white sturgeon from three tagging years; a- 1984, b- 1985, c- 1987.

Although most sturgeon were recaptured within the San Francisco Bay-Delta system, recaptures ranged from Santa Cruz, California in the south to Tillamook Bay, Oregon in the north.Oxytetracycline markers were evident in pectoral fin ray sections and all other calcified structures recovered from recaptured sturgeon when illuminated with UV light, illustrating synchronous growth in all structures. Growth rings and OTC markers found in fin ray sections followed depositional patterns similar to those described in previous sturgeon ageing studies (Brennan and Cailliet, 1989). The OTC marker appeared as a narrow, yellow-gold band that ran somewhat parallel to the periphery of the structure (Figure 11a).

In most samples that were at liberty for greater than one year, the fluorescence was found within the opaque zone, but also occurred on the distal margin of the opaque zone making it difficult to differentiate between the following translucent ring and OTC marker. It was difficult to characterize growth deposited subsequent to the OTC marker in fish at liberty less than 1 year.

Most fin ray sections revealed depositional patterns that supported the hypothesis that one translucent and one opaque ring were laid down each year, although the sample recovered from Tillamook Bay Oregon showed little growth subsequent to the OTC marker. The recapture TL reported for this fish was 7 cm less than the TL measurement when tagged. Thus, it is possible that this fish shrank during the long migration to Oregon, but the dependability of measurements taken by untrained personnel is questionable. Evidence from the fin ray section also suggests that there was little growth (Figure 11b).

A regression analysis performed on data from all sturgeon at liberty for approximately one year or greater supported the hypothesis that one set of translucent and opaque rings were deposited each year (Figure 12). There was a positive, significant linear relationship between the number of years at liberty and the number of rings after the OTC marker ($r2 = 0.69$; $n = 32$; $P < 0.001$). Although the relationship between time-at-liberty and growth increment formation was highly significant for all samples combined, the relationship was strongest for "smaller" fish (100-120 cm TL ; $b = 0.91$; $r = 0.89$; $n = 18$), followed closely by "medium" fish (121-140 cm TL ; $b = 0.54$; $r = 0.87$; $n = 8$) when the samples were arbitrarily separated into three size categories. In contrast, the time-at-liberty to growth increment formation relationship for "large" sturgeon was poor(141+ cm TL ; $b = 1.62$; $r = 0.64$; $n = 6$).

Rate of recapture was significantly higher ($P < 0.001$) for sturgeon injected with OTC compared to sturgeon that were not injected in all three tagging years. Differences ranged from 2.2% to 3.0% during the first year after tagging This could not be attributed to the application of a second tag because of the high shedding rate of these tags. The primary, disc dangler tags attached with stainless steel wire were retained better and remained legible for at least three years subsequent to tagging.

DISCUSSION

The initial step to be taken when conducting any age and growth study is the selection of a bony structure which will provide the most accurate information about the age of a fish. The selection of an optimal ageing structure should be based upon the comparison of a variety of structures using the following criteria : 1) clarity of ring patterns ; 2) percent agreement or precision of pattern interpretability ; 3) ease of collection and processing ; and, ultimately 4) accuracy of ageing a specific structure. The results of the comparative analysis, and preliminary results of laboratory and field studies presented here have at least partially fulfilled the above criteria for determining the true meaning of depositional patterns found in calcified structures of white sturgeon.

An important assumption inherent in growth studies based on skeletal structures is that fish and structure size are closely related throughout the entire life span (Lee et al., 1983; Smith, 1983). Although many attempts have been made to estimate the age of sturgeons using skeletal structures, few studies have examined this relationship.

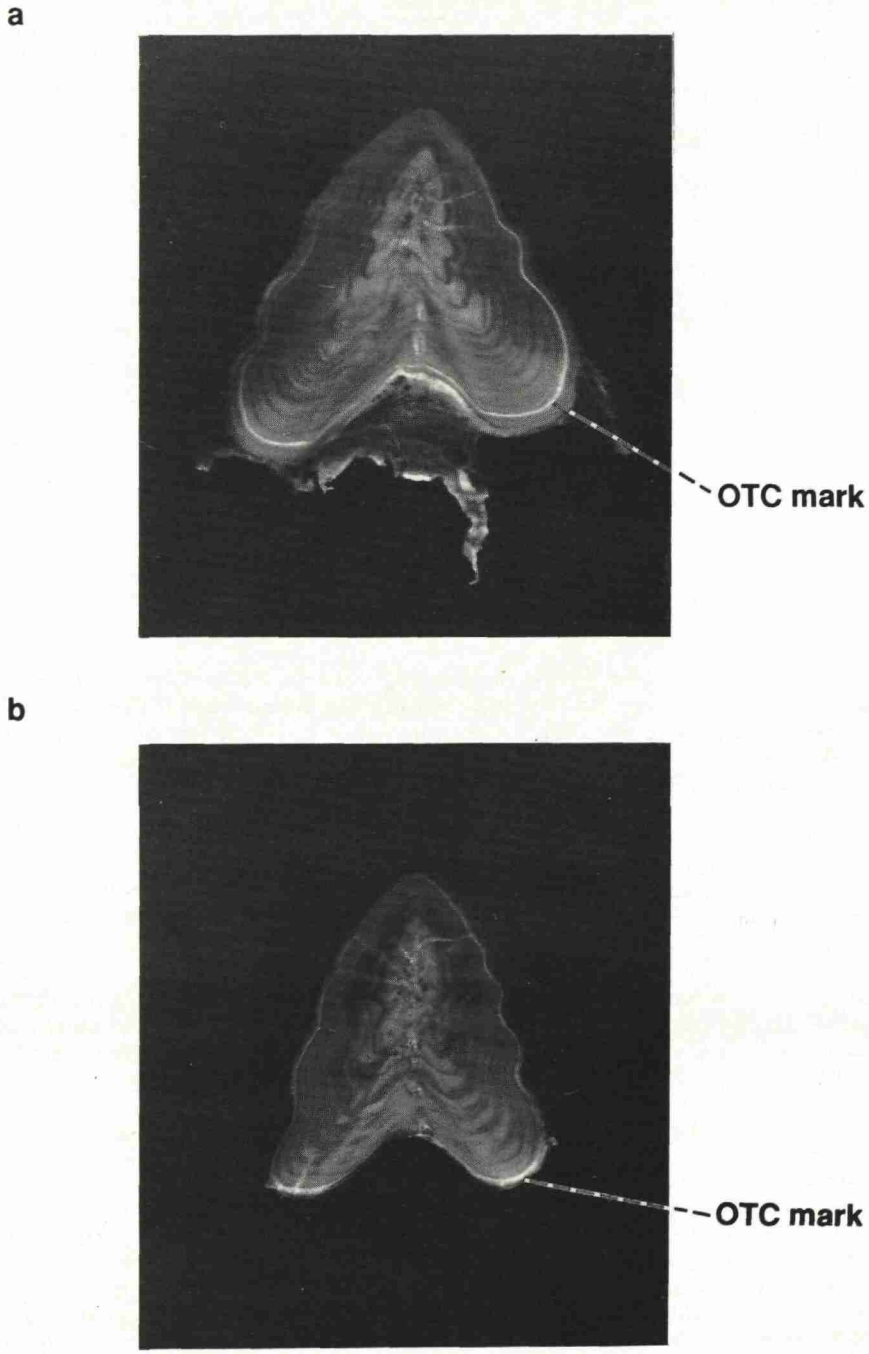


Figure 11- Photographs of pectoral fin ray sections recovered from mark-recaptured white sturgeon. Note the OTC marker and the subsequent growth for a- a sturgeon at large for 554 days and b- a sturgeon recaptured in Tillamook Bay, Oregon, at liberty for 586 days.

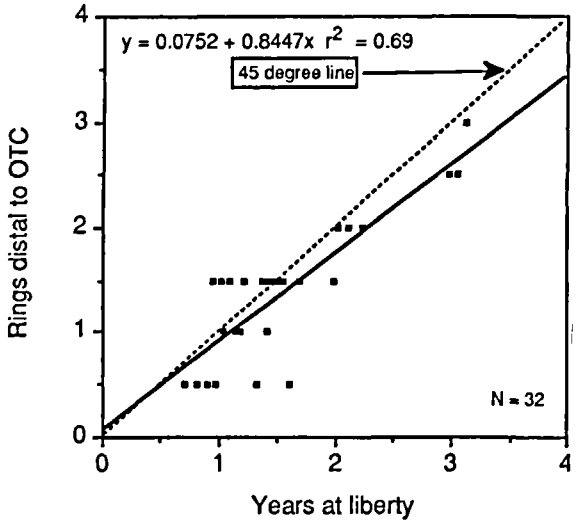


Figure 12- Scatterplot and regression of growth rings distal to the OTC marker versus the number of rays at liberty using pectoral fin ray sections of white sturgeon as the ageing structure (45 degree line, b = 1, is labelled for reference)

Sunde (1961) found a poor relationship between fin section measurements and fork length of lake sturgeon *Acipenser fulvescens*, but Semakula and Larkin (1968) found a significant relationship between fin section width and fork length of white sturgeon (r^2 = 0.81). The positive relationships between total length and the size characteristics of each structure used in our study supported the use of the structures for age estimation. These relationships and usefulness of each structure for age determination are reviewed by Brennan and Cailliet (1989). Other studies have not used this procedure for fin sections or any other structure.

Verification and validation of age estimates are now considered to be essential, yet often overlooked, elements in age and growth studies (Beamish and McFarlane, 1983). In this study, individual white sturgeon ages were at least partially verified by comparing age estimates from several structures. The high level of inter-reader agreement for pectoral fin section counts provided good evidence of similar age determinations. In addition, high intra-reader agreement between structures suggests corroboration. Thus, pectoral fin ray sections were used to generate information for a white sturgeon growth model and were used for analysis of depositional patterns in hatchery-reared and field-marked fish.

To fully understand the timing and frequency of ring deposition throughout a year for a particular species, investigators must collect samples throughout the year and correlate edge characteristics with a particular time of the year, or use internal markers of previously marked fish as a point of reference. Inspection of edge characteristics on unmarked white sturgeon fin ray sections revealed no distinct trends in the time of translucent or opaque ring formation. The inability to fit a definitive edge characteristic (translucent or opaque) to the assumed proper season was probably due to the differential width of growth rings. Translucent rings were narrow compared to opaque rings and were very difficult to distinguish at the outer edge, especially if it was early in the time of formation or if the outer ring pairs were narrowly spaced as in larger (older) fish. Also, opaque ring widths varied within samples, generally being wider in early years and more narrow and closely spaced in later years. This may indicate variable rates of growth throughout the life of a particular individual, but may not be a good indicator of time of annulus formation.

The use of hatchery-reared sturgeon for age verification was useful for meeting only one of our original objectives, due to the anomalous growth patterns found in pectoral fin ray sections. Other researchers have also noted that growth increment formation in calcified structures of wild fish were usually clearer and easier to count than the patterns found in laboratory-reared larvae (Laroche et al., 1980 ; Lough et al., 1982 ; Campana, 1984 ; Savoy and Crecco, 1987). Factors that may have contributed to the deformities observed in the fin ray sections include growth rate, food and feeding schedule, water quality, and confinement. Although the effects of the higher growth rate of these fish on normal calcification processes has not been determined, the visibly abnormal patterns suggests a possible cause and effect relationship. In addition, the ad libitum feeding with a commercial feed may have contributed to the higher growth rate, but may also not be a suitable diet for the nutritional needs of sturgeon.

Water temperatures seemed to follow ambient seasonal fluctuations, but may not have been similar to wild conditions. Water temperatures have been shown to play an important role in larval fish otolith depositional pattern formation (Nielsen and Geen, 1982 ; Campana, 1984) and therefore, may have contributed to the anomalous patterns found in fin ray sections of hatchery-reared sturgeon. In addition, the early movement of wild fish into brackish water may be a part of the stimulus to grow, take up more calcium from the water, and lay down depositional patterns in calcified structures. Also, the freshwater used to rear sturgeon may not have had enough calcium available for the formation of normal growth patterns.

Confinement and crowding may also have been factors that influenced the anomalous growth of pectoral fin rays. It could not be determined if the deformed pectoral fin rays resulted from an incomplete diet or from rubbing on the bottom and sides of the fiberglass holding tanks. Lateral scutes were also eroded on several of the grow-outs.

Although the hatchery-reared, known-age sturgeon samples provided poor evidence of early depositional patterns, the use of field mark-recaptures have provided some evidence for defining seasonal patterns. Based on the assumption that translucent and opaque rings were deposited seasonally, it was also assumed that the OTC would provide some evidence for growth zone formation during that period. Our preliminary findings support both of these assumptions with the exception of the sturgeon recaptured in Oregon. For samples that were at large for less than 1 year, growth subsequent to the OTC marker was difficult to describe, possibly because the period of marking was to be followed by the deposition of a narrow, translucent ring. The OTC markers were found within or near the marginal edge of an opaque zone in those samples at liberty for more than 1 year. This suggests that the months of October through November may be a transitional period. It also suggests that growth ring deposition may be variable between individual sturgeon, or that extrinsic factors (i.e. temperature) occurring during the various tagging months and years may have influenced depositional timing.

In addition, size/age of the fish may influence the readability of fin sections that may, in turn, indicate a difference in timing of growth increment formation. The strong relationship between time-at-liberty and growth increment formation for small and medium size sturgeon support the hypothesis that one set of translucent and opaque rings are formed each year. Analysis of large sturgeon fin sections did not support this hypothesis. This may have been due to the small number of samples from the largest size group which may be insufficient for this type of analysis, or may be a result of the increased difficulty in reading fin sections from larger/older sturgeon. Regardless, this difference in relationships illustrates the need to validate for all sizes and age classes with sufficient sample sizes.

The usefulness of OTC for mapping calcium deposition and determining timing and frequency of formation is illustrated by the 100% success rate of marking and its persistence in the bone for at least three years. Both hatchery-reared and wild fish incorporated OTC into the pectoral fin rays and other calcified structures, revealing a fluorescent, yellow-gold ring when illuminated with UV light. This technique also enables fisheries biologists to back calculate with greater accuracy when attempting to determine year class and early growth.

Although depositional patterns found in the hatchery fin rays did not provide information on early growth, the preliminary results of the field mark-recapture studies suggest that depositional patterns are seasonal and annual in nature with few exceptions. Until more marked fish from a wide size range have been recovered for analysis, timing of ring formation in calcified structures of white sturgeon must be assumed.

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SPAWNING POPULATION CHARACTERISTICS OF *Acipenser sinensis* IN YANGTZE RIVER JUST BELOW GEZHOUBA DAM

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RESUME

L'esturgeon chinois est anadrome. Quelques caractéristiques de la population en migration génésique sont les suivantes: la longueur des mâles est comprise entre 161 et 264 cm pour des poids de 47 à 195 kg et des âges de 9 à 26 ans, celle des femelles est comprise entre 222 et 346 cm pour des poids de 109 à 410 kg et des âges de 14 à 30 ans. La proportion de poissons matures a varié entre 13,5% et 78% après la construction du barrage de Gezhouba. Cependant, la structure de cette population, avec un sex-ratio d'environ 1:1, n'a pas changé significativement depuis l'interruption du courant en 1981. Bien que l'existence de l'espèce ne soit pas menacée, l'interdiction de la pêche commerciale devrait être maintenue car la puberté est tardive (au moins 9 ans pour les mâles et 14 pour les femelles) et les animaux se reproduisant pour la première fois représentent la majorité de cette population.

Mots clés: *Acipenser sinensis*, reproducteurs, âge, biométrie, barrage

ABSTRACT

The chinese sturgeon is anadromous. Some characteristics of the genetic migrated population are as follow: the males ranged in size from 161-264 cm BL (47-195 kg) at ages of 9-26 years and females from 222-346 cm BL (109-410) at ages of 14-30 years. The proportion of mature fish fluctuated from 13,5% to 78% after the construction of the Gezhouba dam. However, the structure of its spawning population with a sex ratio of about 1:1 has not changed significantly since the interception of the flow in 1981. Though the existence of the species is not threatened by daming, commercial fishing should be banned as they mature at an advanced age (at least 9 for males, 14 for females) and recruit population took up a majority of spawning population.

Key-words: *Acipenser sinensis*, breeders, age, biometry, dam

INTRODUCTION

Acipenser sinensis Gray is an anadromous fish of enormous size, it may reach a maximum body weight of 550 kg. It inhabits the continental shelf of the east cost of Asia. Every year in june and july, individuals approaching maturity ascend the Yangtze river and spawn in the fall of the following year. During this migration, their gonads mature to the final stage. Their larvae descend the river and occur mostly at the estuary in may and june of the year after. Its former spawning ground was distributed mainly in the lower reaches of the Jinsha river and in the upper reaches of the Yangtze. The annual catches of the sturgeon in the provinces along the Yangtze averaged 400 to 500 fish in the 1970's, with an output of 60 to 75 tons (Anonymous, 1988).

The first investigation on the spawning ground was primarily in 1964 by Fisheries Institute of Changshou Lake. A little information about this species was recorded by Institute of Hydrobiology (Anonymous, 1976). A considerable survey was carried out from 1972 to 1975 concerning a wild field of the sturgeon (Anonymous, 1988). Interest increased when it became apparent that the construction of the Gezhouba Hydro-electric Project would block the migratory passage of this species.

The objective of our study is to evaluate the present condition of its spawning population which is blocked below the dam (Fig. 1).

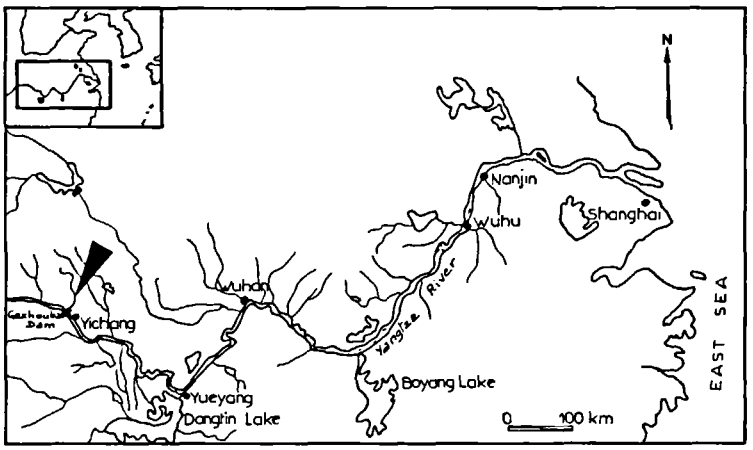


Figure 1- Map of the middle and lower reaches of Yangtze river.

Fish were captured with setlines of 50 to 100m, carrying 200 to 400 hooks (Fig. 2). The setlines with a single hook of 8 cm long (0.2 cm in diameter) are considered to be not selective.

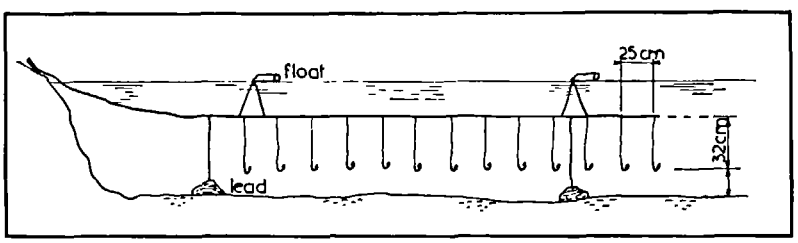


Figure 2- Shematic setlines used for Acipenser sinensis catches

Investigations were carried out in the fall from 1981 to 1982 and from 1986 to 1989 covering some 20km just below the dam. The gonads' developmental stages of fish were determined according to the description of Zhou Chunsheng et al.(1985). As all the fish caught along the Yangtze have an advanced maturity, the first stages were absent and we only observed the following ones:

III: Ovaries brown. Oocytes imbeded in fat, difficult to be separated from ovaries. Oocytes' diameter : 2,5 - 3,7mm.
Testicles: pink and fatty.

IV: Almost no more fat in ovaries. Oocytes black, animal pole very clear. Diameter: 3,7 - 4,9mm.
Testicles white, very fine and plump

V: Ova and milt are flowing.

VI: Sexual products deposited.

Age determination have been made by using the first pectoral fin rays collected in 1981, 1982, 1986 and 1987. It had been reported that *A.transmontanus*, *A.stellatus* and *A.gueldenstaedti* had got reproduction marks in the pectoral fin rays (Roussow, 1957; Veshchev, 1977,1979; Shilov et al.,1971,1977). These authors regarded the zones of abruptly crowding of the annuli, forming a "belt" of closely spaced annuli as a reproduction mark. An example is shown in figure 3.

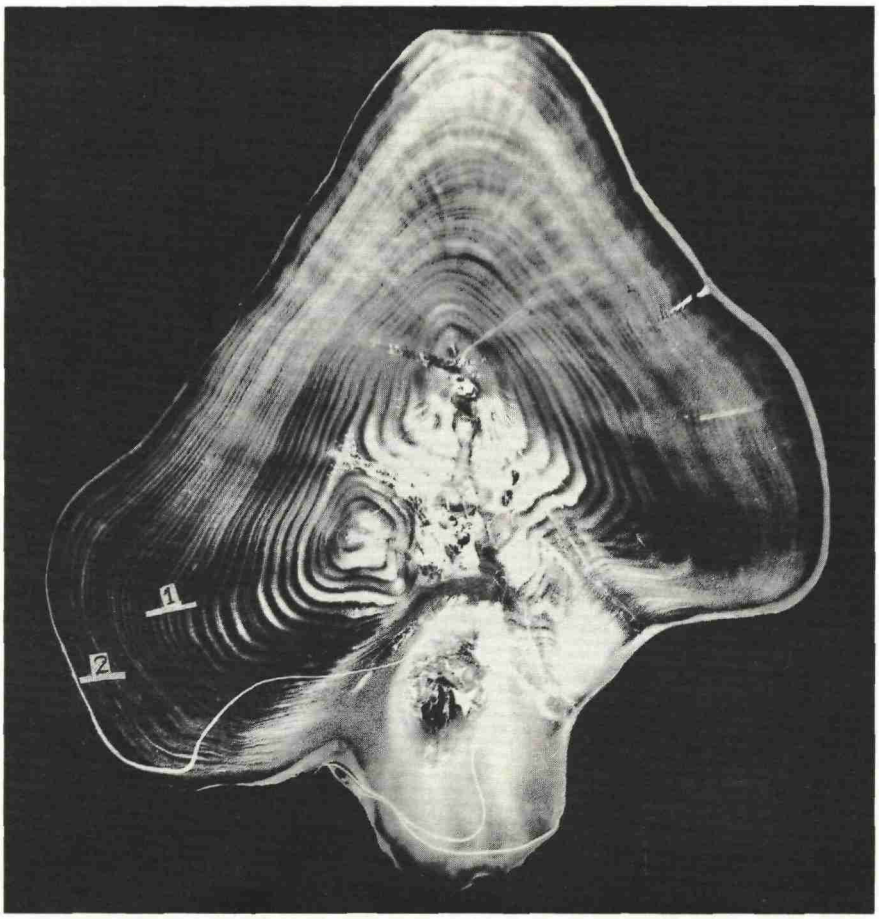


Figure 3- Section of first pectoral fin ray of a 33 years old chinese female sturgeon, showing successive "belt" of crowding annuli. 1- First mature mark, 2- Second mature mark.

RESULTS

Body length and Weight

The body length of 469 adult sturgeons was recorded during the study, 247 of them were male and 222 female. The body length of males ranged from 161 - 264cm (Table 1). The majority (92%) lays in the range of 170 to 230cm, with a mean length of 202cm. For the females the body length is in the range of 222 - 346cm. The majority (93%) lays in the range of 220 - 300cm with a mean body length of 263cm (Table 1).

Table 1- Body-length frequency distribution of male and female in the spawning ground of A.sinensis

Length (cm)	Male					Female				
	1972 1975	1981 1982	1986 1987	1988 1989	Total	1972 1975	1981 1982	1986 1987	1988 1989	Total
170	1	3	5	3	12	--	--	--	--	--
180	1	8	8	7	24	--	--	--	--	--
190	7	20	13	22	62	--	--	--	--	--
200	6	21	20	26	73	--	--	--	--	--
210	6	24	8	17	55	--	--	--	--	--
220	15	8	12	11	46	--	--	1	4	5
230	8	11	5	1	25	--	5	8	6	19
240	5	4	--	2	11	1	6	10	4	21
250	3	2	--	--	5	6	15	10	9	40
260	--	1	--	1	2	10	11	11	8	40
270	--	--	--	--	--	9	18	8	7	42
280	--	--	--	--	--	8	10	2	8	28
290	--	--	--	--	--	11	10	5	6	32
300	--	--	--	--	--	11	10	3	1	25
310	--	--	--	--	--	4	2	--	1	7
320	--	--	--	--	--	--	2	1	--	3
330	--	--	--	--	--	1	1	1	--	3
340	--	--	--	--	--	--	--	--	1	1
350	--	--	--	--	--	--	--	--	1	1
Total	52	102	71	90	315	61	90	60	56	267
mean	211	201	204	201		274	266	262	262	
σ_{n-1}	19	18	16	16		20	22	24	26	

The body weight of 291 sturgeons was recorded from 1981, 1982, 1986 and 1987, 155 of them were male and 136 were female. The weight of males ranged from 47 - 195kg (Table 2). The majority (77%) ranged from 60 - 120kg, with a mean weight of 94kg. The weight of the females ranged from 109 - 410kg. The majority (76%) lays in the range of 150 - 280kg with a mean weight of 215kg(Table 2).

The frequency distribution of the body-length seems to shift slightly towards smaller fish throughout the investigations. But it has not changed significantly since the interception of the flow in 1981. Whereas the distribution obtained in the upstream before damming differs from that after damming, shifting towards bigger fish (Table 3).

Percentage of mature individuals and sex-ratio

The proportion of sturgeon with mature gonads fluctuated from 60% in 1986 to 45% in 1989 (Table 4). It was lower during the first three years after the dam's construction, 13,5% in 1981, 12,5% in 1982 and 25,4 in 1983 (Zhao Yan, 1986).

Table 2- Weight frequency distribution of male and female in the spawning population of A.sinensis

	Male			Female		
Weight (kg)	1981 1982	1986 1987	Total	1981 1982	1986 1987	Total
50	--	3	3	--		
60	4	4	8	--	--	
70	10	9	19	--	--	--
80	14	8	22	-	--	--
90	22	7	29	--	--	--
100	15	6	21	--	--	--
110	10	4	14	-	1	1
120	9	5	14	--	--	--
130	4	4	8	--	2	2
140	6	--	6	1	3	4
150	4	1	5	--	3	3
160	3	--	3	8	1	9
170	--	--	--	--	4	4
180	1	--	1	11	5	16
190	1	--	1	--	1	1
200	1	--	1	10	2	12
220	--	--	--	14	4	18
240	--	--	--	15	--	15
260	--	--	--	13	5	18
280	--	--	--	7	3	10
300	--	--	--	6	1	7
320	--	--	--	4	--	4
340	--	--	--	5	--	5
360	--	--	--	2	--	2
380	--	--	--	--	--	--
400	--	--	--	2	--	2
420	--	--	--	3	--	3
Total	104	51	155	101	35	136
Mean	98	90		238	191	
n-1	29	24		62	50	

Table 3 - Chi-square (X2) test for the body-length frequency distribution of A.sinensis.(* P <0.05)

	X2 (male)				X2 (female)			
	1972 1975	1981 1982	1986 1987	1988 1989	1972 1975	1981 1982	1986 1987	1988 1989
1972-1975		21.82*	24.41*	34.68*		13.38	32.89*	27.31*
1981-1982			14.99	12.66			15.25	18.30
1986-1987				10.52				14.22
	X20.05 = 16.92 (df = 9)				X20.05 = 22.36 (df = 13)			

Table 4 - Sex ratio and percentage of mature Acipenser sinensis captured below the dam.

Year	Sex	Num.of Samples	Gonadal development Stage				Mature Fish (%)	
			III	IV	V	VI	By sex[1]	Total
1986	f[2]	30	12	13	5	-	60%	-
	m[3]	32	13	8	10	1	59%	60%
	Sex ratio 1 : 1.1							
1987	f	32	7	25	-	-	78%	
	m	42	9	26	7	-	79%	78%
	Sex ratio 1 : 1.3							
1988	f	28	16	12	-	-	43%	
	m	45	19	22	4	-	58%	52%
	Sex ratio 1 : 1.2							
1989	f	31	22	9	-	-	29%	
	m	49	22	27	-	-	55%	45%
	Sex ratio 1 : 1							

1- from stage IV and over;
2- female;
3- male

These low percentages can be accounted for two facts: the first-one is, before the dam's completion a lot of sturgeon migrated upstream, 827 and 558 fish were captured respectively in 1981 and 1982. The second-one is, most of them could not have a normal development of their gonads as they were blocked in freshwater. After the strict enforcement of the restriction taken about commercial fishing all along the stream in 1983, the percentage had risen approximatly to 50%. During spawning the catch rate of the male was apparently higher than the one of the female. This fact could be explained by two reasons, firstly the males are more active than the female during spawning, so they have more opportunity to encounter the hooks. Secondly, as they take part several times in reproduction, they spent more time in spawning ground, so their probability to be caught is higher than that of females.

Nevertheless, the sex-ratio of unmature sturgeon with stage III gonads was relatively stable, approximating 1:1. Also the data obtained in 1981 and 1982 indicated that the sex-ratio of the spawning population approached 1:1 (Deng Zhonglin, 1985). Because unmature individuals represented over 85% of the total catches in those years, the high catch rate of mature males did not actually modify the ratio.

Age determination

In 1981 and 1982, 111 females and 107 males were used for age determination. Youngest female was 14 and oldest was 33. Most fell between 17 and 26 years old with a mean of 21,5. Youngest male was 9 and oldest 26. Most fell between 10 and 16 years old with a mean age of 13,5.

In 1986 and 1987, 29 females and 39 males were used for the age determination. The youngest female was 14 and the oldest 30. Most fell between 14 and 23 years with a mean age of 18,9. The youngest male was 9 and the oldest 20. most fell between 10 and 15 years with a mean age of 14,7.

First spawning

From the 101 rays collected in 1981 which were examined for this purpose, 56 came from male and 45 from female. The males being at their first reproduction were 47 (84%) with an age in the range of 9 - 17 years. The others, having spawned more than once have an age in the range of 16 - 26 years. From of the 45 females, 34 (76%) were first spawners and have an age in the range of 16 - 26, instead of 24 - 30 for the other female having spawned more than once.

In 1982, we have got, 44 were males and 35 females. 40 males were first spawners (9-18 years), the 4 others (18-26years) had spawned more than one time. Among the female, 30 were first spawners (14-26years) and the 5 others had been repeadly mature (23-33years).

DISCUSSION

Length and weight

The frequency distribution of both body length and weight has shift slightly towards smaller fish. But they did not change significantly. Table 1 shows the body length distribution obtained from 1972-1975 in the upper reaches of Yangtze before damming. There also were no significant changes compared with data recorded in the 1980's.

Age determination

It can be seen from a comparison of these two samples, 1981-1982 and 1986-1987, that there is no significant difference with regard to the age range distribution and mean age.

Age at first spawning

The reason for forming the "belt" is complicated. *Acipenser sinensis* does not feed during spawning migration which takes more than one year. The male lose about 20% and the female 40% of their total weight after spawning. We think that the "belt" close to the first one can not be taken in account as a reproduction mark. Anyhow , the results presented here could not be accurate. But it is certain that the first mature sturgeon took a majority of the spawning population. This may reflect an important characteristic of the population. If so , the population would be very vulnerable to over-catch.

CONCLUSION

Up to now since the construction of the dam ,the spawning population structure of the sturgeon did not change significantly. The population seems to be stable. So limited exploitation is suggested by researchers. We have known that the sturgeon needs quite a long time to reach maturity, at least 9 years for males and 14 for the females. Individuals spawning below the dam so far were mostly born before damming, thus it would be necessary to evaluate the spawning population in the following 5 or 6 years. Furthermore , the new spawning beds which have been confined in comparatively much narrower areas are located mainly in navigation channel where shipping and dredging make the spawning environment even worst. And its population size has not been well known so far. Though it is certain that the dam would not threatened the existance of the species, and the prohibition of commercial fishing should be executed successively.

During the construction, many discussion about building a fish-pass arose hot debates at every sitting of the attestation conference for the salvation of the sturgeon.

As the efficency of the fish-pass built for such large fish still remains in doubt and the sturgeon has less fishery value along the Yangtze, we standed for that no fish-pass should be established on the dam in order to avoid a waste of large amount of money. The survey conducted in 1981 and 1982 after the interception of the flow proved the existance of the fundamental spawning condition below the dam and the fish had reproduced in those years. These evidence provided scientific basis for planning salvation measures. Instead of establishing a fish-pass, the restrictive prohibition of commercial fishing and young sturgeon release were settled as the main protective measures. On the basis of the results observed up to now, the measures are suited for the conservation of the species.

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ANALYSIS ON *Acipenser sinensis* SPAWNING GROUND AND SPAWNING SCALES BELOW GEZHOUBA HYDRO-ELECTRIC DAM BY THE MEANS OF EXAMINING THE DIGESTIVE CONTENTS OF BENTHIC FISHES

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RESUME

Une série d'études sur l'écologie de la reproduction de l'*Acipenser sinensis*, ont été conduite à la section Yichang en aval du barrage de Gezhouba de 1981 à 1986. Les résultats d'études montrent que les conditions hydrologiques et les caractéristiques du lit du fleuve à la section Yichang correspondent aux conditions essentielles pour la reproduction naturelle de l'esturgeon zhonghua. Généralement les activités de fraye ont lieu lorsque la température se situe entre 17 et 19,5° C. Il n'y a pas de différence fondamentale de la température de l'eau, de la teneur en matière en suspension et du niveau de l'eau entre les frayères de Yichang et la frayère traditionnelle, durant les saisons de reproduction. Des activités de reproduction ont été observées à la section de Yichang, chaque année de 1982 à 1986, cela prouve que l'esturgeon peut s'y reproduire naturellement. La stabilité et l'importance de l'efficacité naturelle supplémentaire de la nouvelle frayère de la section de Yichang sont évaluées sur la base des faits suivants : le nombre d'oeufs d'esturgeon récoltés et le nombre de jours consécutifs durant lesquels les oeufs d'esturgeons peuvent être prélevés dans l'intestin de poissons benthiques, la proportion d'esturgeons matures sur le total des captures, l'ampleur des aires de reproduction et l'abondance d'esturgeons juvéniles dans l'estuaire du fleuve Yangtsé au printemps et à l'été de l'année suivante.

Mots clés : *Acipenser sinensis*, frayère, ponte, hydrologie, poissons prédateurs des oeufs.

ABSTRACT

A series of investigations on reproduction ecology of *Acipenser sinensis* was conducted at the Yichang section below the Gezouba Dam from 1981 to 1986. The results show that the hydrological environment and the features of the river bed at the Yichang section correspond with the essential condition for natural reproduction of the sturgeon. Generally the spawning activities take place while the temperature is in the 17-19.5° C range and there is no fundamental difference in water temperature, siltcontent and water level between the Yichang spawning ground and the traditional spawning ground during reproduction seasons. As the spawning activities were observed at the Yichang section each year from 1982 to 1986, it proved the sturgeon could naturally reproduce. The stability and the natural incremental efficiency of the new spawning ground at Yichang section and its spawning scales are assessed on the basis of the following facts : the number of eggs collected and consecutive days during which time the sturgeon eggs can be collected from the intestine of the benthic fish, the proportion of mature sturgeon among the total catches, the range of spawning grounds and the resources of the juvenile sturgeon at the estuary of the Yangtze river in spring and summer of the following year.

Key-words : *Acipenser sinensis*, spawning ground, spawning, hydrology, egg-eating benthic fish.

INTRODUCTION

Acipenser sinensis Gray 1834 commonly call Zhongua sturgeon is a large migratory species. The breeders travel annually during June and July from the sea up into the Yangtze river with their gonads gradually maturing, and continue their trip upstream to spawn. However, due to the construction of Gezhouba hydro-electric dam on the river (Fig. 1), these fish have been confined below the dam, unable to reach their traditional spawning sites upstream. The impact of the damming on the fish has so far catched the widespread attention of both biologists and the public because of its uniqueness and its potential economic value.

Acipenser sinensis has been the subject of many investigations since 1981 when the first survey on the reproduction's ecology of the fish was conducted within the river section from Yichang to Shishou below Gezhouba Dam. Based on the morphological and histological characteristics of the fish's gonads, it was found for the first time that the gonads of the six breeders caught had reached stage IV, which demonstrated that the gonads of the sturgeon which are blocked below Gezhouba dam can normally develop to reach maturity. These findings led us to discover the natural reproduction of the sturgeon below Gezhouba Dam.

From 1983 to 1986, further investigations were carried out on the new spawning ground of this species below Gezhouba Dam and the spawning activities were observed every year during the period in Yichang section. This work have been realized in connection with the one of Deng Xin et al.(1990), about the spawning population's characteristic.

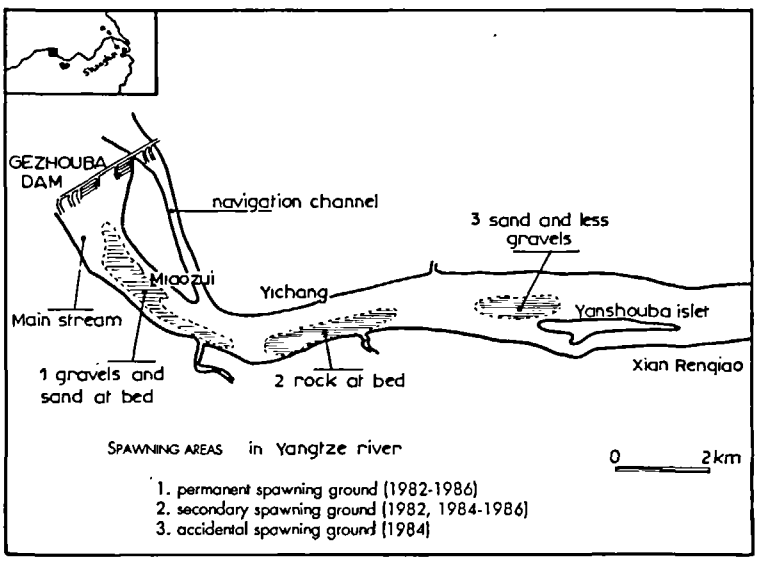


Figure 1 - Portion of spawning ground of Acipenser sinensis downstream from Gezhouba Dam.

MATERIALS AND METHODS

The fertilized eggs released are adhesive and stick tightly to gravel bars and rocks at the bottom of the river bed. As soon as the eggs' adhesive force has disappeared, they are dispersed among gravel bars, continuing their development. After hatching, the larvae either remain among the gravel bars or drift to slowly moving waters to continue their growth.

The spawning time and place can be reliably determined by the eggs stuck to gravel bars in the river bed. However, because of the strong current in the Yangtze River, the sturgeon eggs are extremely difficult to collect from gravels or artificial substrata, as they are swept off in the course of being lifted out of the water. Thus we decided to use an original method which consist in observation of digestive contents of benthic fish.

Over a 5 years period we inventoried 6 species of benthic fish which devour sturgeon eggs: *Coreius guichenoti* Sauvage et Dabry,1874; *C. heterodon* Bleeker,1865; *Leiocassis longirostris* Günther,1864; *Pseudobagrus vachelli* Richardson,1846; *Rhinogobio cylindricus* Günther,1888; and *Rh. ventralis* Sauvage et Dabry,1874.

C. guichenoti inhabits the area of strong current, so the spawning waters of the sturgeon often correspond to the former's living field. While the sturgeon is spawning, the gathering of this fish is enhanced and fishermen can capture a great amount of them. Sturgeon eggs can usually be found by examining its digestive contents. *C. heterodon* lives in the "secondary" fast flow waters and the number of sturgeon eggs it devours is generally less than those of *C. guichenoti*. The others normally live in a slow current.

Most of the devoured sturgeon eggs from the intestine of benthic fish, as far as we noted, were in a half-digested state, some with only a membrane left. But a small number of them located at the entrance of the digestive system were intact. The development stage of the fertilized eggs can then be determined by stripping the membrane under a microscope. It is known that the eggs can hatch after 113-130 hours when the water temperature is in the 16.5-18°C range (Anonymous, 1988). Thus, with the embryonic stage, the water temperature data and the time at which the benthic fish are caught, the spawning time of the sturgeon can be deduced (Tab. 1). Drift nets having a stretch mesh of 5.5 cm, a depth of 120 cm, and a length of about 70-90 m are used to capture benthic fish. They are lowered close to the bottom of the water and drift down the flow with a boat. The Yichang fishermen operate in a fairly regular area of about 1-2 km, taking normally 20-30 minutes . So the spawning site of the sturgeon can usually be defined by the location in which the egg-eating benthic fish are caught.

We consider that it is difficult to compare the yearly abundance of egg-production with only one index. So a comprehensive index has been adopted to judge approximatly the respective spawning scales of each year. This comprehensive index is based on : the number of consecutive days during which time the sturgeon eggs can be collected from the intestines of those egg-eating fish after each spawning, the species of the egg-eating benthic fish, the proportion of mature fish with the gonads of stage IV among total catches, the range of the spawning ground and the number of sturgeon fingerlings captured at the estuary of the Yangtze River in the spring and summer of the following year.

All the hydrological information on water temperature, flow velocity, water level, and silt content are furnished by the Yichang Hydrological Station under the Yangtze river Valley Planning Office (YVPO).

RESULTS

Reproduction periods and areas

Since the first observation of the sturgeon's natural reproduction in 1982, the fish's spawning activities below the dam were also recorded during the next five years by the examination of the intestinal contents of the dissected benthic fish. In 1984, 4 larvae were successfully hatched from the eggs freshly devoured by a *Pseudobagrus vachelli*, while in 1985 a still living larvae was obtained from the intestine of a *Coreius guichenoti*. Table 1 shows the spawning date and area below the dam from 1982 to 1986.

During those 5 years of investigations, the earliest spawning occured on Oct. 14 and the latest on Nov. 13. The regions where the sturgeon eggs were collected each year are shown on table 1 and Fig. 1. The eggs were collected from the Changjiang Ship-building Plant (CSP) to Miaozui in 5 years and also in the area of Mojishan hill except in 1983. It is thought that the two sections above are the spawning ground. The former may be called a "permanent spawning ground" and the latter "secondary spawning ground".

Additionally, a few sturgeons have reproduced at the area of the Yanshouba Islet 9 km below the dam (in 1984) and in the region of the Jiangjunmao 15 km down from the dam (in 1986) when the water level was high (Fig. 2). These were named tentatively the "accidental spawning grounds".

Table 1- Spawning date and area of Zhonghua Sturgeon below GEZHOUBA Dam from 1982 to 1986

Year	Collecting dates of eggs (date)	(hour)	Developmental stage of eggs	Water Temp. (° C)	Time from fertilizing (hour)	Spawning Date	Catch's area of benthic fish
	Nov.1	17:00	mid-gastrula	18.7	20	Oct.31	Mojishan section
1982	Nov.6	6:00	32-64 cells	18.6	8	Nov.5	same as above
	Nov.9	6:00	blastula	18.2	10	Nov.8	Wan Shouchiao CSP*
1983	Nov.8	6:00	early gastrula	17.9	22	Nov.7	Shilihong CSP
1984	Oct.19	8:00	neurala	18.7	40	Oct.17	Yanshouba CSP
	Nov.14	17:00	cleavage-blastula	17.0	8	Nov.13	Wulong CSP
1985	Oct.15	17:00	blastula early	19.5	20	Oct.14	Wan Shouqiao CSP
	Nov.7	8:00	4 cells-blastula	17.5	2-10	Nov.7	Mojishan CSP
1986	Oct.22	9:00	mid-gastrula	18.2	20	Oct.21	Jiangjunmao CSP

* Changhang Shipbuilding Plant

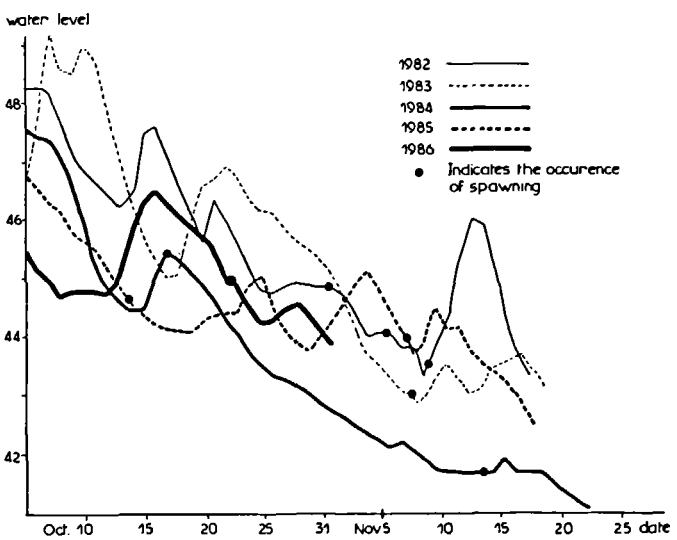


Figure 2 - Fluctuation of water level at Yichang during Acipenser sinensis spawning.

Annual spawning scales

3040 eggs were collected for the first time below the Gezhouba Dam, during three spawning runs in 1982. The bentic fish which eat the sturgeon eggs can only be found within two days after each spawning, 3 at most. *Coreius guichenoti* was the only egg-eating benthic fish that was caught within a section of 7 km. This suggested that the sturgeon had a small spawning scale during each spawning run that year.

In 1983, 338 eggs were collected during a spawning run. The two egg-eating species (*Coreius guichenoti, Pseudobagrus vachilli*) were caught within two days after the spawning between Changhang Shipbuilding Plant and Miaozui section. Among 76 *Coreius guichenoti* dissected only 3 were found with the sturgeon eggs. This again showed a small spawning scale in the year.

During the period from 1984 to 1986, 5 spawning runs were recorded. More than 4000 eggs were collected after each spawning. By dissecting benthic fish, we noted that the eggs could be continually found in their intestines within 4-6 days after each spawning. It can be estimated that most of the eggs, devoured by the benthic fish which were caught on the 4th 6th day after each spawning, had approached or almost reached hatching. This fact demonstrated a large spawning scale, a longer period of spawning activity and the increased chances for the eggs to be devoured. Based on the areas in which the benthic fish were caught, it is believed that the spawning ground had extended to 9 km downstream. The additional support for this belief could be the fact that 4 larvae were successfully hatched from the eggs freshly eaten by a *Pseudobagrus fulvidraco* Richardson,1846 caught at Yanshouba (Fig. 1).

In 1986, the spawning ground was further extended to 15 km downstream (Table 2). If we include Hu Yiatan section within the range of Yichang spawning ground (a breeder was caught there) then it can be assumed that the spawning area covers a section of 20 km.

Table 2 - Some Index of Egg-productivity of Acipenser sinensis Below Gezhouba Dam from 1982 to 1986

Year	Spawning Date	Range of egg-devouring fish collected (km)	Num. of species	Duration of eggs collected (days)	Num. of eggs collected	Total Num. of fish caught
1982	31/10	3	1	2	445	43
	5/11	3	1	2	3	21
	8/11	7	2	3	2592	73
1983	7/11	4	2	2	338	76
1984	17/10	9	5	5	4027	232
	13/11	6	5	4	4001	339
1985	14/10	7	6	6	5645	324
	7/11	5	6	6	7555	513
1986	21/10	15	5	6	5233	628

Riverbed condition and hydrological environment

From the end of the 1960's to the 1970's, a series of surveys on the hydrological conditions of *Acipenser sinensis*' spawning ground in the upper reaches of the Yangtze River were initiated by the fisheries' research ecologists. The results have revealed that the ideal sites for the sturgeon's spawning are those of fast flowing narrow river troughs with rocks piled at the upper end and rocks and gravel at the lower end (The Changjiang Aquatic Resources Survey Group, 1988).

The information provided during this work by YVPO show that the bottom of the river section between Changjiang Shipbuilding Plant to Juanhechiao is mainly composed of gravel bars and rocks.

In this section of the river, the main course runs close to the south bank with the Bija and Guanyin Hills jutting into the river along the way. Pits of various depths and reefs can be found in the trough. The right side of the river bottom between Shilihong to Mojishan comprises a mixture of gravels, sands and rocks with the water depth of 8-26 m. Most of gravels and rocks are of a diameter size of 3-10 cm. In Yanshouba section, there is a mixture of cobblestones and sands with fast water and an unstable flow.

Downstream from the Dam, the availability of the gravels and rocks for the sturgeons to release their eggs upon and the unstable water flow which is favourable for the dispersal of the eggs meet all the essential conditions of Zhonghua Sturgeon spawning grounds. It was noted that the water fluctuation did not have any significant influence on the spawning activity of the sturgeon in the area of Yichang spawning ground. 4 ebbs, 3 rises and 2 stable water levels were recorded during runs (Table 3). During the survey made from 1971-1975 at Sankuaishi spawning ground in the upper reaches each years from the middle of Oct. and Nov., we recorded some water fluctuations: water temperature in the 17.0 - 20.1°C range ; maximum flow velocity within 2.68 and 3.03 m/s with the mean flow velocity ranging between 1.82 and 2.42 m/s and a silt content of 0.375 to 0.550 kg/m^3 ; all the spawning activities taking place at the time of ebbing.

Table 3 - Hydrological characteristics during spawning period at Yichang section from 1982 to 1986

Year	Spawning (month)	date (day)	Water temp. (°C)	Siltcontent (kg/m^3)	Rise and fall of water level (cm/day)	Flow velocity (m/sec.) mean	max
	10	31	18.7	0.287	3	1.23	1.64
1982	11	5	18.6	0.413	4	1.17	1.56
	11	8	18.2	0.287	13	1.03	1.40
1983	11	7	17.9	0.192	-	0.98	1.30
1984	10	17	18.7	0.430	39	1.42	1.72
	11	13	17.0	0.170	-	0.82	1.20
1985	10	14	19.5	0.670	22	1.31	1.67
	11	7	17.5	0.270	41	1.15	1.52
1986	10	21	18.2	0.890	28	-	-

If we compare the above hydrological data to the ones from 1982 to 1986, we can notice a basic correspondence between the two spawning areas in water temperature, silt content and fluctuation during spawning seasons with the only exception of flow velocity. From this information that water temperature might be an important factor for the spawning of sturgeon ; that is to say a water temperature above 17.0° C would be required to trigger a spawning activity.

DISCUSSION

Capability of Acipenser sinensis in reproducing naturally below Gezhouba Dam.

It has been proved since 1981, that the sturgeon gonads can develop normally to reach the stage IV below Gezhouba Dam. The discovery of the sturgeon eggs and

larvae collected below the dam in 1982 indicates that like other *Acipenseridae* species (Vladimov, 1957) when their migratory route is blocked, the zhongua sturgeon can still adapt to and reproduce in new habitats when provided with suitable hydrological conditions for spawning.

In the first two years after the damming, the breeders trapped below the dam represent 827 fish in 1981, and 558 in 1982. These catches are almost six time higher than the average catch in the same river section before the damming while the total catches along the stream during that period averaged about 400-500 fish (The Changjiang Aquatic Resources Survey Group, 1988). This may account for a much lower proportion of mature sturgeon in the following year (12.5% in 1982 ; 25.4% in 1983)(Deng Xin et al., 1990). However, the juvenile fish could still be collected each year at the estuary of the Yangtze River, despite the rapid depletion in the breeders population. The number of the young sturgeons caught by Yuan Fishing Team with barrier nets in certain areas at the mouth of the river showed that there were more juvenile fish present at the mouth of river after 1982 (137 fish were collected in 1982, 477 in 1984 and 571 in 1985) (Zhao Yan, 1986). So the common opinion held by the early investigators that all the eggs have been devoured by benthic fish in an altered habitat should be reconsidered. From 1982 to 1985, 5 spawning activities were observed consecutively in the Yichang section. These observations asserted that suitable hydrological environments which meet the required conditions for *Acipenser sinensis* to mature and to reproduce naturally exist below the Gezhouba Dam. In the future, the incremental efficiency of Yichang will be gradually increased with the execution of the sturgeon Protection Act, and by maintaining a stable environment for the fish.

Recommendations

Acipenser sinensis has a late sexual maturity, it requires 9 years for the male and 14 years for the female. Undoubtedly unrestricted commercial fishing will lead to a disastrous decline in the sturgeon stock, which will be impossible to recover. So the protection of spawning ground and the maintenance of the fish's self-sustaining reproductive populations should be our primary concern both at present and in the future.

For the protection of Zonghua Sturgeon spawning ground at Yichang, we recommend :

1) prohibit quarrying along Yichang river section ;
2) close the section between Miaozui and Changjiang Shipbuilding Plant to commercial fishing to preserve the spawning ground ;
3) impose regulations on the use of barrier nets and any other fishing gears to which the young sturgeons are vulnerable ;
4) limit ship speed in Yichang section to create a peaceful environment ;
5) prevent any kind of netting at the estuary of the river between late March and early July, when the young sturgeon congregate in great density in the area, in order to allow the young to grow in the sea.

Advocation on research

The information and the scientific data on the sturgeon's reproduction populations are still lacking. By blocking the migratory route of *Acipenser sinensis* the Gezhouba Dam has had a negative impact on the sturgeon population. We have been persistent in our proposal for developing artificial propagation techniques to maintain its population. Progress in the same field has also been made in China. After the successful experiment on the artificial propagation of the sturgeon conducted by the concerned fishery agencies and the Gezhouba Artificial Propagation Research Institude in 1983 and 1984, 4 releases have been made in the next four years. This has paved the way for a large-scaled release of the artificially propagated sturgeon larvae in the future.

The current problem facing us now is that the survival rate of the artificially propagated juvenile sturgeon is low in winter since the sturgeon is a fall spawner, therefore, we have a great task before us to carry on the research in the fields of artificial propagation, fish diseases, feed, management, and artificial reproduction by controlling fish's gonads development .

We firmly believe that with the implementation of the protective measures the *Acipenser sinensis* population will have a bright outlook.

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DONNEES ACTUELLES SUR LA BIOLOGIE D'*Acipenser sturio* DANS L'ESTUAIRE DE LA GIRONDE

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RESUME

L'esturgeon Acipenser sturio qui est une espèce menacée a fait l'objet d'une étude dans le bas estuaire de la Gironde de 1981 à 1988. Cette étude a essentiellement porté sur la population de juvéniles (25 à 145 cm environ) qui nous renseigne sur l'existence de reproduction dans le milieu naturel (recrutement) et les potentialités en géniteurs qui ne sont plus capturés accidentellement qu'en quantité très faible. Entre 1981 et 1988, 1847 individus ont été marqués avec différents types de marques et 687 recapturés, dont 62 en mer. La croissance effective mesurée sur un échantillon de 82 poissons marqués et recapturés environ un an plus tard est nettement supérieure à celle déduite des travaux de Magnin (1962) entre 65 et 105 cm environ. L'analyse des effectifs montre une forte proportion d'individus de taille comprise entre 72 et 114 cm et un vieillessement de cette population de juvéniles. Une première estimation du stock de juvéniles par la méthode de Jolly (1965) est tentée. Le schéma migratoire déduit de nos résultats est légèrement différent de celui décrit par Magnin (1962). Dès l'âge d'un an les juvéniles rejoignent le groupe qui effectue la remontée saisonnière dans l'estuaire et regagnent la mer avec eux. Vraisemblablement, les juvéniles ne font pas que séjourner entre mai et octobre dans l'estuaire ; ils effectuent des va-et-vient entre mer et estuaire. L'aire de répartition en mer est beaucoup plus étendue que ne le laissaient supposer les données anciennes. Les perspectives de restauration de l'espèce sont brièvement discutées en conclusion.

Mots clés : estuaire, *A. sturio*, juvéniles, marquage, croissance, migrations, estimation de stock.

ABSTRACT

The sturgeon (Acipenser sturio) which is an endangered species has been investigated in the Gironde Estuary from 1981 to 1988. The studies have been realized in the low part of the Gironde Estuary and have concerned mainly the juvenile population (about 25 cm to 145 cm). The juveniles provides information on the existence of reproduction in the natural environment (recruitment) and on potential genitors which are only caught accidentally in a very limited quantity. From 1981 to 1988, we tagged 1847 individuals with several types of tags and registred 687 recaptures, 62 of which were made in the sea. The effective growth, measured on a sample of 82 fishes tagged and recaptured about 1 year later is distinctly more important than the one deducted from Magnin (1962) between 65 and 105 cm. The analysis of the frequency distribution shows a high proportion of individuals which size ranged from 72 to 114 cm and an ageing of this juvenile population. A first estimate of the juvenile stock is given, using Jolly's method. The migratory cycle deducted from our results is lightly different from the one described by Magnin (1962). The juveniles join the group who makes the seasonal migration upstream, right from the age of one year, and get back to the sea with them. Probably, the juveniles don't just stay in the estuary between may and october ; they come and go between sea and estuary. The sea distribution area extends way beyond the French coast. The prospects of restoration of the species are briefly discussed in a conclusion.

Key-words : estuary, *A. sturio*, juveniles, tagging, growth, migrations, stock assessment.

INTRODUCTION

Situation de l'espèce et cadre de recherche

L'esturgeon *Acipenser sturio* était recensé dans la plupart des mers et des grands fleuves d'Europe au début du siècle ; en 1962 il était encore présent dans le Guadalquivir (Magnin, 1962). Actuellement, on ne le trouve plus, de façon certaine, que dans le Rioni (Ninua, 1976), qui se jette en mer Noire et dans le bassin

Garonne-Dordogne, où sa migration de reproduction s'arrête actuellement au niveau de Golfech et de Bergerac (figure 5).

L'esturgeon a toujours été recherché pour sa chair dans les grands cours d'eau français. Mais c'est seulement au début du siècle que les ovules des femelles mûres furent récupérés pour la fabrication du caviar. Ainsi, de 1920 à 1975 environ, le bassin Garonne-Dordogne a fourni le seul caviar français. La production atteignait 8 à 10 tonnes environ pour une centaine de tonnes de poissons dans les meilleures années ; elle était encore de 3 tonnes en 1950, pour 50 tonnes de poissons. Elle a ensuite fortement chuté puisqu'on totalisait juste 0,250 tonne de caviar pour 200 poissons environ en 1963 et elle est devenue pratiquement nulle en 1980.

Le même déclin des captures a été observé en mer par Letaconnoux (1961) ; les débarquements des chalutiers de La Rochelle enregistrés en criée sont passés de 275 individus environ en 1936 à 120 en 1948, puis à 14 en 1956.

Les causes probables de cette régression sont multiples : pêche trop intensive des immatures et adultes, réglementation inadaptée, contrôles déficients, dégradation du milieu (zones de nourricerie et de fraie) par les extractions de granulats, diminution de la qualité de l'eau liée aux pollutions physico-chimiques et organiques, limitation des migrations de reproduction par les barrages.

A partir de 1980, des mesures de protection de l'espèce ont été prises et un programme de restauration a été lancé selon deux axes :

- acquisition de connaissances biologiques sur l'espèce dans le milieu naturel et suivi des stocks ;

- production d'alevins en vue du repeuplement.

Nous ne nous occupons ici que du premier axe de recherche.

Rappel sur les travaux antérieurs et objectifs

Magnin (1962) a étudié l'âge et la croissance d'*Acipenser sturio* à partir de la lecture des coupes de rayons pectoraux de plus de 300 spécimens mesurant entre 34 et 255 cm (longueur totale). Il a établi des courbes de croissance linéaire et pondérale.

Il indique que la majorité des mâles deviennent matures lorsqu'ils atteignent une longueur de 145 cm, vers 14-15 ans et la majorité des femelles, lorsqu'elles atteignent une longueur de 185 cm, vers 20-22 ans.

La seule phase du cycle biologique d'*Acipenser sturio* que Magnin (1962) a pu déterminer avec certitude est la migration de reproduction :

février-mars :	montée de quelques précurseurs
avril-mai :	pénétration dans l'estuaire et migration anadrome
10 mai - 10 juin :	ponte sur les frayères dans les fleuves
juin :	retour vers la mer
juillet :	il ne reste presque plus de géniteurs dans le fleuve.

Pour les autres phases du cycle biologique d'*Acipenser sturio* concernant les périodes alevin et juvénile, Magnin (1962) s'est appuyé sur des "sondages" qu'il a

effectués sur les fleuves, l'estuaire et l'embouchure, sur des témoignages de pêcheurs et sur les données de Roule (1922) et Letaconnoux (1961). Voici ses conclusions :

"Les alevins descendent très vite vers les eaux saumâtres et dès l'âge d'un an (36 cm), jusqu'à 3 ans (50 cm), ils se trouvent entre Mortagne et Talmont de novembre à février ; il y a une véritable migration saisonnière annuelle de la mer vers l'estuaire entre fin juin et fin septembre des immatures (50 à 145 cm) qui ne dépasse pas les Callonges ; ces immatures se trouvent en mer sur le plateau continental d'octobre à début juin".

Les données acquises sont irremplaçables du fait de la rareté actuelle de l'espèce et ces travaux constituent la base de nos investigations.

Cette communication présente les premiers résultats obtenus sur la croissance, les migrations et l'évaluation du stock des juvéniles.

MATERIELS ET METHODES

La rareté de l'espèce impliquait en premier lieu de localiser la population existante et en particulier le groupe de juvéniles (tailles de 25 à 145 cm environ) qui selon Magnin (1962) effectue une migration saisonnière annuelle de la mer vers l'estuaire appelée "Mouvée de la Saint-Jean".

C'est aussi l'étude de cette partie de la population qui pouvait nous permettre d'obtenir à la fois des informations sur l'existence des reproductions dans le milieu naturel (recrutement) et sur les potentialités en géniteurs qui n'étaient plus capturés qu'en quantités très faibles (quelques unités par an) lors des pêches des autres migrateurs.

Pour cela nous avons mis au point une méthode d'échantillonnage par pêche et une technique de marquage présentés dans ce chapitre.

Secteur d'étude

Il s'agit de la partie aval de l'estuaire de la Gironde formé par la jonction des fleuves Garonne et Dordogne. Ses limites, déterminées à partir des données de Magnin (1962), des connaissances des anciens pêcheurs d'esturgeons, des contraintes liées aux aménagements de l'estuaire et aux pratiques de pêche sont (figure 1) :

- les Callonges à l'amont, que les juvéniles ne dépassent pas dans leur remontée selon Magnin (1962) ;

- Royan à l'aval, port d'embouchure ;

- le chenal de navigation qui condamne la rive Médoc sur deux kilomètres de large environ.

Ce secteur d'une longueur de 45 km environ se situe donc côté Saintonge et couvre plus de la moitié de l'estuaire. Il est découpé en quatre zones de pêche, au départ des ports de Talmont, Mortagne, Maubert et Vitrezay.

La salinité croît en moyenne de l'amont vers l'aval de 5 °/₀₀ à 30 °/₀₀ avec des variations importantes en fonction du cycle des marées et du volume d'eau douce sortant. Ces eaux douces s'écoulent préférentiellement en rive Saintonge et les eaux salées pénètrent en rive Médoc.

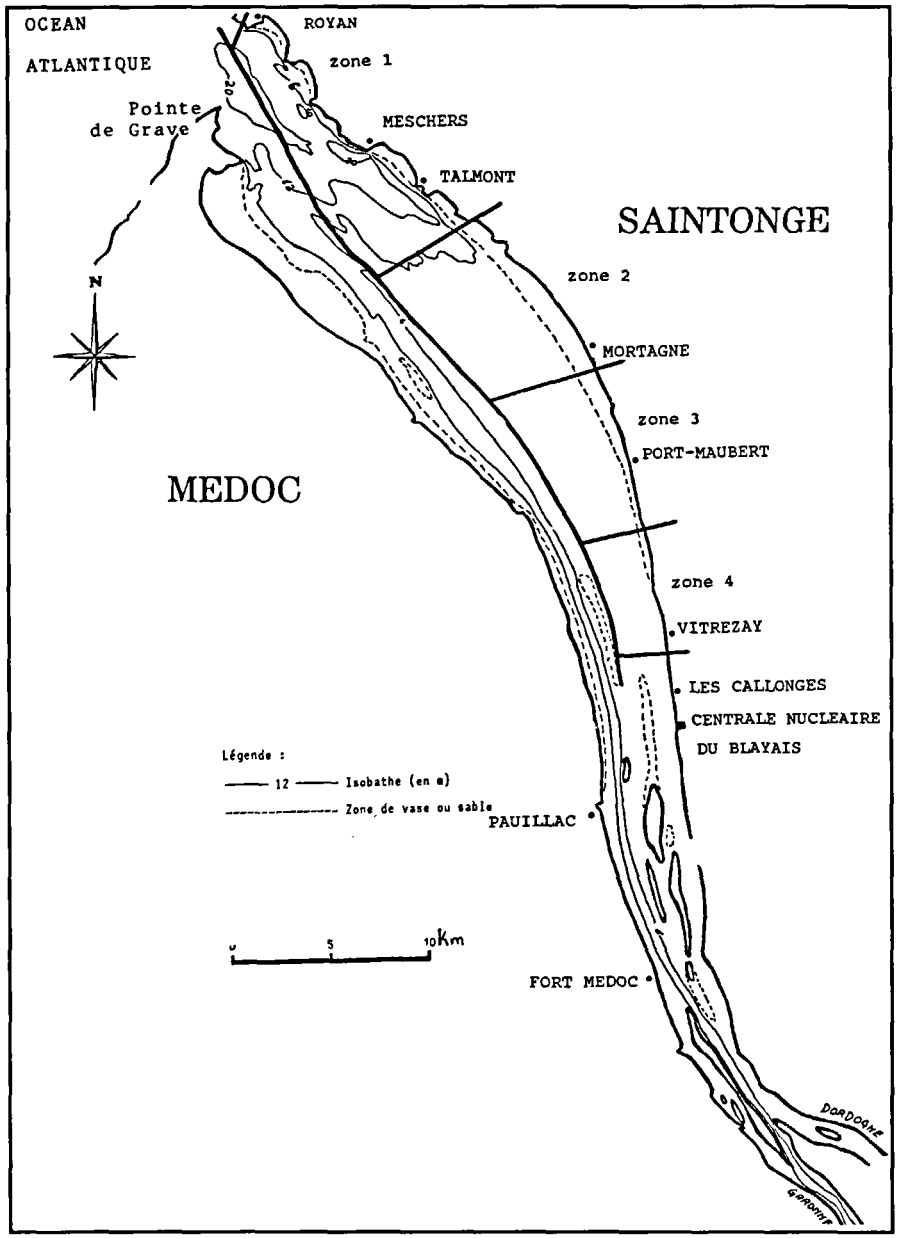


Figure 1 - Estuaire de la Gironde, secteur d'étude, zones de pêche.

Le débit de l'intrusion de la masse d'eau marine dépasse largement les débits cumulés des deux fleuves qui varient entre 300 m^3/s et 1200 m^3/s. Des valeurs de plusieurs milliers de m^3/s peuvent être cependant enregistrées en période de crue (Castaing, 1981).

La concentration moyenne des matières en suspension des eaux fluctue de 0,1 à 1 g/l ; elle atteint 10 g/l dans le bouchon vaseux qui se forme sous l'action de courants résiduels (Castaing, 1981).

La température de l'eau varie en moyenne de 8°C en hiver à 20-25°C en été ; on peut observer des différences entre surface et fond de 1 à 4°C.

L'estuaire de la Gironde n'est pas trop industrialisé ; c'est le seul estuaire français qui a conservé son ichtyofaune spécifique. Nous manquons cependant de données rigoureuses et exhaustives pour apprécier la qualité de l'eau.

- 60 à 70 marins-pêcheurs professionnels exercent toute l'année dans l'estuaire et une bonne partie d'entre eux se trouvent dans le secteur d'étude.

Stratégie d'échantillonnage

Deux types de pêche ont permis la capture, le marquage et la recapture de juvéniles :

- des pêches expérimentales où nous embarquions avec des pêcheurs professionnels rémunérés à cet effet. Les filets utilisés étaient des tramails dérivants de fond, de 120 m de long, de 2,50 m de hauteur et de 70 à 110 mm de côté de maille.

Le nombre de ces pêches pratiquées dans le secteur d'étude est donné au tableau 1 et le tableau 2 montre qu'elles ont été centrées sur les mois de mai-juin-juillet jusqu'en 1983 puis progressivement développées pour couvrir la période d'avril à août en 1984 et de février à novembre en 1988. Ces pêches ont eu lieu uniquement en zones 1 et 2 de 1981 à 1985 ; la zone 3 a été échantillonnée à partir de 1986 et la zone 4 à partir de 1987 ;

- des pêches commerciales ciblées sur d'autres espèces d'intérêt halieutique (*Alosa alosa, Petromyzon marinus, Argyrosomus regius, Dicentrarchus labrax, Solea solea*) qui occasionnent des captures accidentelles d'esturgeons. Pour chacune des quatre zones de pêche (figure 1), un pêcheur avait reçu la responsabilité de mesurer, de relever le numéro de marque ou de marquer ces esturgeons capturés accidentellement par lui-même ou par ses collègues (5 à 7 pêcheurs par zone).

Ces pêches commerciales ont lieu toute l'année et dans l'ensemble de l'estuaire accessible, mais les captures accidentelles d'esturgeons n'ont été enregistrées de façon exhaustive qu'à partir de 1984-1985, lorsque les pêcheurs ont vraiment coopéré à la suite de l'effort de sensibilisation et d'information engagé. Le nombre de pêches commerciales qui a donné lieu à au moins une capture accidentelle d'esturgeon est précisé au tableau 1.

On peut considérer qu'en 1986, 1987 et 1988 nous aboutissons à un échantillonnage correct du fait que les déficiences constatées dans la couverture spatiale et temporelle du secteur d'étude et de l'estuaire par les pêches expérimentales sont compensées par l'importance des pêches commerciales, correctement suivies.

Nous avons retenu comme unité d'effort de pêche, l'effort développé par un tramail en action de pêche pendant 2 h 30 mn en moyenne ; nous n'avons pas pu mettre en évidence de sélectivité en fonction des différents maillages utilisés.

Les captures par unité d'effort de pêche (CPUE) sont donc exprimées en nombre de poissons capturés par tramail pendant 2 h 30 mn. Elles sont calculées mensuellement uniquement pour les pêches expérimentales, à partir du nombre de captures (nombre de fois qu'un poisson est sorti de l'eau) ; sur une année le nombre total de capture (tableau 2) est supérieur au nombre total de juvéniles capturés (tableau 1), du fait que certains individus marqués les années antérieures ou dans l'année, peuvent être recapturés plusieurs fois successivement.

Marquage

Plusieurs types de marques ont été testés au cours de l'étude. Après chaque changement de système de marquage, les poissons recapturés avec l'ancienne marque ou ceux l'ayant perdue sont marqués à nouveau.

La perte d'une marque peut parfois être identifiée sur un individu car il subsiste une plaie maintenue ouverte par une partie de l'attache qui reste fixée ou bien un bourrelet caractéristique formé par la cicatrisation.

Le tableau 1 résume le marquage :

- de 1981 à 1983, 348 juvéniles ont été marqués pour la première fois avec des étiquettes pour salmonidés (ET) de Presadom (France) placées à la base de la nageoire dorsale ; ces marques sont difficiles à placer et sont très peu résistantes ; elles se cassent au niveau des fils d'attache, du noeud et du marqueur. Elles ont été abandonnées fin 1983 ;

- en 1984 et 1985, 693 juvéniles ont été marqués pour la première fois avec des disques Petersen (PT) de Neal and Son (Grande Bretagne) implantés avec une attache classique à la base de la nageoire dorsale ; ces marques sont solides mais les noeuds s'accrochent dans les mailles des filets et rendent les poissons plus vulnérables. Cela nous a conduit à transformer le système de fixation fin 1985 ;

- en 1986 et 1987, 687 juvéniles ont été marqués pour la première fois avec des disques Petersen montés sous la forme étiquette (PTB) fixés à la base antérieure de la nageoire dorsale par un fil métallique formant une boucle de 30 mm de diamètre environ, fermée par un noeud torsadé ; ce système de fixation a permis de limiter l'accrochage dans les filets, mais ne l'a pas éliminé complètement car celui-ci s'est reporté en partie au niveau du disque et du noeud de liaison. L'expérience acquise nous a conduit à rechercher un fabricant susceptible de créer une marque spécifique ;

- en 1988, 119 juvéniles ont été marqués pour la première fois avec des marques spéciales (HL) confectionnées par Hallprint (Australie) basées sur le principe de fixation par une boucle fermée par un noeud torsadé. Cette marque n'offre plus qu'une aspérité au niveau de la torsade et il semble que le phénomène d'accrochage soit négligeable, mais il est évidemment prématuré de poser un diagnostic.

Au total, si l'on excepte les poissons marqués recapturés auxquels nous avons changé leurs marques, nous avons marqué de 1981 à 1988, 1935 juvéniles (tableau 1) dont 88 avaient été identifiés comme ayant perdu leur marque. Ainsi, 1847 juvéniles vierges ont en fait été marqués pour la première fois en estuaire.

- 687 de ces poissons marqués ont été recapturés une ou plusieurs fois :

  - en estuaire :

    . 292 recapturés l'année de marquage ;
    . 241 recapturés, marqués les années antérieures ;
    . 88 marques perdues ;

  - en mer :

    . 62 recapturés.

Tableau 1 - Récapitulatif des pêches, du marquage et des recaptures des juvéniles en estuaire et en mer (1981-1988)

ANNEES	Type de Marque	Zone Estuaire	Nombre sorties Pêches		Juvéniles Estuaire					Juvéniles Mer
			Echantillonnage	Commerc.	capturés 2+3+4 (1)	vierges marqués (2)	marque perdue (3)	marqués antérieurement et recapturés (4)	marqués dans l'année recapturés (5)	marqués estuaire recapturés Mer (6)
1981	ET	1,2	2	0	43	43	-	0	0	0
1982	ET	1,2	2	10	62	61	-	1	4	2
1983	ET	1,2	10	13	252	244	3	5	26	1
1984	PT	1,2	22	15	252	228	16	8	10	4
1985	PT	2	25	14	532	465	29	38	65	12
1986	PTB	1,2,3	23	45	383	332	6	45	59	16
1987	PTB	1,2,3,4	30	19	468	355	16	97	93	16
1988	HL	1,2,3,4	68	13	184	119	18	47	35	11
				Total	2176	1847	88	241	292	62

ET = Etiquette
PT = Petersen
PTB = Petersen boucle
HL = Hallprint

Recueil d'information sur les captures

Une information sur nos études a d'abord été diffusée sous forme d'affiches, d'articles de presse, de plaquettes illustrées et de réunions, sur le bassin Garonne-Dordogne-Gironde et sur les côtes Aquitaine et Charentaise. A partir de 1984, cette information et les sollicitations de renseignements sur les captures d'esturgeons marqués ou non, ont été systématisées en France et dans les principaux pays européens concernés (Castelnaud, 1988). Une récompense a été attribuée pour tout retour de marque ou d'information sur un poisson marqué.

Mesures et prélèvements

Les poissons capturés ont été mesurés (longueur totale LT) au cm près ; à partir de 1988, ils ont aussi été pesés à 200 g près avec un dynamomètre à cadran et un prélèvement d'une tranche du premier rayon de la nageoire pectorale a été effectué pour la détermination de l'âge (Rochard et Jatteau in Acipenser 1989).

Croissance en taille et en poids

Sur l'ensemble des poissons marqués et recapturés lors des pêches expérimentales, nous sélectionnons ceux pour lesquels l'intervalle entre les recaptures successives est d'environ 12 mois et nous obtenons ainsi une croissance individuelle réelle. Nous avons comparé les croissances et la relation taille-poids obtenues à celles de Magnin (1962) avec le test U de Mann et Whitney. (Logiciel PCSM 1987 - Programme Conversationnel de Statistiques pour les Sciences et le Marketing. Deltasoft, Grenoble, France). La relation unissant la taille et le poids est calculée pour 1988 par régression moyenne géométrique (Ricker, 1980).

Migrations

La répartition et les mouvements migratoires de l'espèce sont étudiés à partir des résultats de pêche et des recaptures de poissons marqués.

Evaluation du stock

Afin de respecter les postulats principaux des méthodes d'évaluation de stock par marquage, nous ne pouvons estimer en toute rigueur que la partie de la population de juvéniles qui fréquente l'estuaire de la Gironde.

Nous n'avons pas utilisé la méthode de Petersen car elle suppose une relative stabilité dans le temps de la population que l'on veut estimer ; elle ne permet pas simultanément des entrées d'individus dans le secteur d'étude et des sorties. De plus les estimations ne se font qu'en considérant simultanément deux périodes consécutives et on ne peut pas obtenir de cette façon une estimation sur l'ensemble de la période de séjour des juvéniles en estuaire.

Nous avons donc choisi la méthode stochastique de Jolly (1965). Elle nous semble, pour l'instant, la plus adaptée (marquages individuels étalés sur de longues périodes, recaptures multiples, taux de survie variable dans le temps, émigration et immigration possibles). Celle-ci fait appel pour certains éléments de calcul à la méthode de Petersen affinée par Bailey (1951). On se reportera, pour les symboles utilisés, à Begon (1979).

La méthode de Jolly (1965) implique que chaque poisson marqué et recapturé, soit réutilisé dans les calculs avec la nouvelle date de capture.

Tableau 2 - Zones de pêche, captures, efforts, CPUE par mois pour les pêches expérimentales et captures accidentelles (x) par mois pour les pêches commerciales

		JANV.	FEV.	MARS	AVRIL	MAI	JUIN	JUIL.	AOUT	SEPT.	OCT.	NOV.	DEC.	Ens./an.
1981	Zones						1,2							
	Captures						43							43
	Effort						2							2
	C.P.U.E.						21.50							21.50
1982	Zones					1	1,							
	Captures					11	4 (16)							15 (52)
	Effort					1	4							5
	C.P.U.E.					11.00	1.00							3.00
1983	Zones						1	1,2			2			
	Captures						73 (89)	89 (26)			3			165 (115)
	Effort						15	5			4			24
	C.P.U.E.						4.87	17.80			0.75			6.87
1984	Zones					1	1	2		2	2			
	Captures					0 (10)	62 (93)	26 (11)		41	0 (21)			129 (135)
	Effort					6	29	18		7	5			65
	C.P.U.E.					0	2.14	1.44		5.85	0			1.98
1985	Zones				2	2	2	2				2		
	Captures				26 (3)	0 (13)	187(21)	323 (3)	(39)			4 (1)		540 (80)
	Effort				41	14	35	34				5		129
	C.P.U.E.				0.63	0.00	5.34	9.50				0.80		4.19
1986	Zones				1,2	1,2	1,2	2,3	2,3		2,3			
	Captures				6	11 (2)	267	35 (63)	0 (18)	(42)	0 (12)			319 (137)
	Effort				12	28	22	48	5		9			124
	C.P.U.E.				0.50	0.39	12.14	0.73	0		0			2.57
1987	Zones			1,2	1,2	1,2,3	1,2,3	1,2,3	1,2,3,4		4	2		
	Captures	(1)		1	1	426(24)	110(15)	4 (3)	0 (4)	(6)	0 (1)	0		542 (54)
	Effort			3	18	42	32	27	12		4	3		141
	C.P.U.E.			0.33	0.06	10.14	3.44	0.15	0		0	0		3.84
1988	Zones		1,2	2	1,2	1,2,3	1,2,3	1,2,3	2,3,4	2,3	2,4	1		
	Captures		0	0 (2)	14	49	49 (8)	69 (8)	24 (2)	4 (7)	1	0 (4)		210 (31)
	Effort		2	10	18	27	48	44	31	21	6	5		212
	C.P.U.E.		0	0	0.78	1.81	1.02	1.57	0.77	0.19	0.17	0		0.99

La clé des calculs repose dans l'estimation de M_i (nombre de marques encore en circulation à l'instant i) qui constitue la première étape de l'analyse. La seconde étape consiste en une estimation de la taille N_i de la population en i.

Les périodes de temps retenues pour regrouper les captures sont déterminées de façon à ce que les y_i (nombre d'animaux marqués et relâchés en i et repris par la suite) et z_i (nombre d'animaux marqués avant i, non encore repris en i mais recapturés par la suite) ne soient pas nuls, ce qui bloquerait les calculs ; ces périodes ne sont pas de durées égales.

Nota : L'ontogenèse d'*Acipenser sturio* étant mal connue, nous utiliserons, en fonction des données de Magnin (1962) et de nos observations, les appellations suivantes selon la terminologie de Balon (1975) :

- période alevin jusqu'à 25 cm environ, taille à laquelle les caractères morphologiques définitifs (nageoire caudale) sont mis en place ;

- période juvénile pour les individus de taille comprise entre 25 et 145 cm environ ;

- période adulte pour les individus de taille supérieure à 145 cm, tout en sachant que selon le sexe, la première maturation des gonades a lieu entre 145 et 185 cm.

RESULTATS ET DISCUSSION

Croissance en longueur et en poids

De 1983 à 1988, nous avons sélectionné 82 esturgeons marqués et recapturés environ un an plus tard. Le tableau 3 rassemble à titre d'exemple les 19 poissons retenus pour l'année 1988. Le temps écoulé entre le marquage et la recapture se distribue pour l'échantillon de 82 individus selon une loi normale de moyenne 365 jours et d'écart-type 18 jours. Les individus mesuraient lors du marquage, de 63 à 141 cm et lors de la recapture, de 80 à 143 cm.

Après avoir regroupé nos 82 individus d'après leur taille au marquage, dans les classes de taille correspondant aux classes d'âge IV à XI déterminées par Magnin (1962), nous avons calculé la croissance moyenne annuelle (tableau 4).

On s'aperçoit que pour les classes IV à IX, c'est à dire pour des tailles comprises entre 55 et 105 cm, cette croissance est importante ; elle s'échelonne entre 12,3 et 23,2 cm. Pour les classes X et XI, c'est à dire pour des tailles comprises entre 106 et 123 cm elle est moitié moindre.

Nous avons comparé cette croissance pour les classes V à IX et XI (tableau 4) à celle déduite des données de Magnin (1962) qui a travaillé sur des esturgeons provenant de la même zone.

La croissance que nous observons est significativement différente au seuil de 2 % (test U de Mann et Whitney) ; elle s'avère plus rapide pour les classes V à IX et semble plus lente pour la classe XI (figure 2).

L'analyse d'un groupe de petits individus apparus en fin 1988 et dont le détail est donné au § : "Répartition et mouvements migratoires", laisse penser que l'*Acipenser sturio* peut atteindre une taille de 50 cm en 1 an.

Tableau 3 - Poissons marqués, recapturés et sélectionnés en 1988 pour mesurer la croissance effective

MARQUAGE				1ERE RECAPTURE			
N°	Date	Taille en cm	Classe d'âges*	Date	Taille en cm	Classe d'âges*	Croissance en cm
HL 0028	07.05.87	81	VII	13.04.88	90	VIII	9
HL 0035	06.05.87	86	VII	14.04.88	92	" "	6
HL 0305	07.05.87	83	VII	20.05.88	93	" "	10
HL 0114	20.05.87	92	VIII	21.06.88	105	" "	13
HL 0338	19.05.87	94	" "	26.05.88	104	" "	10
HL 0027	07.05.87	95	" "	24.05.88	104	" "	9
HL 0316	17.06.87	95	" "	24.05.88	106	" "	11
HL 0317	19.05.87	95	" "	24.05.88	104	" "	9
HL 0316	21.05.87	96	" "	24.05 88	107	X	11
HL 0322	18.05.87	96	" "	25.05.88	112	" "	16
HL 0337	19.05.87	97	" "	26.05.88	108	" "	11
HL 0314	21.05.87	103	" "	24.05.88	114	" "	11
HL 0321	19.05.87	103	" "	25.05.88	122	" "	19
HL 0328	18.05.87	103	" "	25.05.88	117	" "	14
HL 0323	19.05.87	104	" "	25.05.88	120	XI	16
HL 0119	20.05.87	105	" "	21.06.88	118	" "	13
HL 0031	17.05.87	112	X	25.05.88	119	" "	7
HL 0350	20.05.87	119	XI	06.06.88	125	" "	6
HL 0343	19.05.87	141	XIV	26.05.88	143	XIV	2

* Selon Magnin (1962)

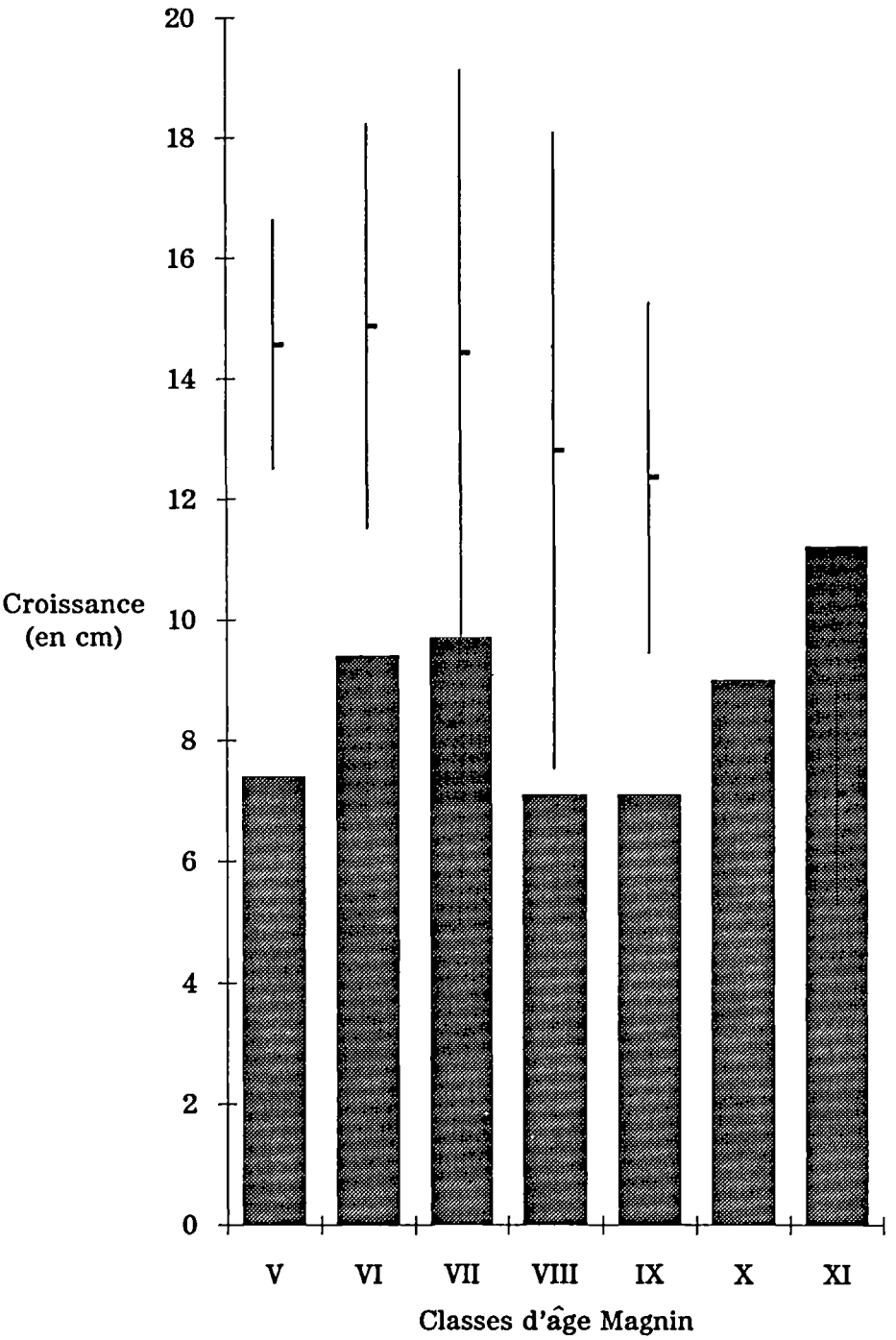


Figure 2 - Croissance moyenne observée et écart type (ŧ) et croissance moyenne déduite des données de Magnin (1962) (▨)

Tableau 4 - Comparaison des croissances moyennes observées et des croissances moyennes déduites des travaux de Magnin (1962), par classe d'âge.

D'après Magnin 1962			D'après marquage de 1983 à 1988	
Classe d'âge	Classe de taille	Croissance moyenne en cm	Nombre d'individus	Croissance moyenne mesurée en cm
I	0 - 35	/	/	/
II	36 - 43	8,3	/	/
III	44 - 54	13,8	/	/
IV	55 - 64	6,9	2	*
V	65 - 71	7,4	6	14,1
VI	72 - 80	9,4	15	14,7
VII	81 - 89	9,7	24	23,2
VIII	90 - 98	7,1	15	12,3
IX	99 - 105	7,1	11	12,4
X	106 - 113	9,0	1	*
XI	114 - 123	11,2	7	7,4
XII	124 - 130	3,6	/	/
XIII	131 - 139	13,6	/	/
XIV	140 - 148	4,6	1	*
			Total : 82	

* Nombre de mesures insuffisant

Nos premiers résultats de lecture d'âge semblent confirmer cette croissance plus importante (Rochard et Jatteau 1989 in Acipenser 1989) puisqu'il apparaît que pour une taille déterminée, l'âge lu sur la coupe de rayon pectoral ne correspond pas à celui prévu par Magnin (1962). Par exemple, un animal de 110 cm aurait 5 ans selon notre lecture d'âge, contre 8 à 10 selon Magnin (1962). Le même décalage est observé par Therrien et al. (1988) sur *Acipenser oxyrhynchus* par rapport à des lectures d'âge effectuées aussi par Magnin (1962).

Lorsque nous aurons validé notre méthode de lecture d'âge grâce à la recapture d'esturgeons marqués et prélevés (opération commencée en 1988) nous pourrons alors établir une clé âge-longueur en accord avec nos mesures de croissance.

A partir de l'effectif 1988, nous avons établi la relation unissant la taille (LT) en cm et le poids en kg :

$$W = 9.48.10^{-6}. (L)^{2,885}$$

Magnin (1962) trouvait $W = 1.949.10^{-6}. (L)^{3.199}$. La différence entre les deux équations n'est pas significative (test U de Mann et Whitney). Le poids que nous obtenons avec notre relation taille-poids est supérieur pour les tailles allant jusqu'à 156 cm et inférieur au-delà. Il faut souligner que dans cet effectif 1988, nous n'avons pas d'animaux de longueur inférieure à 70 cm et très peu de longueurs supérieures à 130 cm. Il sera donc nécessaire d'affiner cette relation dans l'avenir.

Premiers éléments de démographie

Nous avons regroupé par classes de taille, à la figure 3, les effectifs annuels de poissons marqués de 1984 à 1988. L'intervalle de classe retenu est de 7 cm, la classe 1

correspond aux individus dont la longueur totale est inférieure à 23 cm et la classe 24 aux individus dont la longueur totale est supérieure à 176 cm.

On constate une prédominance des classes 9 à 14 (tailles comprises entre 72 et 114 cm) pour l'ensemble des années.

Les effectifs des classes 1 à 6 (moins de 58 cm) sont très faibles ou nuls sauf en 1984 où l'on compte 16 petits individus dans les classes 4, 5, 6.

Le plus petit esturgeon capturé et mesuré en estuaire faisait 22 cm (classe 1) ; il s'agissait d'une capture accidentelle réalisée par un pêcheur commercial dans le haut estuaire, côté Médoc, en janvier 1989.

Le plus grand esturgeon dont nous avons pu contrôler la longueur depuis le début de notre étude (y compris les géniteurs capturés dans les fleuves Garonne et Dordogne) a été capturé en mer au large de l'île d'Ouessant (zone F, Figure 5) à la mi-mars 1985 : il n'était pas marqué et mesurait 222 cm.

Les animaux appartenant à la classe 19 (142 à 148 cm) et au-delà sont représentés dans nos prélèvements depuis 1984 au nombre d'une dizaine chaque année. Ceux-ci ont atteint pratiquement la taille adulte mâle et sont des géniteurs potentiels.

Une distribution polymodale apparaît dans les histogrammes (figure 3) et en s'appuyant sur les résultats de croissance on peut poser l'existence de cohortes composées de 3 classes, la classe centrale en constituant le mode. On peut ainsi suivre la cohorte représentée par les classes 5 à 7 en 1984 ; elle se retrouve dans les classes 7 à 9 en 1985, 9 à 11 en 1986, 11 à 13 en 1987 et 13 à 15 en 1988. On peut aussi suivre deux autres cohortes représentées par les classes 9 à 11 et 14 à 16 en 1984, mais on les perd respectivement après 1987 et 1986. Il apparaît ainsi difficile, voire impossible, d'identifier une cohorte au-delà de la classe 18, soit pour des tailles supérieures à 141 cm. Ces poissons sont cependant présents, comme on l'a montré plus haut, et leur faible nombre peut s'expliquer par un effectif réduit mais aussi par un comportement différent qui les rend moins capturables lorsqu'ils approchent la taille adulte (voir § : cycle biologique).

L'évolution de ces cohortes, notamment la première identifiée qui rassemble l'essentiel des effectifs en 1986, 1987, 1988, semble montrer un vieillissement de la population : les petites tailles sont peu ou pas représentées, vraisemblablement à cause de la faiblesse du recrutement et non à cause d'un problème d'inaccessibilité. Car sinon comment expliquer les résultats de fin 1988 - début 1989 (voir § : répartition et mouvements migratoires) et la quasi absence de prise de petits individus de 1986 à 1988, alors que l'échantillonnage s'est amélioré progressivement depuis 1984 ?

Répartition et mouvements migratoires dans l'estuaire et dans les fleuves

Compte tenu des limites de notre échantillonnage exposées en méthode, l'analyse des résultats de pêche nous donne certaines indications :

- même si l'on ne tient pas compte des années 1981, 1982, 1983 où les pêches ont eu lieu uniquement en mai, juin, juillet, on constate que 75 à 96 % du total des captures de juvéniles sont récoltés à cette période essentiellement en zone 2, à l'aval de Mortagne. Ceci est confirmé par les CPUE (voir tableau 2) : avant mai elles sont faibles et à partir d'août elles chutent;

- sur l'ensemble de l'opération de marquage de 1981 à 1988, les prises les plus précoces ont eu lieu en janvier-février en zone 2 et les plus tardives dans la première quinzaine de novembre en zones 2, 3 et 4 ;

- les pêches expérimentales réalisées en zone 3 à partir de 1986 et en zones 3 et 4 à partir de 1987 n'ont donné lieu à aucune capture. Cependant, lors d'une campagne de pêches scientifiques, le bateau du Centre National de la Recherche Scientifique a capturé au chalut 4 esturgeons le 2 novembre 1988 en zone 3.

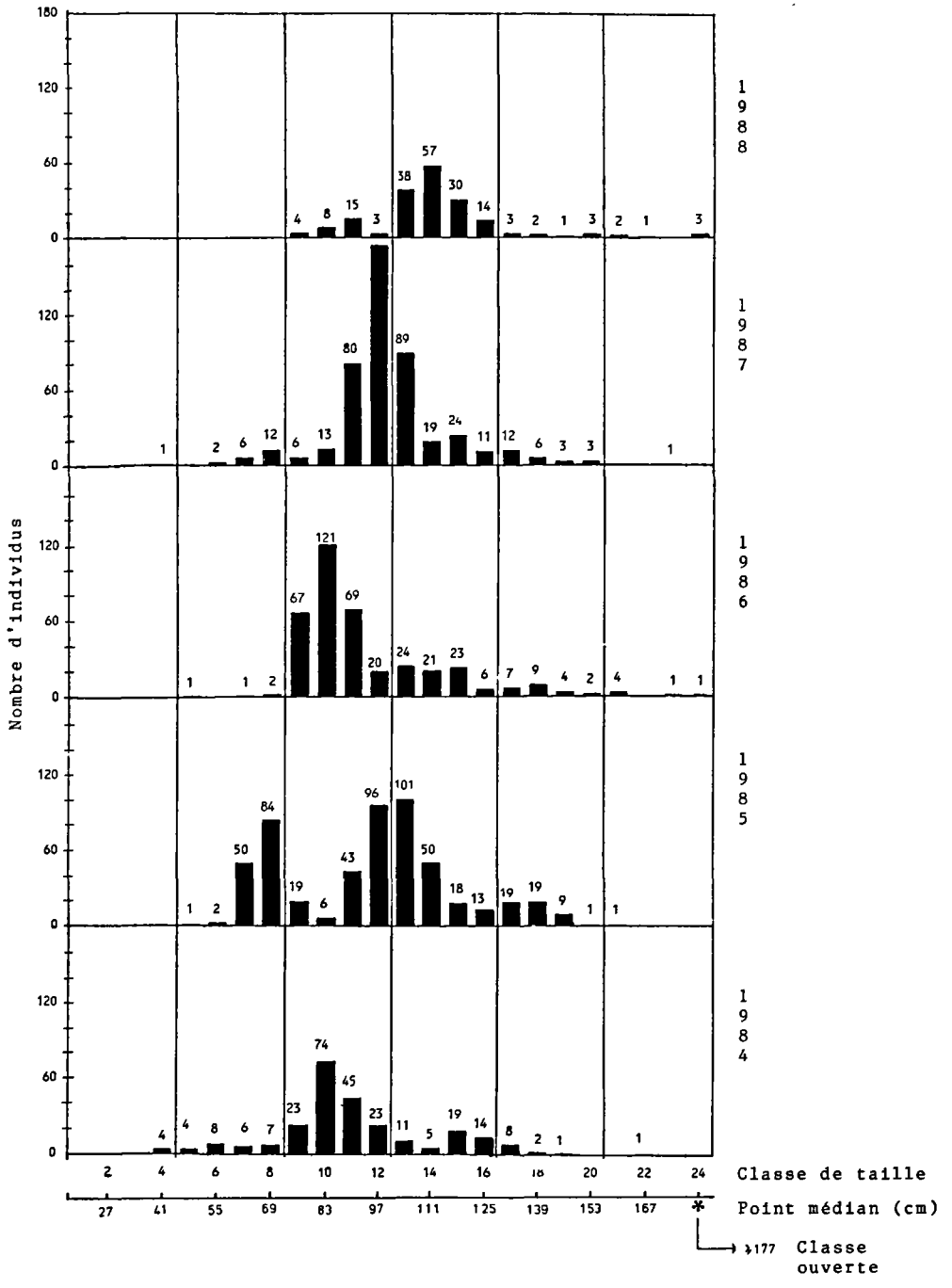


Figure 3 - Distribution des effectifs par classe de taille de 1984 à 1988.

Les pêches commerciales n'ont permis d'enregistrer qu'une prise en zone 3 en juin 1988 et 2 prises en zone 4, l'une en septembre 1986 et l'autre en septembre 1988.

. Sur l'ensemble des juvéniles capturés au moins 2 fois dans l'année en 1986, 1987 et 1988, nous avons reporté à la figure 4 ceux pour lesquels ces captures successives sont distantes d'au moins 15 jours ; 161 poissons sont concernés dont 52 en 1986, 71 en 1987 et 38 en 1988 ; 8 individus ont été capturés 3 fois une même année et 1 individu 4 fois.

On peut émettre l'hypothèse que ces 161 poissons ont séjourné plus de 15 jours dans l'estuaire, la majorité au moins un mois, un tiers deux mois et plus, une dizaine 5 à 6 mois et l'un d'eux plus de 9 mois. Il est à noter que ce dernier, capturé successivement le 10 janvier 1987 et le 21 septembre 1987 (voir figure 4), avait été marqué le 1er octobre 1986 ; n'aurait-il pas quitté l'estuaire pendant toute cette période ?

Le positionnement de la majorité des captures-recaptures en mai-juin-juillet, qui apparaît sur la figure 4 reflète le constat concernant la répartition des captures totales évoquée précédemment.

Il apparaît donc que dès février-mars, des juvéniles pénètrent dans l'estuaire ; on les y trouve en quantité importante en mai-juin-juillet et de septembre à novembre ils regagnent la mer.

. A partir de novembre 1988, nous avons récolté essentiellement grâce aux pêches commerciales des petits individus dont nous avons pu suivre l'évolution.

De novembre 1988 à fin janvier 1989, 16 esturgeons mesurant entre 22 et 31 cm ont été capturés sur la côte médocaine entre Fort Médoc et Pauillac.

En février et mars 1989, 10 esturgeons mesurant entre 26 et 36 cm ont été capturés dans la même zone et au niveau des Callonges.

En avril, mai et juin 1989, 96 esturgeons mesurant entre 33 et 48 cm ont été pris entre les Callonges et Talmont.

Si l'on se réfère aux travaux de Roule (1922) et à ceux de Rochard et Jatteau (in Acipenser 1989), il s'agit d'esturgeons nés lors de la reproduction 1988, qui atteignent l'âge de 1 an environ en juin 1989.

. 35 géniteurs récupérés entre 1981 et 1988 pour les essais de reproduction artificielle ont été capturés dans les lieux et aux saisons suivantes :

- Estuaire 10 ; fleuve Garonne 14 ; fleuve Dordogne 10
- avril 7 ; mai 14 ; juin 11 ; juillet 1.

Le seul géniteur capturé fin juillet (à Talmont) était une femelle qui avait frayé.

Nous n'avons pas pu mettre en évidence sur ce faible échantillon de relation entre ces lieux de capture et ces dates.

Cependant ces données s'inscrivent bien dans le calendrier de Magnin (1962). Notons que les captures en mer de poissons de grande taille (supérieure à 1,50 m) marqués ou non que nous avons recensées ont toutes été effectuées loin de l'estuaire, bien au-delà de la zone de concentration de Letaconnoux (1961), ce qui confirme les suppositions de Magnin (1962) sur leur pénétration en pleine mer.

Répartition et mouvements migratoires en mer

Entre 1982 et 1988, 102 captures d'esturgeons nous ont été signalées en mer ; 66 avaient été marqués en Gironde, dont 62 juvéniles (tableau 1) et 4 géniteurs relâchés après des essais de reproduction artificielle (voir plus loin). Le tableau 5 donne à titre d'exemple le détail des captures recensées en 1988. La figure 5 montre la répartition de l'ensemble de ces captures.

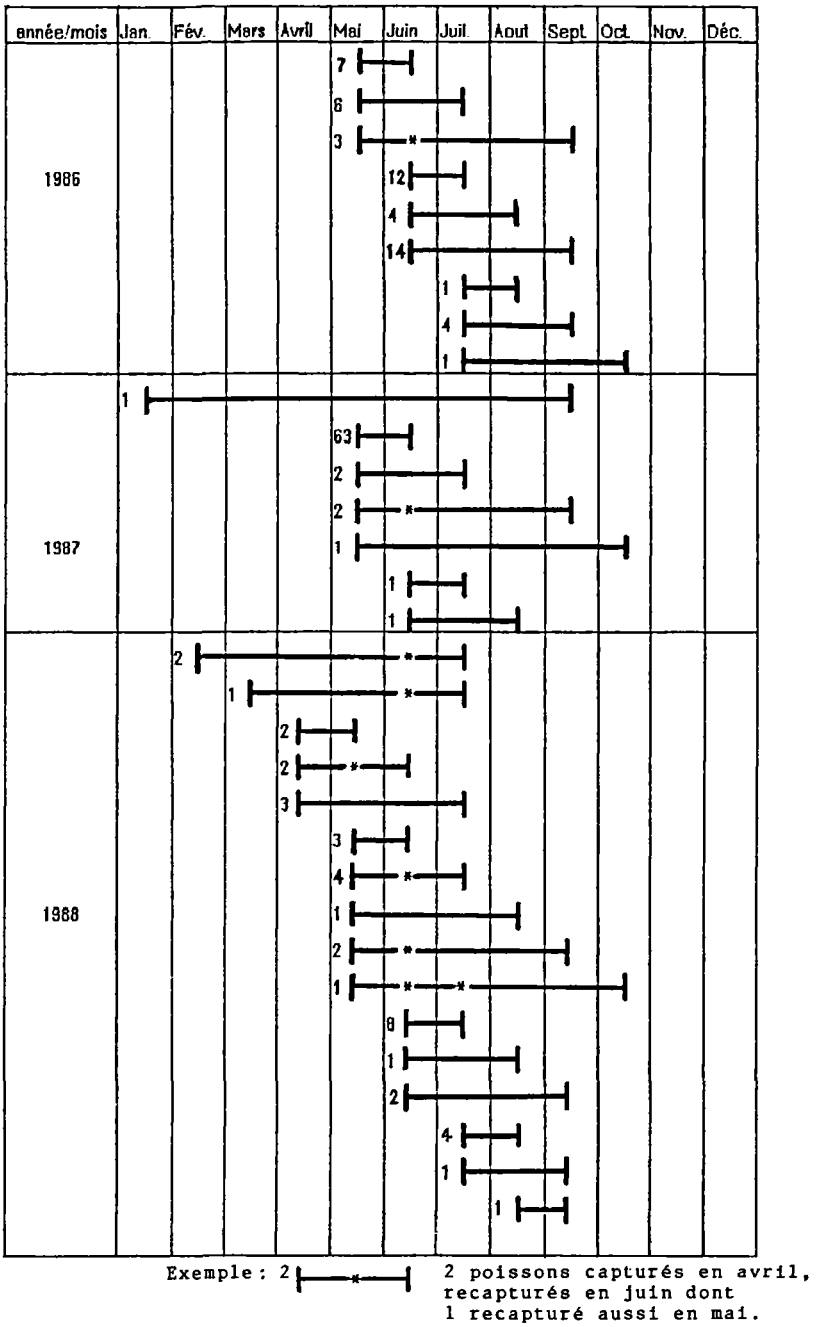


Figure 4 - Captures successives dans l'année espacées d'au moins 15 jours pour 52 poissons en 1986, 71 poissons en 1987 et 38 en 1988.

Nous avons constaté que la proportion des poissons non marqués par rapport au total des captures en mer signalées est passée de 10 % en 1986 à 30 % en 1987 et 50

% en 1988. Ceci peut être une conséquence de la campagne d'information et de sensibilisation effectuée pendant le 1er trimestre 1988. Mais ce retour d'informations est insuffisant : le braconnage persiste dans la zone côtière proche de l'estuaire de la Gironde et l'activité halieutique intense développée dans l'embouchure, passage obligatoire des migrateurs, doit intercepter un nombre d'esturgeons supérieur à celui déclaré.

L'esturgeon capturé au niveau du cercle polaire n'était pas marqué ; par contre les captures réalisées en mer du Nord et près des côtes anglaises concernent surtout des poissons marqués.

On s'aperçoit ainsi que l'aire de répartition en mer de la population qui effectue des migrations dans le bassin Garonne-Dordogne est très importante, beaucoup plus que ne le laissaient supposer les seules données de référence de Letaconnoux (1961). Elle s'étend en fait du golfe de Gascogne à la mer du Nord.

Les 2/3 des captures sont enregistrés loin de l'estuaire au-delà de la zone de concentration des captures des 200 milles circonscrite par Letaconnoux (1961). Ce n'est pas surprenant puisque celui-ci a travaillé uniquement à partir des débarquements des chalutiers de La Rochelle (analyse de 253 captures ramenées en criée entre 1947 et 1960) ;

90 % des captures s'effectuent à moins de 10 km des côtes à des profondeurs inférieures à 40 m ; les autres captures sont réalisées à des profondeurs n'excédant pas 100 m ; cela confirme l'analyse locale de Letaconnoux (1961).

Aucune capture ne nous a été signalée sur les côtes espagnoles. Il se peut que l'information ne nous soit pas transmise ou bien que l'*Acipenser sturio* de Gironde ne dépasse pas le Gouf de Capbreton près de Bayonne (véritable canyon perpendiculaire à la côte française dont la profondeur devient très vite supérieure à 500 m) qui représenterait une barrière difficilement franchissable pour les esturgeons, poissons de fond.

Globalement les 2/3 des captures enregistrées hors estuaire se réalisent au printemps et en été, le reste se répartissant à peu près équitablement sur l'automne et sur l'hiver. Ce constat est contraire à celui de Letaconnoux (1961) qui indique que 71 % des esturgeons débarqués ont été pêchés d'octobre à mars.

Bien que la majorité des esturgeons capturés en mer soit des juvéniles, leur longueur est très rarement inférieure à 90 cm ; notons que les poissons capturés près des côtes anglaises, en mer du Nord et en mer de Norvège sont le plus souvent de grande taille, supérieure à 130 cm.

Les 4 géniteurs utilisés pour des essais de reproduction artificielle ont été recapturés loin de l'estuaire et le dernier mâle signalé s'en est éloigné de plus de 400 km en 13 jours. Ce déplacement assez rapide sur une longue distance se retrouve aussi chez les juvéniles plutôt de grande taille : par exemple l'un d'eux mesurant 121 cm marqué le 19 mai 1987 dans l'estuaire a été repris le 3 juillet 1987 à Saint-Aubin-sur-Mer en zone G (figure 5) après avoir parcouru environ 1000 km en 45 jours.

Les 66 individus marqués, recapturés en mer entre 1982 et 1988, peuvent être regroupés en 4 cas de figure :

1er cas : sur 31 esturgeons présents en mai, juin, juillet en estuaire, 24 ont été recapturés la même année en mer entre fin août et décembre avec un écart de temps de 3 mois et plus (4 cas en 1988 - voit tableau 5) et 7 l'année suivante entre fin août et décembre.

2e cas : 14 esturgeons présents en juin-juillet dans l'estuaire l'année n, ont été recapturés l'année n+1, n+2 ou n+3 entre janvier et avril en mer (1 cas en 1988 - voir tableau 5).

3e cas : 6 esturgeons présents entre avril et juillet en estuaire l'année n ont été repris pratiquement à la même date l'année n+1 en mer pour la plupart très loin de l'estuaire (1 cas en 1988 - voir tableau 5).

4e cas : 11 esturgons ont été recapturés en mer moins d'un mois après leur marquage en estuaire la même année en mai-juin-juillet (4 cas en 1988 - voir tableau 5).

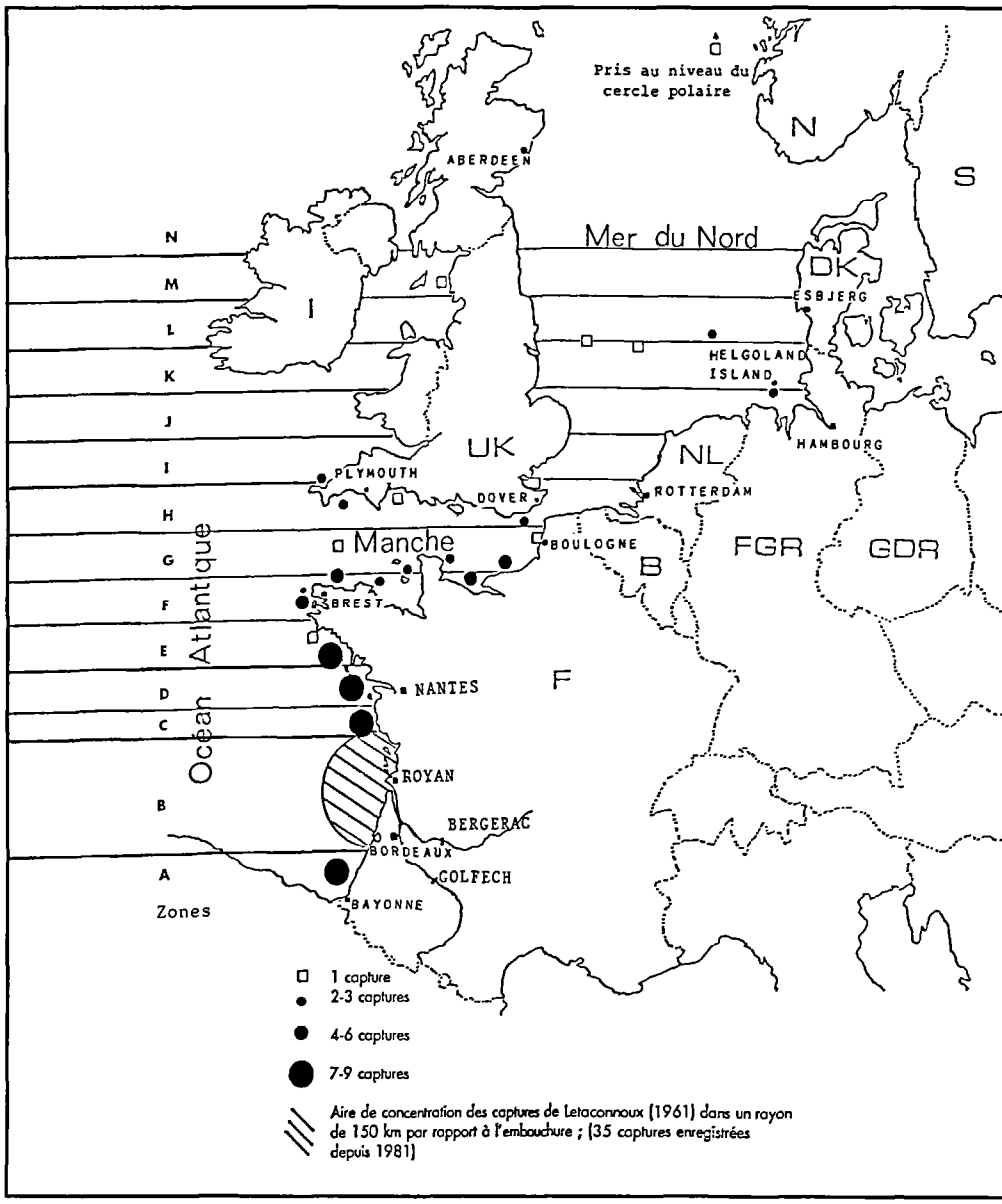


Figure 5 - Répartition des captures d'*Acipenser sturio* en mer avant 1961 d'après Letaconnoux (1961) et depuis 1981 d'après nos travaux.

Tableau 5 - Détail des captures accidentelles en mer recensées pour l'année 1988 et rapportées aux différents cas analysés

Date	Taille rapportée en cm (LT)	L I E U	(ZONE)	Dernière capture estuaire	Taille à la dernière capture	Cas
00 01	?	Saint-Aubin/Mer	(G)	Non marqué	/	/
19.01	105	3 milles W Port bail	(F)	Non marqué	/	/
20 02	115	Sortie baie de Douarnenez	(F)	Non marqué	/	/
00.03	80	6 milles de Pourville	(G)	Non marqué	/	/
05.05	~100	Embouchure Gironde	(B)	18.05.87	92 cm	3ème
08.05	~100	S.O. Cordouan	(B)	Non marqué	/	/
13.05	?	Large Hourtin	(B)	16.06.86	76 cm	2ème
16.05	117	Large Montalivet	(B)	Non marqué	/	/
20.06	108	Embouchure Gironde	(B)	26.05.88	106 cm	4ème
22.06	?	Nord Cornouaille	(I)	16.05.86	162 cm	/
20.07	~120	Large Cap Ferret	(B)	6.07.88	112 cm	4ème
21.07	105	Large Lacanau	(B)	24.06.88	106 cm	4ème
02.08	150	Est Belle-Ile	(D)	Non marqué	/	/
03.08	93	Embouchure Gironde	(B)	Non marqué	/	/
23.08	125	Nord Villers/Mer	(F)	6.06.88	125 cm	1er
08.09	100	S.O. Sables d'Olonnes	(C)	17.08.88	102 cm	4ème
16.10	113	Large Hourtin (35 km)	(B)	22.07.88	111 cm	1er
16.10	~ 50	Large Hourtin	(B)	Non marqué	/	/
?	~ 50	Large Hourtin	(B)	Non marqué	/	/
15.11	~110	2 milles sud Cordouan	(B)	25.07.88	116 cm	1er
09.12	109	1 mille sud Antifer	(F)	21.06.88	105 cm	1er

Les premier et deuxième cas qui regroupent plus des 2/3 des individus, laissent penser que des juvéniles rentrent dans l'estuaire en avril-mai, y séjournent jusqu'à août, retournent en mer fin août-septembre et se retrouvent en mer en automne-hiver.

Le troisième cas indique qu'un même individu peut se trouver entre avril et juillet une année dans l'estuaire et l'année suivante loin en mer à la même époque. Le quatrième cas montre que certains poissons peuvent repartir en mer dès le mois de mai ou bien faire une ou plusieurs (?) brèves incursions dans l'estuaire.

Il est clair que ces déductions ne s'appliquent qu'à l'échantillon analysé et qu'il est difficile d'extrapoler ; il y a bien des échanges entre l'estuaire et l'océan, mais leur importance ne peut être estimée actuellement.

Cycle biologique

Sur la base de nos résultats, nous sommes amenés à penser que les alevins dévalent rapidement les fleuves et atteignent la taille juvénile lorsqu'ils pénètrent dans l'estuaire amont. Ils y passent leur premier hiver et au printemps ils se dirigent progressivement vers l'aval pour atteindre les zones 1 et 2 au début de l'été (figure 1).

Les autres juvéniles de 60 à 145 cm environ qui se trouvent en mer les rejoignent en masse entre mai et juillet, avec quelques précurseurs en février-mars. Ils regagnent vraisemblablement tous la mer à partir de septembre et on observe des retardataires jusqu'en octobre-novembre. Le cycle biologique de l'espèce qui en découle est présenté à la figure 6. Il apporte des précisions par rapport aux données de Magnin (1962) en ce qui concerne la dévalaison des alevins et des juvéniles des fleuves vers l'estuaire, leur séjour en estuaire et leur départ en mer.

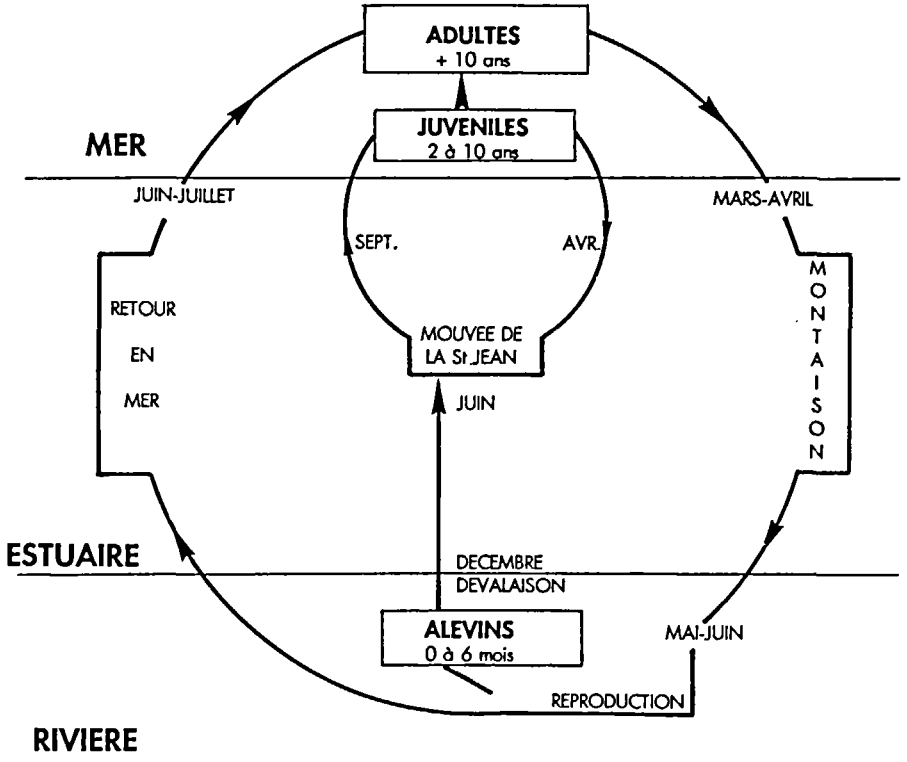


Figure 6 - Cycle biologique d'*Acipenser sturio*.

Le port des Callonges constituerait bien la limite amont de remontée de l'estuaire par le groupe de juvéniles qui effectue la mouvée de la Saint-Jean si l'on se réfère aux quelques captures que nous avons enregistrées en fin de saison en zones 3 et 4. Mais il n'est pas exclu que d'autres zones de regroupement existent, à l'amont de notre secteur d'étude ou sur la rive gauche à l'aval de Pauillac où toute pêche est impossible.

Un certain nombre de facteurs peuvent avoir modifié cette migration ou tout du moins l'avoir influencée : la sédimentation vaseuse côté rive droite du chenal de navigation consécutive à la construction des ouvrages de canalisation des courants de marée jusqu'en 1970, les dépôts près de ces ouvrages côté rive droite de vases issues du dragage du chenal de navigation, l'emprise de la Centrale Nucléaire du Blayais mise en service en 1981 à l'amont immédiat du port des Callonges (figure 1).

Nos données de marquage montrent que des juvéniles sont présents toute l'année en mer et qu'ils effectuent bien des migrations, parfois longues entre mer et estuaire. Les captures plus abondantes au printemps et en été en mer et l'analyse de certains cas de recaptures de poissons marqués, ainsi que les résultats d'estimation du stock par la méthode de Jolly (voir § : évaluation du stock) peuvent laisser penser que les juvéniles effectuent des allées et venues entre mer et estuaire pendant la saison de migration. Cela devrait être vérifié dans l'avenir, éventuellement par radiopistage.

On peut supposer avec Magnin (1962) que les individus qui deviennent adultes lorsqu'ils atteignent la taille de 145 cm environ, ne font plus la migration saisonnière vers le bas estuaire apparemment caractéristique des juvéniles et restent en mer, pour effectuer par la suite uniquement la migration de reproduction. Cela a été montré pour *A. oxyrhynchus*, espèce assez voisine, par Smith (1985). Un phénomène d'évitement des filets peut aussi être évoqué, de même que la colonisation préférentielle de la rive gauche, mais cela reste peu probable.

Evaluation du stock de juvéniles

Pour chaque saison de 1984 à 1988 nous calculons par la méthode de Jolly les effectifs estimés N_i d'*Acipenser sturio* sur le secteur de pêche qui correspondent au stock en i.

Les résultats sont donnés au tableau 6.

Les périodes sont repérées par leur date de début (exemple : en 1984 la période 4 va du 22 juin au 2 juillet). La première et la dernière période pour une année donnée ne permettent pas le calcul des N_i.

Au cours d'une même saison, comme d'une saison à l'autre, les effectifs montrent des variations très importantes.

Elles ne peuvent s'expliquer, au moins en partie, que par un phénomène d'"entrée-sortie" des juvéniles dans la zone de pêche.

Si on analyse les résultats intermédiaires par exemple pour la saison 1985 (tableau 7) on remarque les faits suivants : en période i = 3 il n'y a aucune recapture d'animaux marqués en j = 1,2 ($m_{31} = 0$, $m_{32} = 0$ donc $m_3 = 0$) ceci malgré la capture de r_3 = 153 individus. Du fait que $m_3 = 0$ et que l'on recapture par la suite en i = 4, 5, 6 des animaux marqués en i = 1,2 on aboutit à partir de la formule de Jolly à une valeur N_3 = 8415 très élevée. Lors de la saison 1986 on observe ce phénomène deux fois de suite avec N_3 = 2663 et N_5 = 1765.

Si on ne regroupe pas les jours k de pêche en période on constate que dans la plupart des cas, les $m_{(k+1)k}$ (nombre de poissons capturés en k+1 avec une marque de k) sont nuls ; en effet, il n'y a que très rarement capture du même animal deux jours de suite ; par contre, il est courant qu'il soit repris quelques jours plus tard. Ceci peut être dû à un stress occasionné par le marquage.

L'incapturabilité induite vraisemblablement par le stress sur une période très courte entraîne un biais dans les estimations. On tente de minimiser ce biais en choisissant judicieusement les périodes afin qu'elles ne soit pas trop courtes, mais il

nous est difficile de savoir de quelle façon il perturbe le modèle mathématique d'évaluation de stock utilisé.

Une analyse fine des captures montre que certains individus sont fréquemment capturés ensemble, ce qui laisserait supposer une dispersion non aléatoire. Dumont et al., (1987) observent des regroupements semblables chez *Acipenser fulvescens*. Un tel comportement grégaire pourrait intervenir dans le phénomène d'"échappement" lié au stress et dans le mouvement de va-et-vient évoqué plus haut et au § : répartition et mouvements migratoires.

Tableau 6 - Estimations de l'effectif par la méthode de Jolly.

Années	i	Dates	Mi	Ni	SE_{Ni}
	1	01/01			
	2	18/06	57	1625	2217
1984	3	20/06	16	219	214
	4	22/06	57	427	605
	5	02/07	50	956	1148
	6	01/08			
	1	01/01			
	2	24/05	62	505	329
1985	3	07/06	55	8415	# #
	4	22/06	160	1280	426
	5	07/07	372	1380	748
	6	21/07			
	1	01/01			
	2	02/06	70	844	384
	3	14/06	129	2363	1168
1986	4	27/06	279	958	437
	5	03/07	302	1765	1122
	6	23/07	346	864	512
	7	03/09			
	1	01/01			
	2	07/05	70	3583	3594
	3	18/05	264	1865	667
1987	4	19/05	366	2216	777
	5	20/05	305	1050	357
	6	01/06	1050	2030	2029
	7	02/06	668	1707	1741
	8	17/06			
	1	01/01			
	2	04/06	59	275	106
1988	3	22/06	205	822	462
	4	22/07	101	327	208
	5	01/08			

: Valeurs ne pouvant pas être calculées.
i : Période
Mi : Nombre de poissons susceptibles d'être récupérés en i, vivants, avec une marque.
Ni : Estimation de l'effectif de la population selon la méthode de Jolly.
SE_{Ni} : Erreur standard sur Ni

Tableau 7 - Calcul des mij (nombre de poissons capturés en i avec une marque de j (j<i)) pour la saison 1985.

Périodes i	ri	Périodes j 1	2	3	4	5	mi	Mi	Ni
1	42								
2	56	6					6	62	505
3	153	0	0				0	55	8415
4	143	4	3	10			17	160	1280
5	114	1	1	14	14		30	372	1380
6	109	0	1	4	7	4	16		

ri : nombre de poissons marqués, relachés en i.
mi : nombre de poissons marqués, capturés en i.
Mi : nombre de poissons susceptibles d'être récupérés en i, vivants avec une marque.
Ni : estimation de l'effectif selon la méthode de Jolly.

CONCLUSION

Grâce aux travaux réalisés de 1981 à 1988, nous avons pu vérifier la plus grande partie des données antérieures (Roule, 1922 ; Magnin, 1962 ; Letaconnoux, 1961) et apporter des éléments de connaissance nouvelle, notamment en ce qui concerne la croissance de l'espèce et ses migrations en estuaire et en mer.

Pour ces aspects de la biologie de l'espèce, comme pour celui de l'importance du stock, certaines interrogations subsistent ou se posent dans de nouveaux termes.

Le stock dont nous avons tenté d'estimer la portion qui fréquente l'estuaire de la Gironde a évidemment diminué depuis les précédentes investigations et certaines caractéristiques biologiques ont pu se modifier.

Les croissances observées entre 72 et 105 cm, nettement supérieures à celles trouvées par Magnin (1962) pourraient s'expliquer en partie par ce phénomène. D'ailleurs, Therrien et al. (1988), qui ont aussi rencontré un décalage entre leur relation taille-âge et celle établie par Magnin sur *Acipenser oxyrhynchus* se demandent si "les taux de croissance observés sont dus à une différence de lecture d'âge, à une réponse adaptative de l'espèce face à la diminution des stocks ou à une combinaison des deux".

Depuis 1984, nous capturons des individus de plus de 130 cm de long dans nos échantillons et nous observons un vieillissement de la population de juvéniles qui se retrouve dans l'estuaire de la Gironde. Cela suggère qu'elle génère des adultes et que ce phénomène devrait s'accentuer dans un avenir proche. La survie de ces adultes dépend en grande partie de l'importance des pêches accidentelles (ou dirigées ?) dont ils sont l'objet en particulier sur le plateau continental.

Par contre, le renouvellement des petites tailles est resté faible jusqu'en 1989 où une reproduction de l'année précédente est attestée par une cohorte dont nous avons pu suivre l'évolution au travers de la capture de plus de 120 individus de taille comprise entre 22 et 48 cm.

La fraie a donc heureusement toujours lieu dans le milieu naturel, alors que l'intégrité des quelques frayères rescapées de la destruction par les extractions de

granulats est toujours menacée, même lorsqu'elles sont protégées par voie réglementaire.

Au travers des études que nous avons menées, nous avons participé à la sauvegarde d'*Acipenser sturio*. Sa restauration s'est peut être amorcée, mais elle ne pourra aboutir que si les causes de régression sont définitivement stoppées et si les moyens financiers nécessaires à la réhabilitation du milieu et aux recherches scientifiques sont alloués.

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ASPECTS OF THE STRUCTURE AND DYNAMICS OF AN EXPLOITED CENTRAL CALIFORNIA POPULATION OF WHITE STURGEON (*Acipenser transmontanus*)

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RESUME

Le cycle de vie et la dynamique de population de l'esturgeon blanc (*Acipenser transmontanus*) de la Californie centrale ont fait l'objet d'une étude intermittente depuis 1954, date à laquelle on a rouvert la pêche d'agrément, fermée depuis 1917. Effectuées par marquage et recapture, les évaluations de population portant sur les poissons de taille minimum légale (≥ 101,6 cm de longueur totale) se sont élevées de 11.000 en 1954 à 128.000 en 1984. Le taux annuel d'exploitation s'est accru de 2% à 11,5%. Le taux moyen d'exploitation pour les années 1980 a été plus élevé de 41% que précédemment, au fur et à mesure que la pêche d'agrément est devenue, non seulement plus populaire, mais aussi plus efficace. Les évaluations concernant le taux annuel de survie sont montées de 74% à 90%, 1984 étant l'année du taux de survie le plus bas. D'après les données basées sur la récupération des plaques d'identité entre 1974 et 1988, la majorité des esturgeons blancs resteraient toute l'année dans l'estuaire et les affluents. On trouve des concentrations de poissons dans l'eau saumâtre des baies de Suisun et de San Pablo et ces poissons semblent se déplacer en amont ou en aval en fonction des changements de salinité. Les poissons adultes dans leur phase reproductive, qui représentent une faible portion des effectifs, remontent le fleuve Sacramento et, en plus petit nombre, le San Joaquin, pour frayer vers la fin de l'hiver et au printemps. Il y a un rapport positif entre le recrutement et le débit d'eau douce dans l'estuaire ; on en conclut que le contrôle de ce débit pourrait jouer un rôle important dans l'entretien des effectifs d'esturgeon. La règlementation actuelle de la pêche à la ligne a pour effet de réduire légèrement le rapport exploitation/nouveaux effectifs au-dessous du maximum, mais un autre effet est une réduction de 35% dans la production d'oeufs, en comparaison de la production observée quand l'exploitation était plus faible. De récentes augmentations du taux d'exploitation nous amènent à conclure qu'il faut imposer de nouvelles restrictions à la pêche d'agrément pour réduire la prise et protéger les poissons reproducteurs.

Mots clés : Abondance, pêche, taux de mortalité, mouvement, population, recrutement, marquage, esturgeon blanc, rendement par recrue.

ABSTRACT

Life history and population dynamics of white sturgeon (*Acipenser transmontanus*) in central California have been studied intermittently since the sport fishery was reopened in 1954 after being closed since 1917. Mark-recapture population estimates of legal-sized fish (≥101.6 cm total length) have varied from 11,000 in 1954 to 128,000 in 1984. Annual exploitation rates have ranged from 2% to 11.5%. Average exploitation rate in the 1980's has been 41% higher than in earlier years as the sport fishery has become more popular and effective. Annual survival rate estimates have varied from 74% to 90%, with lowest survival in 1984. Tag returns from 1974 to 1988 suggest that most white sturgeon remain year-round in the estuary and tributary rivers. Fish concentrate in brackish water of Suisun and San Pablo bays and appear to move up or downstream in response to salinity changes. Reproductive adults, a small fraction of the population, move upstream in the Sacramento River and, to a lesser extent, the San Joaquin River in late winter and spring to spawn. Recruitment is positively associated with the volume of freshwater flow through the estuary; hence, management of these flows may be important in maintaining the sturgeon population. The present angling regulations result in a slight reduction of yield-per-recruit below the maximum, but reduce egg production by 35% from that exhibited at previous lower exploitation rates. Recent increases in

exploitation rate suggest that further restrictions on the sport fishery are needed to reduce catch and protect spawning stock.

Keywords : abundance, fishery, mortality rates, movements, population, recruitment, tagging, white sturgeon, year-class strength, yield-per-recruit.

INTRODUCTION

Description of the Sacramento-San Joaquin Estuary

The white sturgeon (*Acipenser transmontanus*) population in central California is mostly confined to the Sacramento and San Joaquin rivers and their common estuary (Figure 1). These two rivers are the largest streams in California's Central Valley and, after draining an area of about 153,000 km^2, form a tidal estuary from their junction in an inland delta to San Francisco Bay. Most of the land surface of the delta is below sea level due to years of subsidence, erosion, and oxidation of organic soils and is protected from the surrounding water by levees originally constructed in the late nineteenth and early twentieth centuries. These agricultural islands exist in a network of over 1,100 km of tidal channels varying in width from 50 m to 1.5 km and mostly less than 15 m deep. Suisun, San Pablo, and San Francisco bays downstream of the delta cover an area of about 1,125 km^2. Important marshes occur around Suisun Bay, northern San Pablo Bay, and southern San Francisco Bay. Suisun and San Pablo bays are characterized by extensive shallow flats, the higher portions of which are exposed at low tide. San Francisco Bay historically had less extensive shallow water areas than the upstream bays; shoreline landfill practices associated with urban and industrial development have further reduced the shallows in San Francisco Bay (Nichols et al., 1986).

Historically, the rivers discharged about 34 km^3 of fresh water into the estuary annually. This has been reduced by over 60% by local use along the rivers and exports to the San Joaquin Valley and Southern California (Nichols et al., 1986). Dams regulating flow in the major tributaries modify seasonal flow patterns by storing winter and spring runoff and releasing water for diversion in summer and fall. Although pulses of freshwater flow from storms continue to occur at about the same frequency as in the past (Herrgesell et al., 1983), the current level of water development has decreased the ratio of winter (Dec, Jan, Feb) to summer flow (Jun, Jul, Aug) by over 40% from the ratio without water development (calculated from data in Miller 1982). About 85% of delta inflow originates in the Sacramento River, 10% comes from the San Joaquin River, and 5% is from smaller eastern valley streams. Water is diverted all along the rivers and in the delta, primarily for agricultural use. The United States Bureau of Reclamation's Central Valley Project and California's State Water Project are the largest diverters in the system as they export water from the southern delta (Figure 1). The combined diversion rate of these projects averaged 200 m^3/s in 1987 and is authorized to increase to 270 m^3/s in 25 years (Chadwick, 1977).

A more detailed description of the Sacramento-San Joaquin Estuary is provided by Kelley (1966).

White Sturgeon in the Sacramento-San Joaquin Estuary

White sturgeon is a native anadromous fish in the Sacramento-San Joaquin Estuary and the object of an important and growing sport fishery ; commercial fishing for sturgeon is prohibited in California. Another species, the green sturgeon (*A. medirostris*), is much less common in the estuary and legal-sized ($\geq$101.6 cm total length) fish are seldom caught.

Historical accounts indicate that a commercial fishery greatly reduced the Sacramento-San Joaquin Estuary white sturgeon population in the late 1800's (Skinner, 1962). As a result, all sturgeon fishing was prohibited in 1917 ; the fishery was reopened to sport angling only in 1954. With the exception of the period from 1956 to 1963 when the minimum size limit was raised to 127 cm total length (TL), the sport fishery has had

the same regulations since its inception : a year-round season, 101.6 cm TL minimum size limit, and a one fish per day creel limit.

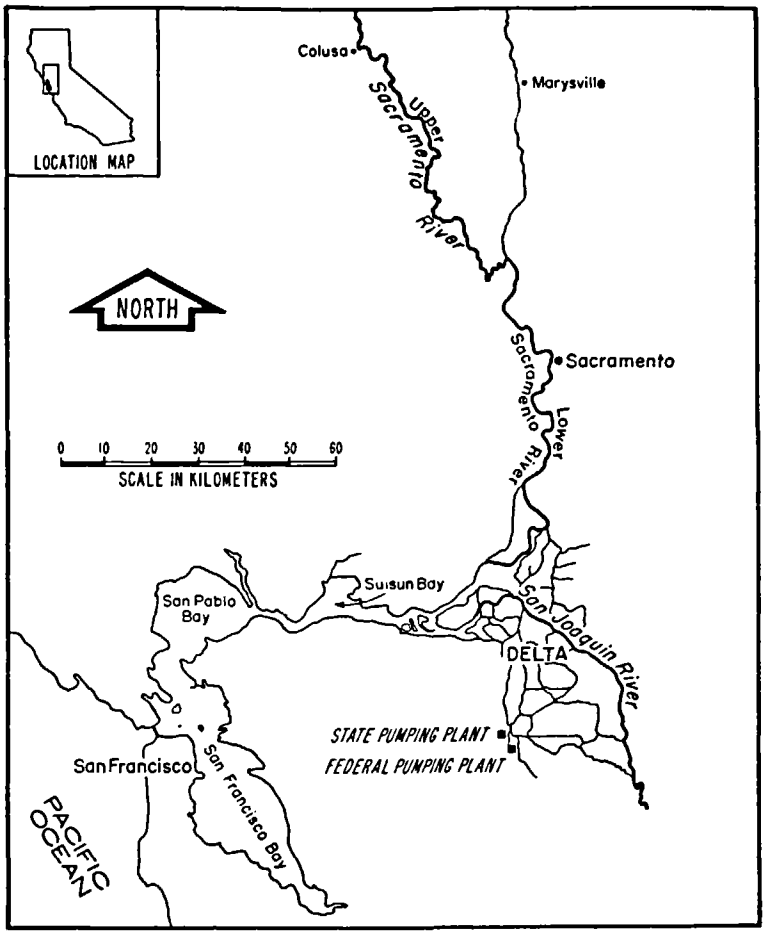


Figure 1 - The primary area inhabited by white sturgeon in California

Initially, the sport fishery was unsuccessful because angler interest was low and effective angling techniques had not been developed. However, in 1964 fishing success improved dramatically when shrimps, *Crangon spp.* and *Palaemon macrodactylus* were first used as bait. Since then, burrowing shrimps of the genera Callianassa and *Upogebia* also have been widely employed as sturgeon bait.

White sturgeon life history and population dynamics have been studied intermittently since the sport fishery reopened in 1954. The general time and location of spawning in the Sacramento River have been described (Stevens and Miller, 1970 ; Kohlhorst, 1976), as have food habits of juveniles (Schreiber, 1962; Radtke, 1966) and adults (McKechnie and Fenner, 1971) and growth rate (Kohlhorst et al., 1980; Brennan and Cailliet, in press). Tagging studies (Pycha, 1956; Chadwick, 1959; Miller, 1972 a, b; Kohlhorst, 1979, 1980; Brennan and Cailliet, 1991) have been used to estimate abundance and mortality rates, to describe movements, and to validate aging techniques.

This report summarizes and updates these abundance and mortality rate estimates through 1988, describes general movement patterns since 1974, examines the effect of hydrologic variables on year class strength, and estimates the effect of sport angling regulations on yield.

METHODS

Tagging

A tagging study was used to obtain mark-recapture estimates of white sturgeon abundance, exploitation rate, survival rate, and movement pattern. In 1954 and 1974, pectoral fin rays were removed from some tagged fish for the purpose of age determination. Tagging methods and results before 1984 were originally reported by Pycha (1956), Chadwick (1959), Miller (1972a, 1972b), and Kohlhorst (1979, 1980), Kohlhorst et al. (1980), and Shirley (1987). Some earlier results are presented here for comparison with recent estimates. The following tagging methods are those used in 1984, 1985, and 1987.

Sturgeon to be tagged were captured in a 20.3 cm stretched mesh trammel net, 366 m long and 6 m deep, that was drifted in the fall in San Pablo Bay (and occasionally in Suisun Bay) (Figure 1). Drifts lasted from 1/4 to 2-1/2 h. Sturgeon were removed from the net, placed in fabric or plastic cradles, and tagged with disc-dangler tags attached with stainless steel wire through the muscle below the anterior end of the dorsal fin using methods described by Chadwick (1963). Twenty-dollar reward tags were used exclusively to assure a high rate of response from anglers catching tagged sturgeon. Only legal-sized fish were tagged. After being tagged and measured to the nearest centimeter total length, fish were immediately released near the site where they were captured.

As part of a study to validate sturgeon aging techniques, some fish each year were injected with oxytetracycline (OTC) at the time of tagging (26% of the tagged fish in 1984, 31% in 1985, and 79% in 1987) (Brennan and Cailliet, 1991). Injected fish were double-tagged with T-bar disc or spaghetti tags in 1984 and 1985, but most of these second tags were shed in the first year after release and were not used in any analysis presented here. All fish were single-tagged in 1987.

Mean total length of tagged fish was calculated for each tagging year from 1974 to 1987. The statistical significance of differences in mean size was tested using one-way ANOVA and Tukey's multiple comparison test (Zar, 1984).

Abundance

In 1984 and 1985, white sturgeon abundance was estimated using Bailey's (1951) modification of the Petersen method because a recapture sample was available from a later year. In 1987, the multiple census method of Schumacher and Eschmeyer (Ricker, 1975) was used because recaptures during tagging were all that were available. Confidence intervals for the Petersen estimates were calculated by assuming tag recaptures followed a Poisson distribution and, for the Schumacher and Eschmeyer estimate, by following Ricker (1975). Abundance estimates before 1984 used one of the above methods, depending on the availability of a recapture sample in subsequent years (Kohlhorst 1980). The assumptions inherent in any mark-recapture experiment are described by Ricker (1975) and Kohlhorst (1980) discusses potential biases in the multiple census technique as applied to white sturgeon. Assumptions of random distribution of tagged fish in the untagged population and equal vulnerability of tagged and untagged fish to the fishing gear are likely violated by the multiple census techniques. Both methods deal with a population that is probably not closed and the proportion of the entire population represented by the estimate is unknown and may vary between estimates.

Abundance was also indexed using catch per net-h during tagging as described by Kohlhorst (1980) for 1967-1979. Here, we add values for 1984-1987.

Exploitation Rate

Exploitation rate was estimated as R_1/M, where R_1 = the number of tags returned by anglers in the first year after tagging and M = the number of fish tagged. We assumed that the use of reward tags assured a 100% reporting rate for all tagged fish recovered by anglers. Exploitation rate estimates would be biased low by failure of this assumption or by unrecognized tagging mortality. Confidence intervals for exploitation rate estimates were calculated assuming a Poisson distribution for tag recoveries (Ricker, 1975). When return rate for OTC-injected fish and uninjected fish differed, the higher return rate was used as the estimate of exploitation rate.

Survival Rate

We estimated annual survival rate for the one-year period following tagging in 1984 with a bias-adjusted maximum likelihood equation (Model 1 of Brownie et al., 1978) applied to returns from 1984 and 1985 tagging.

When a maximum likelihood estimate of survival rate could not be obtained because tagging was not conducted in two successive years, we determined average annual survival rate from a catch curve (Ricker, 1975) using age frequencies of tagged sturgeon. Ages were estimated from length frequencies with an age-length key based on sturgeon aged in 1973-1976. The slope of the straightest portion of the descending right limb of the curve was calculated using regression of the natural logarithm of numbers on age. The antilog_e of the slope is an estimate of survival rate.

Movements

We described movement patterns by recording the month and location of tag recovery reported by anglers from 1974 to 1988. Tag return data were combined to provide a general picture of movement and examined separately by recovery year to detect variations or trends in movement patterns.

Young White Sturgeon Abundance and Year Class Strength

Young-of-the-year and juvenile (ages 1-5) white sturgeon were captured with otter and midwater trawls fished once monthly at 35 locations from southern San Francisco Bay to the western delta (Figure 1) from 1980 to 1987. The semiballon otter trawl (4.9 m mouth opening, 2.5 cm stretched-mesh body, 1.3 cm stretched-mesh cod end) was towed on the bottom for 5 min at each location. The midwater trawl (3.7 m by 3.7 m mouth opening with variable mesh from 20.3 cm stretched-mesh at the mouth to 1.3 cm stretched-mesh at the cod end) was fished in a diagonal tow from bottom (or 12.2 m, whichever was shallower) to surface for 10 min at each location. Captured white sturgeon were measured to the nearest millimeter fork length.

Approximate age was assigned using the Von Bertalanffy growth curve for white sturgeon (Kohlhorst et al., 1980) after first converting fork length to total length by multiplying by 1.08. Abundance and distribution were examined for annual variability and trends.

An index of year class strength for year i was calculated as :

$YCI_i = \Sigma N_o/y$, where
y = number of years in which fish of year class i were available to the sampling gear at ages 0-5 and
$N_o = N_t/e^{-Zt}$, where
t = age at capture,
N_t = number captured at age t,
Z = assumed instantaneous mortality rate of 0.2877 (equivalent to annual survival rate of 0.75), and
N_o = backcalculated catch at age 0 that would be the equivalent of N_t.

With this method, the more ages at which each year class is sampled, the more reliable the estimate of YCI.

Associations between YCI and hydrologic variables (mean daily freshwater outflow from the estuary, mean daily freshwater diversions from the delta, and mean daily percent of inflow diverted) were explored with correlation analysis. These environmental variables were chosen because they affect the production of young by other anadromous fishes in the estuary (Turner and Chadwick, 1972; Stevens, 1977; Stevens and Miller, 1983; Stevens et al., 1985).

Effect of Angling Regulations

To estimate the relative effects of different regulations on yield we computed yield-per-recruit in terms of biomass for various lower size limits and fishing mortality rates. In addition, to evaluate the potential effects of regulations on future recruitment, we computed eggs-per-recruit for various size limits and fishing mortality rates. Because of a paucity of fecundity and spawning frequency data for this population, we have assumed egg production is proportional to weight for these calculations.

RESULTS

Tagging

We tagged a total of 5,952 white sturgeon in 1984, 1985, and 1987 (Table 1). Mean total length of tagged fish since 1974 has ranged from 121 to 135 cm. Fish tagged in 1979 and 1984 were significantly smaller than in other years since 1974 and fish tagged in 1974 were significantly larger ($p<0.001$).

Table 1 - Number, size, and tag reward value of white sturgeon tagged in the Sacramento-San Joaquin Estuary from 1954 to 1987. Only legal-sized (≥ 101.6 cm total length) fish are included. Values for years before 1974 are from Pycha (1956) and Miller (1972a). Kohlhorst (1979, 1980) reported some of the values for 1974 and 1979

Year	Number Tagged	Total Length (cm)			Reward Tag Value($)
		Mean	Range	Conf. Int.[1](95 %)	
1954	994	145	102-239	-	0
1967	1,612	119	102-163	-	5
1968	1,080	124	102-163	-	5
1974	713	134.7	102-239	133.2-136.2	5
1979	1,368	121.4	102-202	120.5-122.2	10
1984	2,551	122.3	102-272	121.8-122.9	20
1985	2,419	126.2	102-214	125.6-126.8	20
1987	982	126.0	102-225	124.9-127.1	20

[1] Conf. Int. = Confidence Interval

Abundance

White sturgeon abundance estimates have varied substantially since 1954 (Table 2). The lowest estimated population of 11,000 legal-sized fish in 1954 (Pycha, 1956) increased about 10 times to 115,000 (Miller, 1972a) by 1967 and then decreased

again to 21,000 in 1974 (Kohlhorst, 1980). Kohlhorst's results indicated another increase to 74,000 fish in 1979. Recent tagging results show abundance reaching the highest level yet estimated (128,000) in 1984 followed by a decrease to 96,000 in 1985 and 84,000 in 1987.

As reported by Kohlhorst (1980), catch per net-h followed a pattern generally similar to the abundance estimates, although the correlation between these two measures of abundance is not statistically significant ($r=0.62$, $n=7$, $p>0.10$). Notable differences in the pattern of change shown by these two measures of abundance occurred in 1968 and 1987 (Table 2). In 1968 the multiple census estimate decreased from the previous year while catch per net-h increased and in 1987 catch per net-h decreased relatively more than the multiple census estimate.

Table 2 - White sturgeon tagging results for the Sacramento-San Joaquin Estuary from 1954-1987. All values pertain to legal-size (≥ 101.6 cm total length) fish only. Results for years before 1984 were originally reported by Pycha (1956), Chadwick (1959), Miller (1972a), and Kohlhorst (1979, 1980).

Year	Catch per Net-h	Abundance		Exploitation Rate		Survival Rate	
		Estimate	95% Conf. Int.[3]	Estimate	95 % Conf. Int.[3]	Estimate	95 % Conf. Int.[3]
		x 10^3	x 10^3	x 10^{-3}	x 10^{-3}		
1954	-	11.2	8.8-15.7	20	13-31	-	-
1967	15.3	114.7	72.4-212.3	73	58-88	0.84[1]	0.65-1.03
1968	19.5	40.0	29.2-63.5	65	49-83	-	-
1974	3.7	20.7	13.1-50.1	56	36-84	0.90[2]	0.82-0.99
1979	8.4	74.5	53.3-123.3	83	68-98	0.82[2]	0.78-0.87
1984	36.2	128.3	100.6-170.0	89	68-114	0.74[1]	0.64-0.84
1985	18.2	96.2	64.2-157.9	115	90-141	0.84[2]	0.80-0.89
1987	6.7	84.0	46.8-411.4	87	67-110	0.86[2]	0.82-0.90

[1] Maximum likelihood estimates from tag returns
[2] Catch curve estimates from age frequencies of tagged fish
[3] Conf. Int. = confidence interval

Exploitation Rate

Based on estimates from 1984, 1985, and 1987 tagging, annual exploitation rate has ranged from 0.087 to 0.115 recently (Table 2). This is higher than previous estimates, and the trend in exploitation rate (with 1954 omitted since present fishing techniques were not in use then) is almost statistically significant ($F=6.46$; $df=1,5$; $p=0.052$).

Survival Rate

Survival rate estimates have been remarkably uniform over the period of record (Table 2). The maximum likelihood estimate of 0.74 in 1984 is the lowest that we have observed. The catch curve estimates of survival (1974, 1979, 1985, 1987) exhibit little variability, possibly because they are essentially averages over several years (Figure 2, Table 2). The greater precision of the catch curve estimates, reflected in the narrow confidence intervals, does not imply greater accuracy of these estimates in relation to the maximum likelihood estimates.

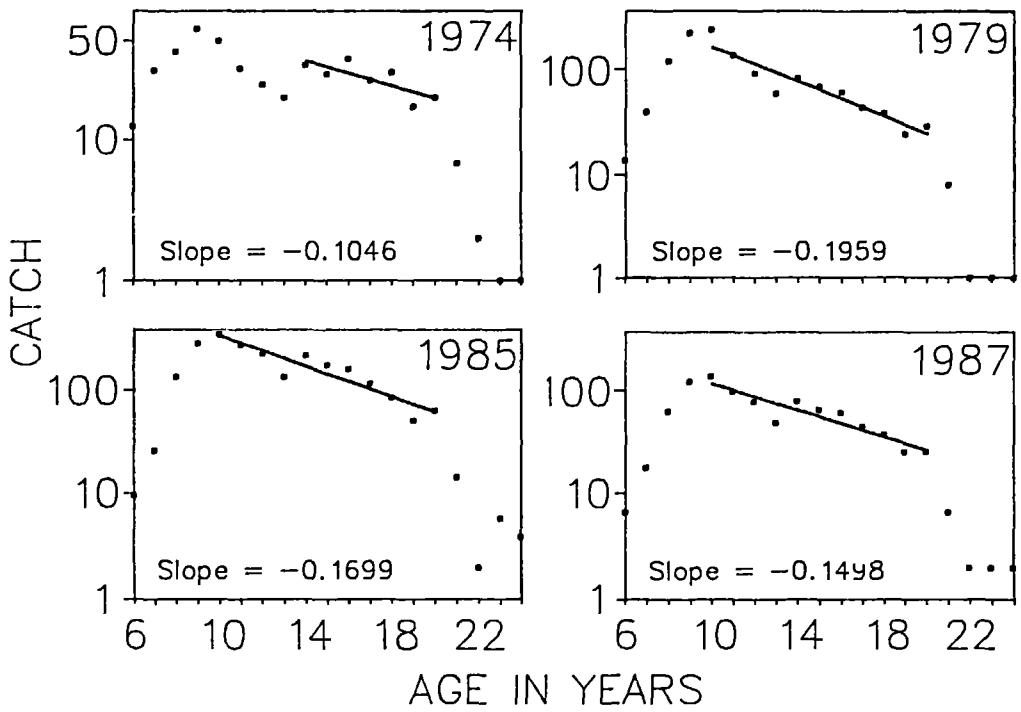


Figure 2 - Catch curves for white sturgeon tagged in the Sacramento-San Joaquin Estuary in 1974, 1979, 1985 and 1987. The slope of the straightest segment of the descending right limb of the curves was used to estimate annual survival rate. The slope of each line is shown. Survival rate = antilog$_e$ (slope). Note that the y-axis scale for 1974 is different from the other years.

Movements

A total of 1,455 white sturgeon tags returned by anglers between 1974 and 1988 was used to describe movement patterns. Over 66% of the tag returns were received from the Suisun and San Pablo Bay area (Table 3). Many sturgeon are found in these two bays throughout the year, but peak fishing in Suisun Bay occurs from November through January ; it occurs from January through March in San Pablo Bay. In San Francisco Bay, over half the annual catch is taken from January through March and almost no fish are caught from August through October.

Some sturgeon move into the lower Sacramento River and the delta in fall and their numbers increase in winter (Table 3). A portion of these fish, presumably those that are mature and ready to spawn, move up the Sacramento River until they are concentrated in the upper river near Colusa (Figure 1) from March through May. The mean total length at tagging of white sturgeon caught in the upper Sacramento River was 145 cm; this is significantly greater ($p<0.05$) than the mean length of 125 cm for the entire tagged sample. As most female white sturgeon in the Sacramento-San Joaquin Estuary do not mature until they reach about 135 cm (Calif. Dept. Fish and Game, unpublished), the large size of fish in the Sacramento River and the known spawning period of late February through May (Kohlhorst, 1976) indicate that the movement up the Sacramento River is a spawning migration.

Movement of white sturgeon into the San Joaquin River in the spring (Table 3) suggests spawning occurs there also, although no sampling to collect sturgeon eggs or larvae has been done in the San Joaquin system. If the number of tag returns from each river is a valid indicator of the relative number of spawning fish, over ten times (spring tag return ratio of 53:5; Table 3) as many white sturgeon spawn in the Sacramento River as in the San Joaquin River. Flows in the San Joaquin River are only about 1/8 those in the Sacramento River.

In recent years, some white sturgeon have moved out of the estuary and migrated up the coast to Oregon and Washington. Chadwick (1959) reported one white sturgeon tagged in 1954 was returned from the Columbia River, Oregon, but no additional evidence of coastwise migration was seen until 1985 when a white sturgeon tagged in 1979 was captured in the Chehalis River, Washington. Since then, 11 more tagged white sturgeon have been caught in six river systems north of California, primarily the Umpqua River, Oregon (five tags recovered) and the Chehalis River, Washington (three tags). Also, one tag return each was received from the Columbia River, Yaquina River, and Tillamook Bay, Oregon and the Willapa River, Washington.

Table 3 - Returns by area and month for white sturgeon tagged in the Sacramento-San Joaquin Estuary and recovered by anglers from 1974 to 1988.

	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC	TOTAL
Washington	-	-	-	-	-	-	2	2	-	-	-	-	4
Oregon	-	2	1	2	-	1	2	-	-	-	-	-	8
Pacific Ocean	-	-	1	-	-	-	-	-	-	-	-	-	1
San Francisco Bay	51	45	81	44	18	5	2	1	1	1	13	43	305
San Pablo Bay	61	70	111	39	37	21	13	8	10	18	23	55	465
Suisun Bay	65	38	39	32	32	42	31	28	27	42	58	71	505
Lower Sacramento River	13	16	11	6	3	2	-	-	1	3	5	7	67
Delta	4	-	3	3	1	1	-	-	3	2	6	5	28
Sacramento River	1	-	5	4	2	-	-	-	-	-	-	2	14
Upper Sacramento River	3	2	12	23	8	3	-	-	-	-	-	2	53
San Joaquin River	-	-	3	1	1	-	-	-	-	-	-	-	5
TOTAL	198	172	267	154	102	75	50	39	42	66	105	185	1455
PERCENT OF TOTAL	13.6	11.8	18.4	10.6	7.0	5.2	3.4	2.7	2.9	4.5	7.2	12.7	

White sturgeon appear to move within the estuary in response to flow, which affects salinity, so that they are farther upstream when saline water encroaches eastward in dry years and farther downstream when brackish water is pushed westward in wet years. In dry years, when freshwater flow was low and salt water encroached upstream, such as 1976, 1977, and 1985, more sturgeon were found in Suisun Bay and fewer in San Pablo Bay than in other years (Figure 3). In wet years with high freshwater flows such as 1978, 1980, 1982, and 1986, more fish were recaptured in San Pablo Bay. The percentages of annual tag returns from these two bays are significantly correlated (Suisun Bay : $r=-0.64$, $n=13$, $p<0.02$; San Pablo Bay :

r=0.80, n=13, p<0.01) with year type when year type is quantified by ranking from critically dry (1) to wet (4). More fish also tend to use San Francisco Bay in wet years, but the association between the percent of annual returns from there and ranked year type was not significant (r=0.39, n=13, p>0.10).

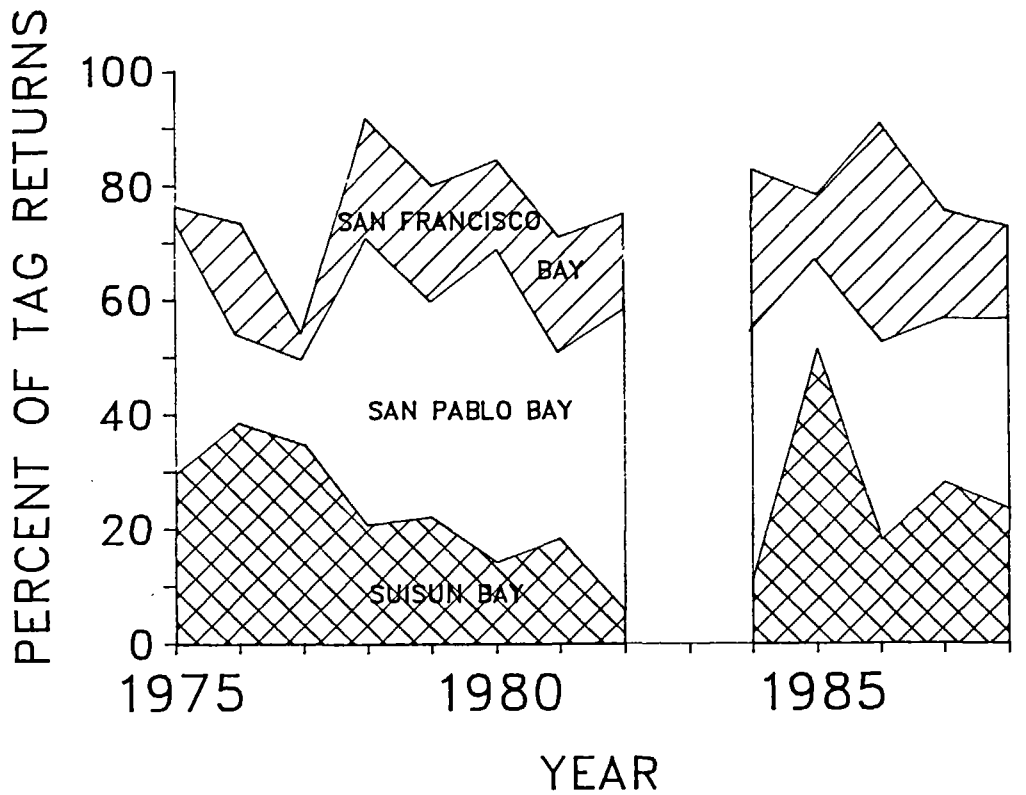


Figure 3 - Annual percent of white sturgeon tag returns by sport anglers from Suisun, San Pablo, and San Francisco bays in the Sacramento-San Joaquin Estuary. We omitted 1983 because only 17 tags were returned that year.

Young White Sturgeon Abundance and Year Class Strength

A total of 429 white sturgeon was captured in otter and midwater trawls from 1980 to 1987. These fish ranged from 31 to 1,382 mm total length (Figure 4), but fish greater than age 5 (approximately 815 mm) were omitted when indexing year class strength for 1975 to 1986. Estimated production from the 1982 and 1983 year classes was substantially greater than for other years in this period (Figures 4 and 5). These were both years of very high spring and early summer freshwater outflow from the estuary and are responsible for the significant (p<0.001) correlation between year class index and outflow in all months from April to July (Table 4, Figure 5). These correlations with mean daily flow decreased slightly from April to July, suggesting spring flow affects production more than summer flow. Correlations of the year class index with the mean daily volume of diversions by the State and Federal water projects and delta agriculture were not significant. Negative correlations with the percent of inflow that is diverted increased from March to July, but only the July value was significant (p<0.001).

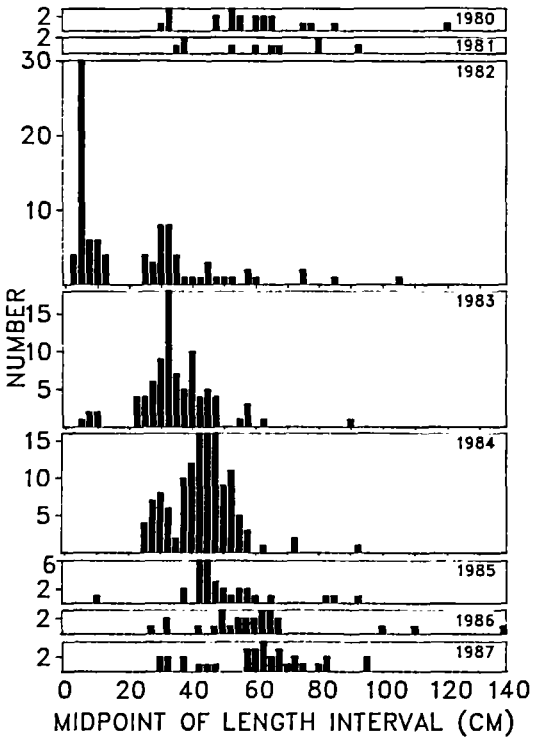


Figure 4 - Annual length frequencies of white sturgeon caught in otter and midwater trawls in the Sacramento-San Joaquin Estuary from 1980 to 1987.

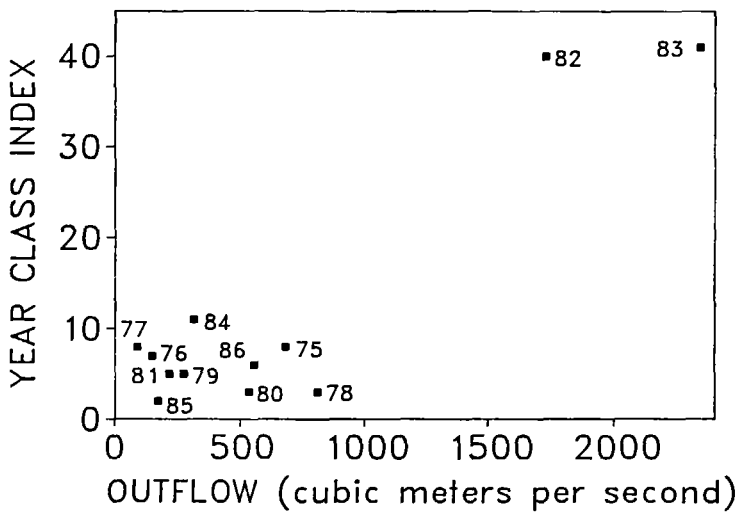


Figure 5 - Scatterplot of white sturgeon year class index versus mean daily outflow for April to July in the Sacramento-San Joaquin Estuary. Numbers adjacent to points designate year classes.

Table 4 - Product-moment correlation coefficients between the white sturgeon year class index and hydrologic variables in the Sacramento-San Joaquin Estuary for the years 1975-1986. *Asterisks* denote statistical significance at the 0.001 level. Experimentwise error rate over all 21 correlations is approximately 0.02. Mean daily values are used for all hydrologic variables.

Variable	Correlation with Year Class Index
March Outflow	0.579
April Outflow	0.882*
May Outflow	0.842*
June Outflow	0.831*
July Outflow	0.828*
April-May Outflow	0.890*
May-June Outflow	0.849*
June-July Outflow	0.833*
April-May-June Outflow	0.898*
May-June-July Outflow	0.852*
April to July Outflow	0.903*
March Diversions	0.379
April Diversions	0.209
May Diversions	-0.097
June Diversions	-0.234
July Diversions	-0.376
March % of Inflow Diverted	-0.231
April % of Inflow Diverted	-0.423
May % of Inflow Diverted	-0.445
June % of Inflow Diverted	-0.678

Effect of Angling Regulations

Yield-per-recruit calculations indicate that observed values of instantaneous fishing mortality rate and minimum size limit are near a relatively flat part of the yield surface ; hence, biomass yield is relatively insensitive to small changes in these parameters (Figure 6). For earlier fishing mortality rates (0.06), the size limit that would produce maximum yield was near 120 cm, but the existing limit (101.6 cm) reduces yield only 3%. For more recent fishing mortality rates (0.09), a size limit of 130 cm maximizes yield, but the existing size limit reduces yield only 8%.

In addition to maintaining high yields per recruit, we also wish to maintain high recruitment rates. Because the relationship between stock and recruitment is not known for white sturgeon, we have resorted to the goal of providing for adequate egg production (Botsford and Hobbs 1986). On the basis of egg-per-recruit calculations, the recent increase in instantaneous fishing mortality rate (from 0.06 to 0.09) has reduced egg production by 35%.

Returning to the previous level of egg production would require either :
(1) increasing the size limit to 130 cm,
(2) returning fishing mortality rate to its former value, or
(3) a combination of changes such as increasing the size limit to 120 cm and reducing instantaneous fishing mortality rate to 0.075.

The first would increase biomass yield by 8%, the second would reduce yield by 7%, and the third would increase yield by 3%.

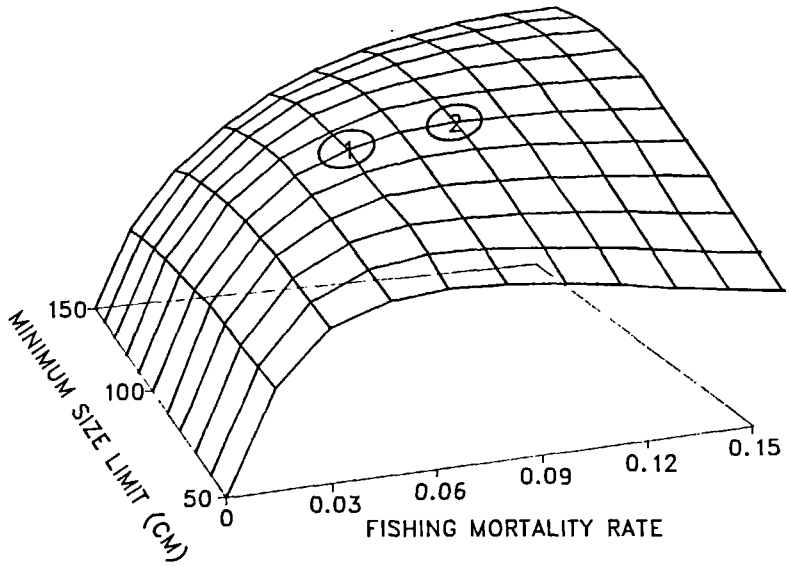


Figure 6 - Biomass yield for the Sacramento-San Joaquin Estuary population of white sturgeon. Ellipse 1 reflects fishing mortality rates in the 1960s and 1970s and ellipse 2 reflects current higher fishing mortality rates.

DISCUSSION

White sturgeon abundance in the Sacramento-San Joaquin Estuary has varied dramatically in the last 35 years, while total mortality rates during much of that period have been relatively stable. This suggests that the abundance of legal-sized sturgeon has been controlled primarily by variations in recruitment. If, like many other fishes, white sturgeon year class strength is set early in life, then recruitment is directly related to previous production of young fish.

We have presented evidence that the production of young sturgeon may be associated with freshwater outflow from the estuary ; in years with very high outflow in spring and early summer, more young-of-the-year sturgeon are produced. Year class strength may also be weakly associated with the fraction of freshwater inflow to the estuary that is diverted in the delta, but this is likely the result of an inverse relationship between percent diverted and flow when diversions are relatively constant, as they were for the 1975-1986 period when year class indices are available. The association of sturgeon year class strength with outflow is consistent with relationships observed for several other anadromous species inhabiting the estuary, specifically striped bass (*Morone saxatilis*) (Turner and Chadwick, 1972; Stevens et al., 1985), chinook salmon (*Onchorynchus tshawytscha*), American shad (*Alosa sapidissima*), and longfin smelt (*Spirinchus thaleichthys*) (Stevens and Miller, 1983). High flows may improve young sturgeon survival by transporting larvae to areas of greater food availability, by dispersing larvae over a wide area of the rivers and estuary to take advantage of all available habitat, by quickly moving larvae downstream of any influence of water diversions in the delta, or by enhancing productivity in the nursery area by increasing the nutrient supply. Additionally, adults may experience a stronger attraction to upstream spawning areas in high flow years and spawn in greater numbers.

While we have demonstrated an association between year class strength and outflow, data are inadequate to evaluate the nature of the spawner-recruit relationship. In this regard, a time series of recruitment estimated from aged samples collected from

the population at four times between 1954 and 1986 depicts a large peak in recruitment in 1938, a broader peak in the mid-1950s and an even broader peak in the late 1960s (Figure 7) (Shirley 1987). This pattern is consistent with an "echo effect" (Leslie 1945 ; Bernardelli 1941) for a species that matures at about age 14. It reflects decaying peaks in reproduction that result from a large recruitment with a positive influence of adult numbers on recruitment. Given this suggestion of a spawner-recruit relationship, it seems prudent, for management purposes, to make the conservative assumption that spawning stock, as well as environmental factors, influence recruitment.

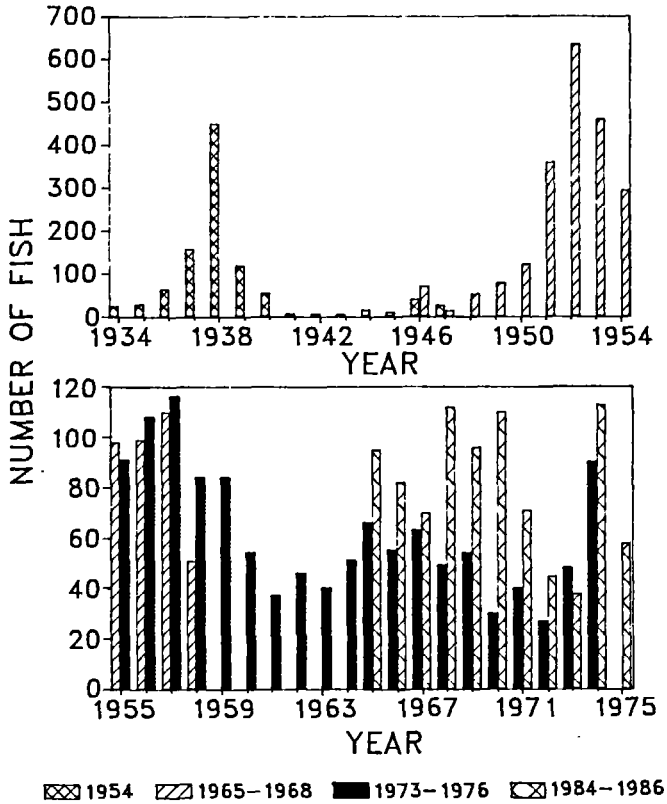


Figure 7 - Recruitment time series from white sturgeon captured in the Sacramento-San Joaquin Estuary in 1954, 1965-1968, 1973-1976, and 1984-1986 plotted on the same axis. Note change of scale on the y-axis between graphs (from Shirley 1987).

Fluctuations in legal-sized white sturgeon abundance have been dependent on recruitment, and not mortality, because mortality rates have been low and relatively stable over most of the period of record. However, mortality now may be increasing. Annual exploitation rate, a major component of total annual mortality rate, has increased from a mean of 0.069 in the 1960's and 1970's to 0.097 in the 1980's. This 41% increase in harvest rate has resulted from burgeoning popularity of the fishery and greater sophistication of anglers in locating fish with sonar and inducing them to bite. The increase in maximum likelihood estimates of annual mortality rate from 0.16 (survival = 0.84) in 1967 to 0.26 (survival = 0.74) in 1984 hints at the effect of the expanded fishery on the sturgeon population, although the correlation between

exploitation rate and total mortality rate ($r=0.38$, $n=6$, $p>0.10$) is not statistically significant.

Our knowledge of the dynamics of the white sturgeon population in the Sacramento-San Joaquin Estuary has implications for water development and management and for fishery management: flows must be adequate to maintain recruitment and fishing regulations must offer sufficient protection to keep harvest rates low and maintain spawning stocks. Unfortunately, data on year class strength are still inadequate (Figure 5) to specify to agencies that regulate water development the flow requirements that are necessary to optimize sturgeon production. We can only qualitatively observe that high flows produce good year classes. Because of this insufficient knowledge, the California Department of Fish and Game has initiated a program to develop better indices of year class abundance from catches of 1- to 5-year-old sturgeon captured in a small-mesh gill netting survey centered in Suisun Bay. Anticipated large catches of juveniles in this gear should yield more reliable indices than provided by the relatively low trawl catches and should allow better quantification of the relationship between year class strength and environmental variables.

Changes in angling regulations to reduce white sturgeon harvest are in order. While we do not know exactly the maximum harvest that the population can support without affecting both present status and future recruitment, the 40% increase in annual harvest rate in the 1980's and its predicted effect on egg production are cause for concern. Sturgeon are readily vulnerable to overharvest and subsequent decline (Bijkov, 1949 ; Dees, 1961) because they cannot rapidly compensate for unusually high mortality. To reduce the possibility of lower future recruitment we wish to maintain egg production at an adequate level. Changes in angling regulations that are being considered to attain this objective include increasing the minimum size limit, imposing a maximum size limit, initiating a "slot limit" that would protect fish in a given size range, and season or area closures.

In the future, management of the white sturgeon resource may require new approaches. These include evaluating the sensitivity of conclusions to parameter estimates and evaluating yield in terms other than biomass, which may not be the best indicator of value for a recreational fishery. In addition, we might attempt to regulate the population at the size limit and mortality rate that maximized yield under the constraint that the current (or higher) level of egg production be maintained (Botsford and Hobbs, 1987). However, even maintaining egg production at a specified level does not guarantee persistence of the population; we must also consider changes in the age structure. Broad, flat age structures provide more stable populations and lower extinction probabilities than narrow peaked ones (Botsford, 1986 ; Murphy, 1968) in variable environments. In addition to the effects of increased harvest on this population, we will consider, in this context, the influences of changes in the random environment as it affects sturgeon recruitment, particularly changes in magnitude and variability of freshwater flows.

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DISTINCTION DES STOCKS D'ESTURGEON JAUNE (*Acipenser fulvescens*) DU LAC DES DEUX MONTAGNES ET DU COULOIR FLUVIAL DU SAINT-LAURENT AU MOYEN DE L'ETUDE DES DEPLACEMENTS, DE LA CROISSANCE ET DE LA MORTALITE

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RESUME

Dans le but de vérifier l'homogénéité des groupements sympatriques d'Esturgeons jaunes de différents secteurs du couloir fluvial du Saint-Laurent et du lac des Deux Montagnes, les déplacements, la croissance et la mortalité totale ont été comparés. Le groupement du lac des Deux Montagnes se distingue très nettement des autres par un degré de sédentarité plus élevé, par une croissance plus lente et un embonpoint plus faible. Contrairement aux esturgeons du bassin de Laprairie, ceux des lacs Saint-Louis et Saint-Pierre affichent un degré de sédentarité relativement élevé, bien que des déplacements significatifs d'individus aient été observés entre les deux secteurs. Les déplacements des esturgeons qui furent étiquetés sur les frayères des rivières des Prairies, l'Assomption et Ouareau, de même que sur certains sites de concentration pré- et post-fraie sont beaucoup plus importants que ceux des individus étiquetés en d'autres endroits ; en effet, des recaptures de ces poissons ont été effectuées dans l'ensemble du couloir fluvial, incluant le lac Saint-Louis et le lac Saint-Pierre. Certaines différences ont été observées entre les groupements du lac Saint-Louis, du lac Saint-Pierre et de la partie du fleuve Saint-Laurent à l'aval du lac Saint-Pierre, au niveau de la structure de la récolte commerciale en taille, masse et âge, de la croissance et de la mortalité totale. L'exploitation commerciale plus intensive au lac Saint-Pierre pourrait être responsable de certaines de ces différences. Ces particularités, couplées aux résultats relatifs aux déplacements, justifient l'attribution, aux fins de la gestion et de l'aménagement, du statut de stock aux groupements d'esturgeons du lac des Deux Montagnes, du lac Saint-Louis (incluant le bassin de Laprairie), du lac Saint-Pierre et du fleuve Saint-Laurent à l'aval du lac Saint-Pierre.

Mots clés : *Acipenser fulvescens*, migrations, frayères, sédentarité, aires d'alimentation, variabilité intersectorielle, stock sympatrique, pêcherie commerciale, gestion.

ABSTRACT

In order to study the homogeneity, within the same hydrographic system, of the lake sturgeon populations of the Saint-Laurent River and of Lac des Deux Montagnes, movements, growth and total mortality were compared. The Lac des Deux Montagnes sturgeon differ from all other groups by their higher degree of sedentarity, slower growth and lower condition factor. Contrasting with those of Bassin de Laprairie, Lac Saint-Louis and Lac Saint-Pierre sturgeon show a relatively high degree of sedentarity, although significant movements of fish between these last two sectors were observed. Movements of sturgeon tagged on the Rivière des Prairies and Rivière l'Assomption spawning grounds, and also on pre- and post- spawning concentration sites are more important, recaptures having occurred all over the fluvial corridor, from Lac Saint-Louis to Lac Saint-Pierre. Length, weight and age composition of the commercial catch, growth and total mortality showed some variation between sectors. Higher commercial exploitation rates in Lac Saint-Pierre could be responsible for some of these differences. Observed differences in growth and total mortality, coupled with results on movements, justify that sturgeon from Lac des Deux Montagnes, Lac Saint-Louis (including Bassin de Laprairie), Lac Saint-Pierre and Saint-Laurent River downstream from Lac Saint-Pierre be considered as distinct stocks for management purposes.

Key-words : *Acipenser fulvescens*, migrations, spawning area, sedentarity, feeding ground, sympatric stocks, commercial fisheries, management.

INTRODUCTION

La pêcherie commerciale d'Esturgeon jaune du couloir fluvial du Saint-Laurent, dans le secteur du lac Saint-Louis (LSTL), du Bassin de Laprairie (BL), du lac Saint-Pierre (LSTP) et du fleuve Saint-Laurent en aval du lac Saint-Pierre (FSTL) (figure 1), dont le tonnage annuel est de l'ordre de 100 à 200 t (15 000 à 30 000 individus), est la plus importante en Amérique du Nord. Depuis le début des années 1980, face à l'accroissement des demandes de permis de pêche commerciale, plusieurs travaux ont été effectués sur la biologie des populations de cet Acipenséridé d'eau douce (Desjardins, 1985 ; Dumont et al., 1987 ; Léveillé et Mailhot, sous presse). Des études similaires avaient été menées, entre 1964 et 1979, au lac des Deux Montagnes (LDMT), dans le but de suivre le processus de restauration de la population d'esturgeons, fortement décimée suite à une période prolongée d'anoxie survenue au cours de l'hiver 1950 (Mongeau et al., 1982). Le présent travail utilise les résultats de ces études, quant aux déplacements, à la croissance et à la mortalité, dans le but d'obtenir une réponse à la question suivante : les groupements d'esturgeons du couloir fluvial du Saint-Laurent (CFSL) et du LDMT doivent-ils ou non, être considérés comme un stock unique aux fins de la gestion de la pêche commerciale ? La réponse à cette question est essentielle à l'élaboration de modalités de gestion et d'aménagement de cette importante ressource naturelle. Compte tenu des critères de distinction que nous nous proposons d'utiliser, le statut de stock distinct sera attribué à un groupement de poissons caractérisé par une stabilité spatio-temporelle et par une homogénéité au niveau des paramètres de production naturelle, justifiant ainsi l'application des modèles utilisés en gestion des pêches (Gulland, 1969).

TERRITOIRE A L'ETUDE

Le territoire à l'étude (fig. 1) couvre une superficie en eau d'environ 980 km^2. A l'ouest, il est limité par le barrage de Carillon, situé à l'amont du LDMT, et par le barrage de la Pointe-des-Cascades, situé à l'amont du LSTL. La fin des battures de Gentilly, situées à environ 20 km à l'aval de la ville de Trois-Rivières, en constitue la limite est. La distance maximale entre les deux extrémités est d'environ 240 km. Ce territoire comprend un réseau complexe de chenaux, de rivières, d'îles et de lacs de plaine formés par des élargissements de la rivière des Outaouais (LDMT) ou du fleuve Saint-Laurent (LSTL, BL et LSTP).

Des eaux brunes, relativement acides de plusieurs tributaires de la rive nord, issus du bouclier canadien, alimentent le LDMT et parviennent au fleuve Saint-Laurent dans le territoire à l'étude où elles s'écoulent le long de sa rive nord.

Des eaux vertes, transparentes et alcalines, en provenance des Grands Lacs, s'écoulent au centre et le long de la rive sud du LSTL, du BL et du fleuve Saint-Laurent. Aucun obstacle physique, naturel ou artificiel n'empêche le passage des poissons d'une étendue d'eau à l'autre.

Les eaux de ce secteur sont généralement bien oxygénées mais elles sont sérieusement contaminées par des rejets toxiques, des éléments nutritifs et des microorganismes. A l'exception de quelques fosses naturelles et du chenal maritime du Saint-Laurent, qui est creusé et entretenu à une douzaine de mètres, la profondeur est généralement inférieure à six mètres. Sauf dans des baies et des secteurs abrités, la vitesse du courant est relativement élevée : elle peut atteindre de 1 à 2 $m.s^{-1}$ dans le chenal maritime et plus de 4 $m.s^{-1}$ dans les secteurs de rapides. La faune piscicole y est abondante et diversifiée, particulièrement dans les secteurs qui furent le moins affectés par la perte d'habitats riparien. Plus de 90 espèces dulçaquicoles, anadromes ou catadromes y ont été répertoriées (Mongeau et al., 1974).

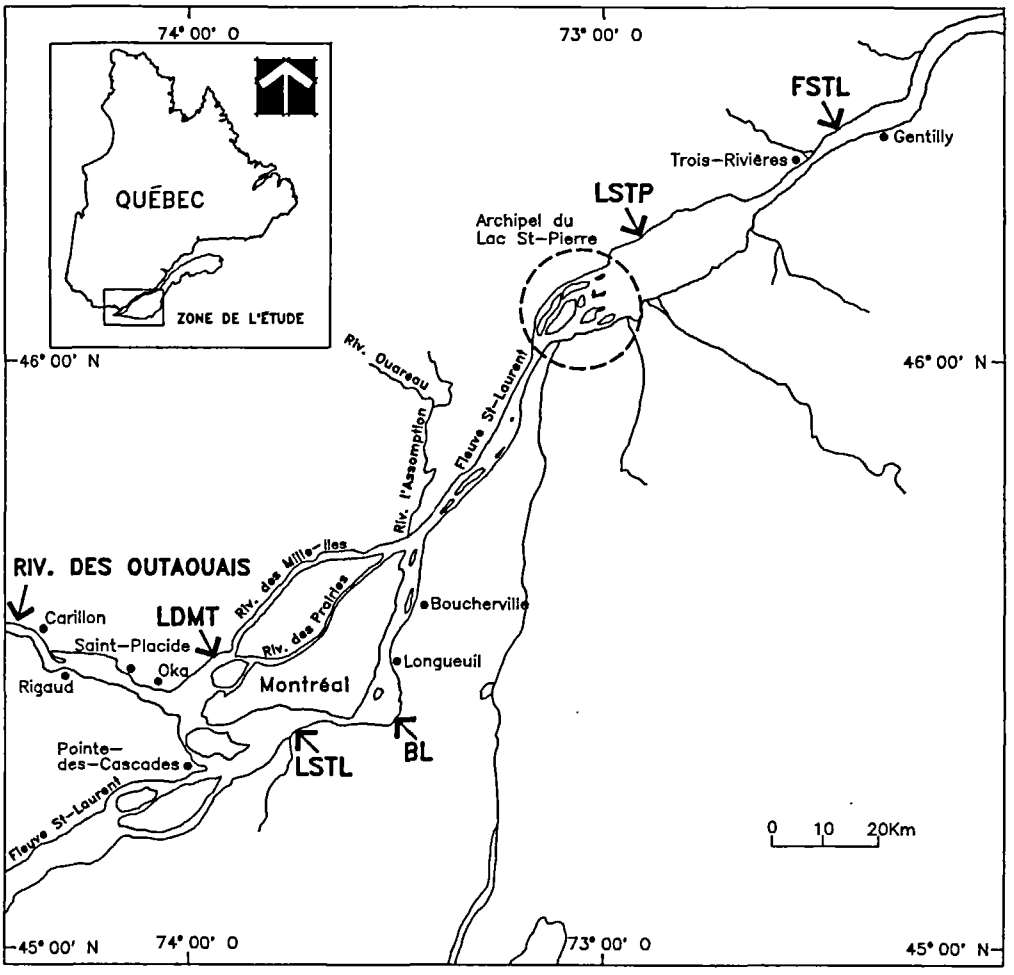


Figure 1 - Localisation du territoire à l'étude. LDMT : lac des Deux Montagnes ; LSTL : lac Saint-Louis ; BL : bassin de Laprairie ; LSTP : lac Saint-Pierre ; FSTL : fleuve Saint-Laurent en aval de Trois-Rivières.

MATERIEL ET METHODES

Déplacements

Le tableau 1 montre que tous les spécimens étiquetés, à l'exception de 200 rejets de la pêche commerciale (rejets p. com.), ont été capturés par nos équipes de terrain, à l'occasion de pêches expérimentales au filet maillant. Trois types d'étiquettes, toutes fixées au travers de la base des premiers rayons de la nageoire dorsale, ont été utilisés. Il s'agit de l'étiquette spaghetti fixée selon la méthode de White et Beamish (1972) (n=7244, tableau 1), de l'anneau métallique (n=2224) et de l'étiquette spaghetti avec ancrage nylon en T (n=520). Les deux premiers types d'étiquettes, qui furent les plus utilisés, affichent probablement un taux de rétention satisfaisant puisque des recaptures ont été enregistrées plus de 10 ans après le marquage.

Tableau 1 - Etiquetages d'Esturgeons jaunes au lac des Deux Montagnes (LDMT) et dans le couloir fluvial du Saint-Laurent (CFSL) en vue de l'étude des déplacements. FM : filet maillant. W&B : White et Beamish (1972). Deux cents individus de taille sublégale (rejets p.com.) ont été étiquetés en 1981-1982.

Endroit	Années / mois	Mode de pêche	Etiquette employée	n	Auteurs
LDMT	1964 à 1969 Juillet-octobre	Expérimentale FM 3.8-25.4 cm	Anneau métallique	2224	Mongeau et al. (1982)
LDMT	1978,1983,1985 Juillet-octobre	Expérimentale FM 3.8-25.4 cm	Spaghetti W&B	1972	Mongeau et al.(1982) Rousseau, M. (Comm. pers.)
CFSL	1981 à 1986 Mai-juin	Expérimentale FM 15-30.5 cm + rejets p.com.	Spaghetti W&B	5272	Dumont et al. (1987) Desjardins (1985)
CFSL	1981,1982 Mai-juin	Expérimentale FM 15-30.5 cm +rejets p.com.	Spaghetti ancrage T	520	Desjardins (1985) Dumont et al. (1987)

Au LDMT, 4196 individus (3396 de 1964 à 1978 ; 800 en 1983 et 1985) dont environ la moitié mesuraient plus de 800 mm (longueur totale minimale permise pour la pêche commerciale dans le CFSL) ont été étiquetés en trois sites principaux, soit Rigaud, Saint-Placide et Oka. Dans le CFSL, le marquage fut étendu à tout le territoire à l'étude, excepté au FSTL. Cependant, comme en témoignent les nombres d'individus étiquetés aux différents sites (figures 2 et 3), l'effort déployé n'a pas eu partout la même intensité. Sur le total de 5792 spécimens étiquetés, qui mesuraient pour la plupart au-delà de 750 mm, environ 34% l'ont été sur les aires d'alimentation, lesquelles sont également des secteurs de pêche commerciale (LSTL, BL et LSTP, figure 1) et 66% sur des sites utilisés comme frayères ou comme lieux de concentration pré- et post-fraie (Rivières des Prairies, l'Assomption et Ouareau, fleuve Saint-Laurent au niveau de Longueuil et des îles de Boucherville, figure 1). Les spécimens furent pesés et mesurés (longueur totale, longueur à la fourche) préalablement à leur remise à l'eau.

Les recaptures ont été effectuées principalement par nos équipes de terrain à l'occasion des pêches réalisées en vue de l'étiquetage, et par les pêcheurs commerciaux qui, à compter de 1983, se sont vu offrir une récompense de cinq dollars par recapture. Quelques recaptures ont été rapportées par les pêcheurs sportifs. Tous les pêcheurs commerciaux de la rivière des Outaouais et du CFSL furent contactés chaque année (1981-1986) par téléphone ou par courrier, et ceux du territoire de l'étude furent régulièrement visités individuellement. Des affiches décrivant le projet ont en outre été disposées sur les sites des principaux débarcadères publics, utilisés par les pêcheurs sportifs, de même que sur ceux des pourvoyeurs de pêche.

Caractérisation des captures

Au LSTL, au BL, au LSTP et au FSTL, les groupements ont été caractérisés au moyen d'échantillons prélevés à même la récolte commerciale. L'examen des spécimens a eu lieu aux points de débarquement. Les deux principaux engins de pêche commerciale étaient le filet maillant (dimension minimale permise de la maille étirée : 19 cm ; mailles les plus utilisées : 19 et 20 cm) et la ligne dormante (dimension des hameçons variant de 7/0 à 9/0). La caractérisation a eu lieu principalement en 1981, 1982, 1985 et 1986 au LSTL, en 1981 et 1982 au BL, en 1984 au LSTP et en 1988 au FSTL (tableau 2). Notons qu'en ce dernier site, les spécimens provenaient d'une pêche expérimentale menée en collaboration avec un pêcheur commercial, et réalisée au moyen de filets maillants (mailles étirées de 18.0, 20.3 et 21.0 cm).

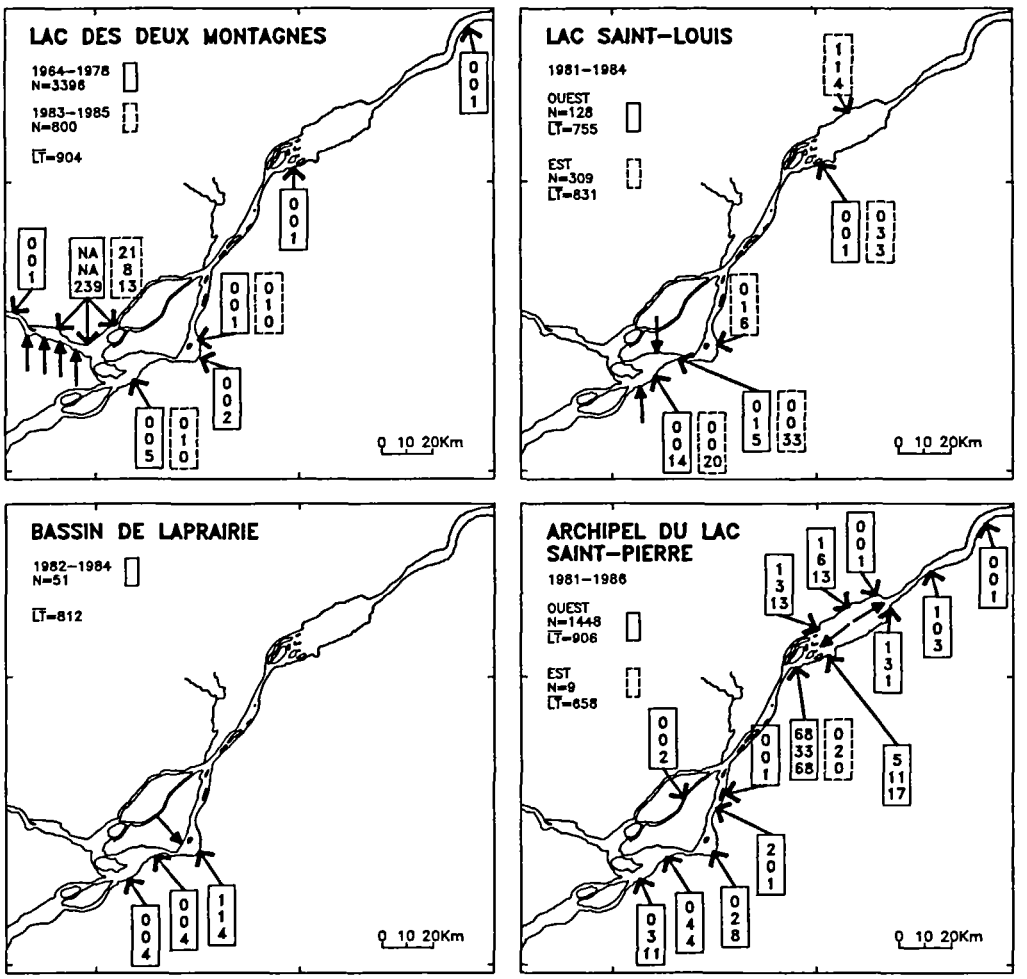


Figure 2 - Localisation des recaptures des esturgeons étiquetés dans les secteurs de pêche commerciale et au lac des Deux Montagnes. Le nombre ainsi que la taille moyenne des poissons étiquetés sont indiqués. Les flèches à pointe étroite identifient le lieu de marquage ; les flèches à pointe large accompagnant les rectangles identifient les lieux de recapture. Pour chaque site de recapture, le premier nombre indiqué dans le rectangle correspond aux recaptures effectuées moins de 30 jours après le marquage, le second, à celles effectuées entre 30 et 299 jours et le troisième, à celles effectuées après au moins 300 jours. N.A. : données non disponibles.

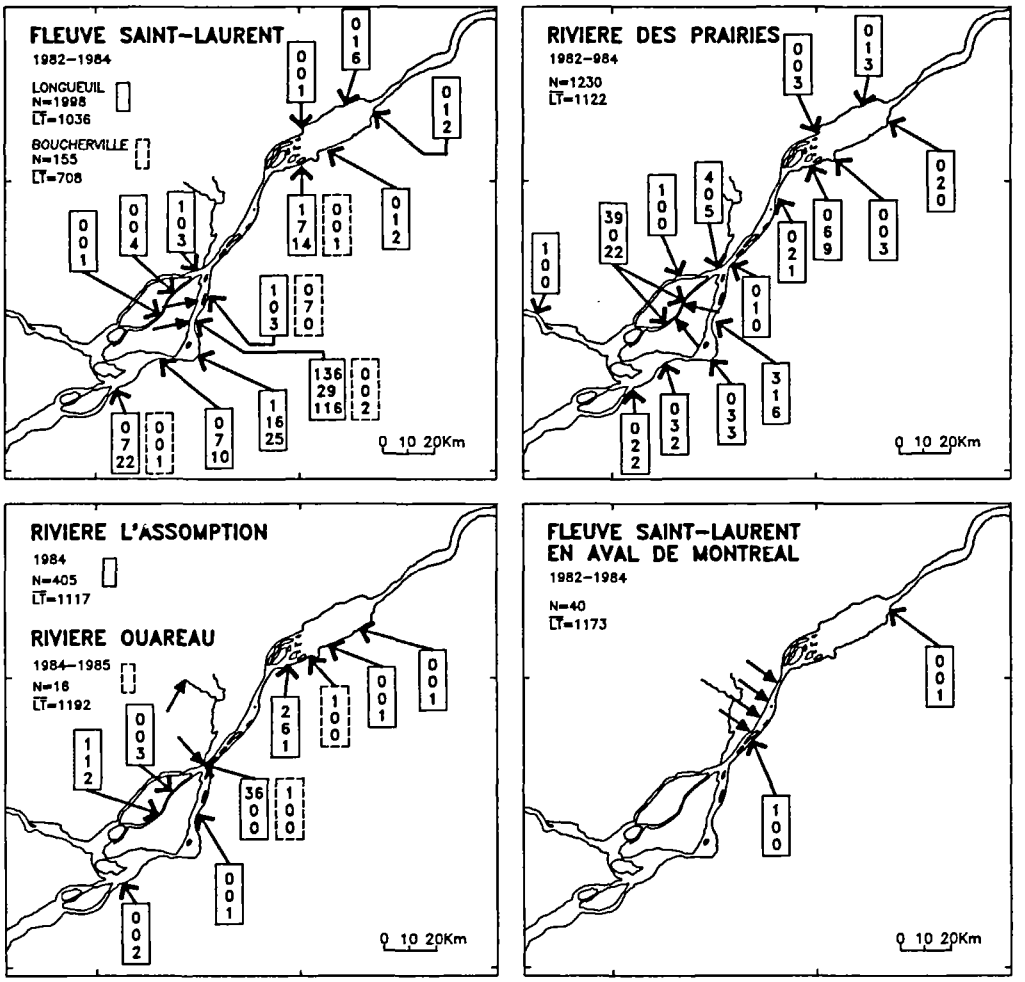


Figure 3 - Localisation des recaptures des esturgeons étiquetés sur les frayères ou sur les sites de concentration pré- et post- fraie. Légende : voir figure 2.

Compte tenu du faible effectif, les poissons de ce secteur ont été regroupés indépendamment de l'engin de pêche aussi bien pour les comparaisons des caractéristiques de la récolte commerciale que pour le calcul du taux de mortalité totale. L'échantillon du LDMT comprend uniquement des spécimens capturés à la pêche expérimentale (filets maillants à mailles étirées variant de 3.8 à 25.4 cm) ; il inclut les spécimens étiquetés en 1985 mentionnés au tableau 1.

Tableau 2 - Répartition du nombre de spécimens selon les secteurs de pêche, les années de collection et les données traitées. LT, longueur totale ; M, Masse ; FM, Filet Maillant ; LD, Ligne Dormante. LSTL : lac Saint-Louis ; BL : bassin de Laprairie ; LSTP : lac Saint-Pierre ; FSTL : fleuve Saint-Laurent, aval du lac Saint-Pierre ; LDMT : lac des Deux Montagnes.

Secteur de pêche	Années	Engin	LT	M	Age
LDMT	1964	FM	786	786	3
	1965	FM	240	240	1
	1966	FM	312	312	1
	1968	FM	565	563	44
	1969	FM	332	332	20
	1978	FM	1310	1309	1298
	1979	FM	599	3	
	1985	FM	721	718	
LSTL	1981	FM	181	2	249
	1982	FM,LD	639	580	427
	1983	FM	56		
	1984	FM,LD	28	28	193
	1985	FM	553	550	524
	1986	LD	194	194	189
BL	1981	FM	41	15	49
	1982	FM	312	270	129
	1983	FM	8	8	8
LSTP	1981	FM	73	30	100
	1984	FM,LD	1772	1754	995
FSTL	1988	FM	431	431	399

Les spécimens échantillonnés furent mesurés (longueur totale, longueur à la fourche) et pesés. Le sexe fut déterminé par examen macroscopique des gonades. Le rayon marginal de la nageoire pectorale fut prélevé en étant sectionné le plus près possible du corps. Ultérieurement, des coupes d'environ 450 à 500 µ d'épaisseur, pratiquées transversalement au rayon, furent utilisées pour la détermination de l'âge suivant la méthode utilisée par Cuerrier (1951), Probst et Cooper (1954) et Roussow (1957). Certains pêcheurs commerciaux ont collaboré en prélevant eux-mêmes les rayons. Dans ce cas, seul l'âge des spécimens étant disponible, les données ne purent être utilisées que pour l'évaluation de l'âge moyen et des taux de mortalité. Les quantités de poissons qui ont été mesurées, pesées et dont l'âge a été déterminé figurent au tableau 2.

Traitement statistique

Les comparaisons des moyennes des différentes variables considérées ont été effectuées au moyen de tests non paramétriques (Kruskal-Wallis) ou paramétriques (Student-Newman-Keuls) suivant que les distributions de fréquence différaient ou non

de la normalité (tests de Kolmogorov-Smirnov). Les différences dans les distributions de fréquence des variables ont été éprouvées au moyen de tests de Chi^2 (X^2). Les taux annuels de mortalité totale ont été estimés à partir de la pente des courbes de capture des deux principaux engins de pêche commerciale, ainsi qu'au moyen de la méthode de Robson et Chapman, qui permet aussi l'évaluation de la variance et de l'intervalle de confiance (Ricker, 1975). Des analyses de covariance ont permis la comparaison de la relation masse-longueur, entre les secteurs. Toutes les comparaisons statistiques ont été effectuées au moyen du progiciel SPSSX. Les paramètres des équations de von Bertalanffy ont été estimés au moyen du programme FISHPARM, de la banque FSAS (Saila et al., 1988).

Du fait qu'à l'intérieur de la distribution des tailles et des âges des spécimens, il n'existait généralement pas de différences significatives entre les deux sexes, quant à la plupart des variables considérées, les comparaisons inter-sectorielles furent effectuées en regroupant tous les individus. Au BL et au LSTP, nous n'avons pas observé de différences inter-annuelles significatives au niveau des tailles, poids et âges moyens de la récolte. Au LSTL et au LDMT, certaines différences ont été relevées. Toutefois, comme celles-ci s'avéraient généralement moindres que celles observées entre les secteurs, les comparaisons inter-sectorelles furent effectuées en cumulant les données disponibles, indépendemment de l'année de capture.

RESULTATS

Les déplacements

La capture et l'étiquetage sont susceptibles d'influencer les déplacements des poissons durant les semaines suivant leur remise à l'eau. Nous avons donc considéré, dans les cas des esturgeons marqués entre 1981 et 1986 dans le CFSL et entre 1983 et 1985 dans le LDMT, trois intervalles de temps pour la compilation des recaptures : moins de 30 jours après l'étiquetage (n=325), de 30 à 299 jours (n=188) et 300 jours ou plus (n=536). Dans la présentation des résultats relatifs aux déplacements, nous utiliserons surtout les recaptures effectuées au moins 30 jours après le marquage. Neuf cent soixante dix individus furent repris une fois, 71, deux fois, sept, trois fois et un, quatre fois. De ce total de 1049 recaptures, 503 ont été effectuées par nos équipes de terrain, 508 par les pêcheurs commerciaux et 38 par les pêcheurs sportifs. Dans les cas des esturgeons étiquetés au LDMT entre 1964 et 1978, les recaptures réalisées l'année même du marquage n'ont pas été utilisées pour l'étude des déplacements ; seules les 250 recaptures réalisées au moins un an après le marquage l'ont été.

Parmi les 1299 recaptures, incluant celles du LDMT, 855 ont eu lieu sur le site de marquage. Toutefois, si l'on ne considère que les recaptures effectuées au moins 30 jours après le marquage, ces chiffres deviennent respectivement 577 et 387. La figure 2 montre que, dans le cas des sites de marquage correspondant aux principaux secteurs de pêche commerciale et d'alimentation, la proportion de recaptures dans le secteur d'origine, au-delà de 30 jours après le marquage, est relativement élevée soit : 95% (260 sur 273) au LDMT, 83% (73 sur 88) au LSTL et 81% (171 sur 211) au LSTP. Par contre, au BL, la situation est différente, puisque la proportion correspondante n'est que de 38% (5 sur 13). On note par ailleurs que les esturgeons ont tendance à se déplacer en demeurant dans leur plan d'eau d'origine ; il en est ainsi notamment des déplacements observés entre les secteurs est et ouest du LSTL, entre l'Archipel du LSTP et les secteurs est et ouest de ce dernier et entre les secteurs d'Oka, de Saint-Placide et de Rigaud à l'intérieur du LDMT (figure 2).

Chez les esturgeons étiquetés dans le fleuve Saint-Laurent, sur le site de Longueuil, considéré comme secteur de concentration pré- et post-fraie, le pourcentage de recaptures à l'endroit même du marquage est plus faible, soit 52% (145 sur 281) (figure 3). Par contre, sur les deux sites de fraie où des nombres significatifs d'esturgeons furent étiquetés, soit celui de la rivière des Prairies et celui de la rivière l'Assomption (cette dernière incluant les données relatives à l'un de ses tributaires,

la rivière Ouareau), les pourcentages de recaptures correspondants sont particulièrement faibles, soit 28% (22 sur 80) et 0% (0 sur 18), respectivement.

Pour ce qui est des 444 recaptures effectuées à un site autre que celui du marquage, la figure 2 montre que certains individus (moins de 20% des spécimens recapturés 30 jours ou plus après le marquage) quittent l'aire d'alimentation où ils ont été marqués (LSTL, LSTP, LDMT) pour être repris en divers secteurs du couloir fluvial, incluant l'extrémité opposée de l'aire d'étude. De tels échanges entre les secteurs d'alimentation et de pêche commerciale impliquent surtout les esturgeons du LSTL et du LSTP, tandis que les poissons du LDMT se déplacent beaucoup moins. Notons que les spécimens marqués dans l'Archipel du LSTP ont tendance à être recapturés en plus grand nombre vers l'aval que vers l'amont (83 et 38 individus respectivement).

La figure 3 montre que les sites de recapture des esturgeons étiquetés dans les secteurs de fraie ou de concentration pré-et post-fraie sont beaucoup plus diversifiés. Par exemple, certains individus étiquetés à la rivière des Prairies ont rejoint et remonté le Saint-Laurent (n=25) pour être recapturés à Longueuil, au BL et au LSTL ; un nombre à peu près équivalent (n=31) a été repris en aval de Montréal, dans le couloir fluvial et au LSTP. Quant aux poissons marqués dans le bassin de la rivière l'Assomption, la figure 3 montre qu'ils ont été repris en plus grand nombre en aval de l'île de Montréal (n=12) qu'en amont (n=3). Des nombres variables d'individus marqués sur une certaine frayère une année donnée ont été repris moins de trente jours, ou plus de 300 jours plus tard, sur une autre frayère (figure 3). Quant aux esturgeons marqués au site de Longueuil, 88 furent repris en amont et 49 en aval.

Principales caractéristiques biologiques des groupements d'esturgeons

Compte tenu 1- du nombre plutôt restreint de spécimens disponibles en provenance du BL, 2- de l'absence de différences significatives observées entre ces poissons et ceux du LSTL, quant à la plupart des variables considérées, 3- du fait que plus de la moitié des recaptures d'esturgeons marqués dans ce secteur ont été effectuées au LSTL, 4- de la proximité géographique entre le BL et le LSTL, les esturgeons de ces deux secteurs de pêche commerciale ont été regroupés aux fins de comparaison des caractéristiques biologiques entre les différents groupements. Dès lors, les groupements considérés sont les suivants : LDMT, LSTL (incluant le BL), LSTP et FSTL.

Un premier classement des groupes de spécimens des quatre secteurs, par ordre croissant de taille, de poids et d'âge moyens des captures réalisées à l'aide du filet maillant de 200 mm de maille étirée, est le suivant : LSTP, LDMT, FSTL et LSTL (tableau 3). Ces variables diffèrent significativement ($p<0.05$) entre les quatre secteurs considérés globalement. Lorsque comparés entre eux, les principaux secteurs de pêche commerciale (LSTL, LSTP, tableau 3) diffèrent significativement au niveau des trois variables en cause ; il en est de même dans le cas du LSTP et du FSTL. Le LDMT affiche aussi quelques différences significatives par rapport aux autres secteurs (tableau 3). Le tableau 3 montre en outre que la taille, le poids et l'âge moyens des spécimens capturés à la ligne dormante au LSTL sont significativement supérieurs ($p<0.05$) à ceux des spécimens du LSTP.

En ce qui a trait à la croissance en longueur, la grande variation dans l'étendue des tailles observées aux différents âges est notoire, comme en témoignent les valeurs élevées de l'écart-type (tableau 4). De plus, l'accroissement de la taille moyenne aux différents âges est très irrégulier. L'ordre croissant des tailles moyennes à l'intérieur des quatre groupements, entre les âges de 9 à 13, est généralement le suivant : LDMT, LSTP, FLSL et LSTL. Cependant, à compter de la 14ème année, l'ordre est inversé dans le cas des deux derniers plans d'eau.

On observe en outre que pour huit des groupes d'âge 8 à 19, les esturgeons du LDMT présentent des tailles moyennes significativement plus petites ($p<0.05$) que celles des esturgeons de chacun des trois autres groupements; par ailleurs, les esturgeons du LSTP sont fréquemment significativement plus petits ($p<0.05$) que ceux du LSTL (huit groupes d'âge) et du FLSL (huit groupes d'âge). Enfin, les esturgeons du LSTL présentent des tailles moyennes supérieures à celles de poissons du FLSL dans le cas de

Tableau 3 - Longueur totale (L, mm), masse (M, kg) et âge (A, années) moyens des Esturgeons jaunes capturés au filet maillant de 200 mm de maille étirée et à la ligne dormante dans les différents secteurs du couloir fluvial du Saint-Laurent et au lac des Deux Montagnes (LDMT). LSTL : lac Saint-Louis (incluant BL) ; LSTP : lac Saint-Pierre ; FLSL : fleuve Saint-Laurent en aval du lac Saint-Pierre. X, sx, n : moyenne, écart-type et effectif. $p<0.05$: groupes différant entre eux au niveau de leurs moyennes (tests de Kruskal-Wallis, KW) et/ou de leur distribution de fréquence (tests de X^2).

	LDMT (1)			LSTL (2)			LSTP (3)			FSTL (4)			
	X	sx	n	X	sx	n	X	sx	n	X	sx	n	p<0.05
Filet maillant de 200 mm													
L	958	175	152	992	121	1662	925	133	1665	990	130	431	KW : 12,13,14,23,34 X^2 : 12,13,14,23, 24,34
M	5.5	2.6	135	6.9	3.6	1389	5.6	2.9	1612	6.6	2.9	431	KW : 12,14,23,34 X^2 : 12,14,23,24,34
A	15.0	5.0	47	15.8	4.6	1419	14.1	3.4	937	14.7	2.6	399	KW : 13,14,23,24,34 X^2 : 13,14,23,24,34
Ligne dormante													
L				1087	214	253	974	144	175				KW: 23 X^2: 23
M				10.1	9.5	175	6.6	4.0	175				KW: 23 X^2: 23
A				20.3	9.2	366	14.8	4.4	158				KW: 23 X^2: 23

deux groupes d'âge (10,11), mais inférieures dans le cas de quatre groupes d'âge (15,16,18,19). Les paramètres des équations de von Bertalanffy diffèrent aussi d'un secteur à l'autre : L_∞ varie de 1477 à 2240 mm, K, de 0.022 à 0.050 et t_o, de -4.9 à -12.3 (figure 4). Toutefois, il apparaît que les différences observées sont attribuables en partie au fait que l'étendue des groupes d'âge disponibles pour l'estimation des paramètres varie beaucoup d'un secteur à l'autre.

Lorsqu'ils sont considérés globalement, les quatre groupes présentent des différences significatives ($p<0.01$) au niveau de la pente de la relation masse-longueur (tableau 5). Ces différences sont causées principalement par deux d'entre eux, soit LSTL (pente : 3.36) et LDMT (3.25). On note également que les quatre groupes diffèrent entre eux ($p<0.01$) au niveau de l'ordonnée à l'origine.

Les variations de la croissance en masse, obtenue en évaluant, au moyen des équations du tableau 5, la masse moyenne correspondant aux longueurs moyennes observées aux différents âges (tableau 4), reflètent bien les variations observées au niveau de la croissance en longueur (figure 4).

Les différences observées au niveau de la relation masse-longueur se traduisent par des variations inter-sectorielles des valeurs moyennes de la masse et du coefficient de condition de Fulton calculées aux différentes tailles. Le tableau 5 montre tout d'abord qu'à l'intérieur de chaque groupe, en conformité avec l'allométrie de croissance suggérée par la pente des équations masse-longueur, le coefficient de condition augmente avec la taille. Les différences significatives ($p<0.05$, test S-N-K) relevées au niveau de la valeur moyenne du coefficient de condition, entre les secteurs, reflètent des différences sectorielles, à la fois au niveau de la longueur moyenne et de l'embonpoint. C'est pourquoi les différences de poids et de condition, entre les secteurs, peuvent être mieux évaluées en comparant ces variables par classe de longueur (tableau 5). L'ordre croissant du poids moyen et du coefficient moyen de condition observés pour les quatre secteurs est généralement le suivant : LDMT, FSTL et LSTP-LSTL, les deux derniers présentant des valeurs très semblables.

Tableau 4 - Longueurs moyennes observées aux différents âges pour les Esturgeons jaunes des lacs des Deux Montagnes (LDMT), Saint-Louis incluant le BL (LSTL) et Saint-Pierre (LSTP), ainsi que du fleuve Saint-Laurent en aval du lac Saint-Pierre (FSTL). $p<0.05$: groupes différant entre eux au niveau de leur longueur moyenne (test de Student-Neuman-Keuls).

	LDMT (1)			LSTL (2)			LSTP (3)			FSTL (4)			
Age	LT	sx	n	LT	sx	n	LT	sx	n	LT	sx	n	p<0.05
2	388	27	11										
3	468	20	9										
4	538	46	70										
5	599	48	249	493		1							
6	634	52	158				940	464	3				
7	663	85	19	804		1	710	35	2				
8	705	71	27	896	31	4	691	71	10				12,23
9	736	83	38	888	88	29	813	107	39	865	58	7	12,13,14,23
10	801	72	64	940	74	29	804	91	59	869	49	9	12,14,23,24,34
11	816	75	261	927	65	77	862	96	87	886	62	20	12,13,14,23,24
12	43	92	58	938	72	116	879	82	135	912	73	32	12,13,14,23,34
13	848	118	26	955	97	148	910	81	138	937	69	82	12,13,14,23,34
14	884	107	24	958	87	140	933	105	153	979	84	56	12,13,14,23,34
15	922	110	34	979	85	158	970	111	117	1020	89	51	12,13,14,24,34
16	936	95	36	1008	82	150	967	102	98	1052	88	47	12,14,23,24,34
17	956	106	67	1014	108	115	1013	113	97	1055	97	51	12,13,14
18	993	122	77	1021	104	95	1019	97	36	1110	97	31	14,24,34
19	981	90	39	1054	97	85	1050	115	24	1158	129	17	12,13,14,24,34
20	995	116	38	1032	102	62	1027	118	10	1103	106	8	-
21	1028	110	33	1062	134	51	1046	153	12	1200	149	4	24
22	1081	109	21	1079	122	28	1130	159	11	1236	39	2	-
23	1089	55	2	1137	119	23	1144	128	8	1100		1	-
24	1134	140	3	1192	174	18	1140	294	6	1331		1	-
25	1270		1	1141	115	16	1325	92	3				
26				1291	131	7	1288	56	4				
27				1307	158	15							
28				1210	141	6	1348	74	2				
29				1245	109	8	1165	7	2				
30				1394	63	6	1383	243	3				
31				1411	159	4							
33				1493	114	5	1432		1				
34				1784		1							
35				1410	57	2							

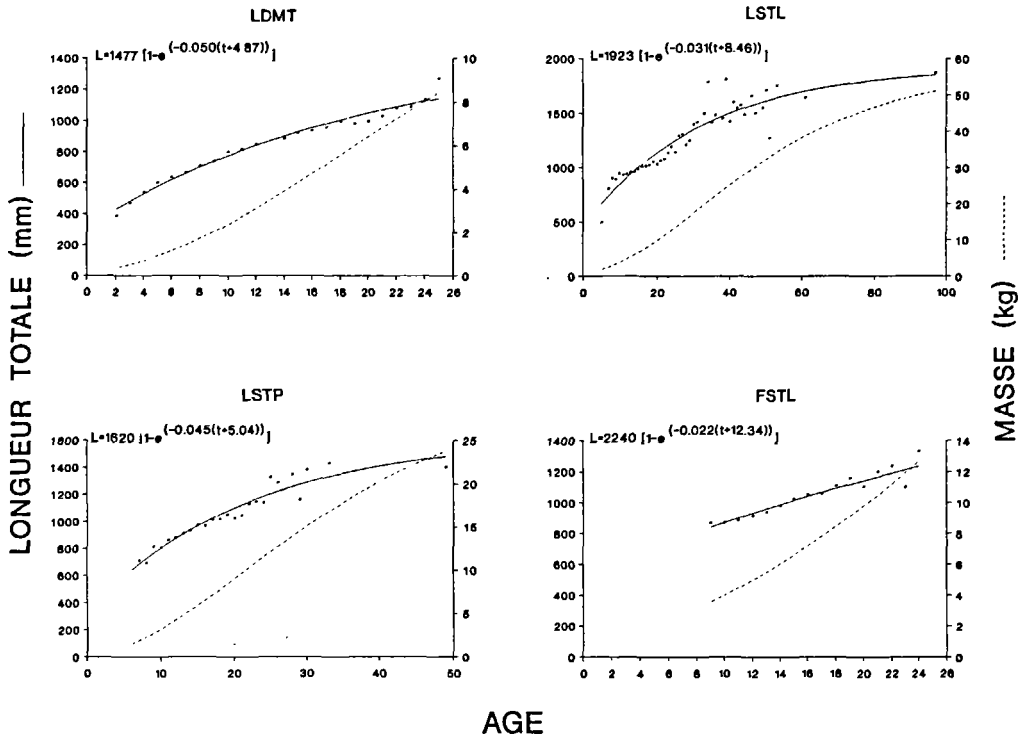


Figure 4 - Croissance en longueur et en masse des esturgeons des différents secteurs. Les points représentent les longueurs totales moyennes observées aux différents âges. La courbe en trait plein correspond à l'équation de von Bertalanffy présentée sur chaque graphique. La courbe en tireté représente la croissance en masse, générée à partir des équations de von Bertalanffy et des équations de la relation masse-longueur présentées au tableau 3. LDMT : lac des Deux Montagnes ; LSTL : lac Saint-Louis (incluant le BL) ; LSTP : lac Saint-Pierre ; FSTL : fleuve Saint-Laurent en aval du lac Saint-Pierre.

Tableau 5 - Comparaison des paramètres de la relation masse-longueur entre les secteurs (A) et variations sectorielles de la masse et du coefficient de condition de Fulton en fonction de la longueur (classes de 100 mm). Les différences statistiques au niveau de la pente et de la position des droites sont indiquées. ** : $p<0.01$; * : $p<0.05$. LDMT : lac des Deux Montagnes ; LSTL : lac Saint-Louis (incluant le BL) ; LSTP : lac Saint-Pierre ; FSTL : fleuve Saint-Laurent en aval du lac Saint-Pierre.

A) Relation masse-longueur : log (masse) = a + m log (longueur)

Secteur	a	m	r	n	Diff. pente 1 2 3 4	Diff. position 1 2 3 4
LDMT (1)	-6.00518	3.24735	0.99	4265	**	** ** **
LSTL (2)	-6.26055	3.35695	0.94	1648	*	** **
LSTP (3)	-6.00319	3.27363	0.96	1791		**
FSTL (4)	-6.12341	3.30782	0.92	431		

B) Valeurs moyennes de la masse (M) et du coefficient de condition de Fulton (K) par classe de longueur (L) de 100 mm:

	LDMT			LSTL			LSTP			FSTL		
L (mm)	M (g)	K	n	M (g)	K	n	M (g)	K	n	M (g)	K	n
250	85	0.37	2									
350	228	0.46	47									
450	458	0.47	227	500	0.42	1	375	0.34	2			
550	823	0.47	685	926	0.52	7	750	0.44	1			
650	1352	0.49	661	1383	0.45	3	1665	0.57	44	1450	0.50	6
750	2200	0.51	711	2995	0.63	14	2734	0.62	198	2767	0.60	12
850	3213	0.52	928	4032	0.62	280	4001	0.64	504	3835	0.61	84
950	4719	0.55	608	5505	0.64	617	5552	0.65	552	5390	0.63	140
1050	6354	0.55	286	7541	0.67	429	7497	0.66	306	7384	0.65	102
1150	8459	0.57	89	10001	0.67	175	10098	0.68	119	9730	0.66	58
1250	11559	0.60	19	13495	0.71	52	13108	0.68	36	13332	0.69	22
1350	12750	0.57	2	18980	0.78	34	18312	0.75	11	14645	0.59	5
1450				20665	0.69	20	21732	0.74	10	18750	0.67	1
1550				30635	0.84	12	27443	0.72	1			
1650				37300	0.84	5	30625	0.72	1			
1750				40833	0.80	3						
1850				56717	0.90	3						
Moyenne	2767	0.51	4265	7381	0.66	1648	5666	0.65	1791	6548	0.63	431

Signalons que des échantillonnages de spécimens mesurant entre 90 et 100 cm effectués au cours de l'été 1988, ont révélé des différences significatives, au niveau du coefficient K de Fulton ($p<0.05$, test S-N-K), entre les poissons du LDMT (0.50, n=27) et ceux de chacun des autres secteurs (LSTL : 0.63, n=38 ; LSTP : 0.60, n=40 ; FSTL : 0.65, n=27) ; des différences ont aussi été observées entre FSTL et LSTP ($0.01<p<0.05$).

Pour le LSTL, les valeurs du taux annuel de mortalité totale A, estimées à partir de la pente des courbes de capture au filet maillant de 200 mm, ont peu varié pour les différentes années d'échantillonnage : 0.20 en 1981 (âges 14-26), 0.23 en 1982 (âges 16-27) et 0.23 en 1985 (âges 16-29). La valeur de A pour la période 1981-1985 est de 0.21 (figure 5). L'estimé correspondant de Robson et Chapman est de 0.24 (écart type : 0.00082). En ce qui a trait aux captures à la ligne dormante, l'estimé de A, obtenu en regroupant les captures de 1982, 1984 et 1986 et en considérant les groupes d'âge 17 à

31 (figure 5), est de 0.13. Notons cependant que le segment de courbe correspondant aux âges 17 à 26, fournit un estimé équivalent à celui observé pour les captures au filet de 200 mm, soit 0.21.

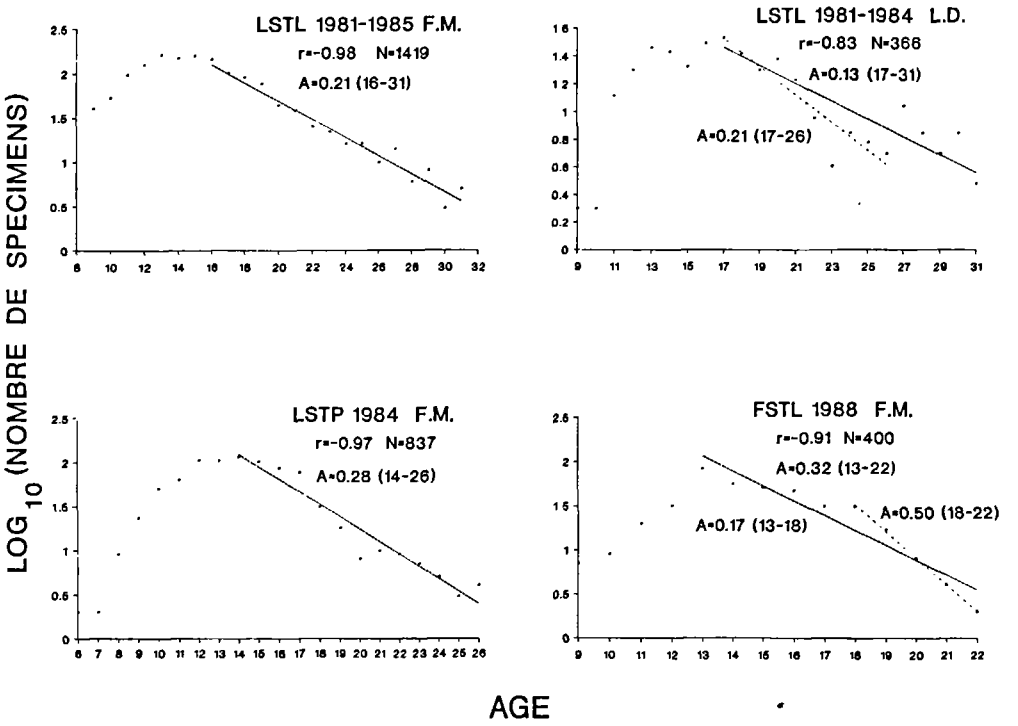


Figure 5 - Courbes de capture du filet maillant (F.M.) de 200 mm et de la ligne dormante (L.D.) utilisées pour le calcul du taux annuel de mortalité A. Chiffres entre parenthèses : premier et dernier âge utilisé dans le calcul ; r : coefficient de corrélation de Pearson ; N : nombre de spécimens impliqués. Secteurs: mêmes abréviations que figure 4.

Le membre descendant de la courbe de capture des esturgeons du LSTP (1984, figure 5) est beaucoup plus irrégulier que celui des esturgeons du LSTL. L'hypothèse d'une pente unique donne, pour les âges 14-26, un estimé de A de 0.28. Il s'agit en fait de la moyenne des valeurs qui peuvent être estimées sur les différents segments de droite correspondant aux âges 14-17 (0.13), 17-20 (0.52) et 21-26 (0.21). L'estimé de Robson et Chapman pour les âges 14-26 est de 0.30 (écart type : 0.0116). Il diffère statistiquement ($p<0.01$) de la valeur équivalente mentionnée plus haut pour les esturgeons du LSTL.

L'hypothèse d'une pente unique, dans le cas des esturgeons du FSTL, donne un estimé de A de 0.32 (figure 5). La courbe semble toutefois être mieux interprétée en considérant deux segments de droite, avec une valeur de A de 0.17 pour les âges 13-18 et de 0.50 pour les âges 18-22. La similitude entre les courbes de capture des esturgeons du FSTL et du LSTP mérite d'être signalée : dans les deux cas, on note un changement de pente très marqué vers les âges 17-18.

En regard de l'objectif défini au début de ce travail, nos résultats comportent certains biais au niveau des différentes variables considérées. En ce qui a trait aux déplacements, les biais sont reliés principalement à la distribution spatiale et temporelle inégale de l'effort de pêche, et à l'effort réduit de publicité à l'aval du territoire à l'étude. Ainsi, la probabilité pour un poisson d'être marqué et recapturé n'est pas répartie uniformément sur l'ensemble du territoire et de la période. Cependant, le fait que les principaux secteurs considérés, à l'exception du FSTL, aient été l'objet d'échantillonnages intensifs, permet de mieux comprendre les composantes "déplacements" et "sédentarité" du comportement de l'esturgeon dans le couloir fluvial du Saint- Laurent et dans le LDMT.

En ce qui a trait aux caractéristiques de la récolte, les principaux biais tiennent à l'inégalité de l'effort d'échantillonnage dans les différents secteurs ainsi qu'au regroupement de données provenant d'un nombre variable d'années. Ce regroupement nous est apparu justifié compte tenu du fait que dans le cas des secteurs comportant les plus longues séries de données (LDMT, LSTL), les variations intra-sectorielles étaient d'une amplitude plus faible que les variations inter-sectorielles. Par ailleurs, cette façon de procéder a pour effet d'augmenter la variance totale des échantillons sectoriels, nécessitant ainsi que les différences inter-sectorielles soient importantes pour atteindre le niveau de signification statistique. Enfin, une dernière source d'incertitude provient du fait que la majorité des données disponibles sur les caractéristiques biologiques des esturgeons du LDMT ont été recueillies avant 1980. La comparaison de ces données à celles recueillies sur les esturgeons du CFSL suppose que les caractéristiques des esturgeons du LDMT, ainsi que les conditions environnementales prévalant dans ce plan d'eau, soient demeurées stables. Nous ne pouvons nous prononcer sur ce dernier point. Par contre, en ce qui a trait aux caractéristiques biologiques de la population, les résultats quant aux déplacements, obtenus à partir des marquages effectués en 1983 et en 1985, suggèrent un degré de sédentarité très élevé (91% de recaptures dans le même plan d'eau), comparable à ce qui avait été observé par Mongeau et al. (1982) pour la période précédant 1980 (95%) ; en outre, l'embonpoint des poissons en 1985 (K=0.51, n=716) et en 1988 (K=0.50, n=28) est équivalent à celui observé avant 1980 (K=0.51, n=3532). Notons cependant que la taille moyenne des poissons capturés au filet maillant de 200 mm semble avoir augmenté entre 1978 et 1985, poursuivant ainsi la tendance observée au cours des années 1960 et 1970 par Mongeau et al. (1982) et appuyant l'hypothèse de la restauration graduelle de cette population.

Les travaux antérieurs sur les déplacements de l'Esturgeon jaune dans le territoire à l'étude, qui étaient basés principalement sur l'observation des juvéniles de petite taille, ont conclu à la sédentarité relative de l'espèce (Roussow, 1955 ; Magnin et Beaulieu, 1960 ; Leclerc, 1984). Nos résultats varient suivant la nature des sites de marquage. Les poissons étiquetés sur les frayères (rivières des Prairies, l'Assomption et Ouareau), de même que sur les sites de concentration pré- et post fraie (Longueuil), effectuent généralement les déplacements les plus importants ; en effet, des recaptures sont relevées dans l'ensemble du territoire à l'étude, et aussi bien à l'amont qu'à l'aval.

Présentement, nos données ne permettent pas de conclure à la fidélité des géniteurs à une frayère particulière et/ou à une aire d'alimentation déterminée. Un tel comportement a pourtant été démontré dans le cas des Esturgeons jaunes du système lac Winnebago-rivière Wolfe, au Wisconsin (Priegel et Wirth, 1971), qui montent dans la rivière sur une distance d'environ 200 km pour frayer en compagnie d'individus venus de différents points du bassin versant, et reviennent ensuite au lac Winnebago. Un schéma similaire pourrait exister dans le cas des esturgeons du couloir fluvial : les géniteurs quitteraient leurs aires d'alimentation, situées en différents points du territoire de l'étude, pour se rendre frayer sur des sites communs, tels que ceux qui sont situés dans les eaux brunes des rivières des Prairies, des Mille Iles, l'Assomption et Ouareau, à mi-chemin entre le LSTL et le LSTP, et reviendraient ensuite dans leur secteur d'origine. Cette interprétation devra cependant être appuyée par des recaptures additionnelles sur les sites de fraie et d'alimentation.

Au LDMT, le nombre très faible de recaptures, aux dépens de poissons marqués sur les frayères du territoire de l'étude (n=1), couplé à l'absence de recaptures, sur ces mêmes frayères, de poissons marqués au LDMT, tendent à démontrer que les esturgeons de ce lac utilisent des frayères particulières. Des mâles (n=98) prêts à frayer ont d'ailleurs été capturés dans le bief aval du barrage de Carillon, à la tête du LDMT, en juin 1983. Rappelons également que ce sont les esturgeons marqués au LDMT qui présentent le degré le plus élevé de sédentarité. Par ailleurs, aucun poisson marqué sur les sites de pêche commerciale (LSTL, BL, LSTP) n'a été recapturé au LDMT. En conséquence, il semble que le degré de cohésion de ce groupement ait de fortes chances d'être très élevé.

Les esturgeons du LDMT présentent en outre des caractéristiques biologiques très différentes de celles des autres groupements : leur croissance en longueur et en masse est plus lente et, à une taille donnée, leur masse et leur embonpoint sont nettement inférieurs. Signalons de plus que ce sont les esturgeons de ce plan d'eau qui présentent les plus faibles coefficients de variation de la taille et de la masse moyenne à chaque âge, et ceci peut être interprété comme un indice additionnel d'une cohésion supérieure entre les individus de ce groupement.

Compte tenu du fait que cette population était encore en cours de reconstitution au moment de nos captures, la faiblesse de leur taux de croissance et de leur coefficient de condition nous apparaît quelque peu étonnante. On aurait pu normalement s'attendre à ce que la diminution des effectifs dont souffrait la population soit compensée par une croissance accélérée et par un embonpoint supérieur. Mongeau et al. (1982) mentionnent que la croissance relativement faible observée au lac des Deux Montagnes, par rapport à celle qui prévaut dans les secteurs où prédominent les eaux du Saint-Laurent, pourrait constituer un indice de la qualité relativement moindre de ces eaux pour la croissance de l'esturgeon. Cette explication pourrait aussi être valable pour d'autres espèces. En effet, Pépin (1986) a montré que le Doré jaune (*Stizostedion vitreum*) grandissait plus lentement dans le lac des Deux Montagnes que dans le lac Saint-Louis. Du fait que les eaux brunes du lac des Deux Montagnes sont plus acides et moins minéralisées que celles du Saint-Laurent, il se peut que leur productivité globale soit inférieure. En outre, il est également plausible que la cause invoquée pour expliquer la mortalité massive de 1950, soit le manque chronique d'oxygène dont est affectée l'eau de la rivière des Outaouais depuis l'installation des industries papetières le long de ses rives (Mongeau et al., 1982 ; Mongeau et Massé, 1976), continue d'agir, bien qu'à des niveaux sublétaux, en produisant suffisamment de stress pour affecter la croissance et l'embonpoint des poissons.

A l'exception des spécimens du BL, les poissons marqués sur les sites de pêche commerciale et d'alimentation du couloir fluvial affichent un fort degré de sédentarité. De plus, la plupart des recaptures répétées (poissons repris à deux, trois et même quatre reprises) d'esturgeons étiquetés au LSTL et au LSTP furent effectuées dans le plan d'eau d'origine du poisson (Dumont et al., 1987).

Ces observations tendent à démontrer que, à l'exception des migrations de fraie, les déplacements sont plutôt limités et qu'ils sont restreints à un petit nombre d'individus. Ce comportement est donc très propice à l'établissement et au maintien de stocks phénotypiques distincts. Toutefois, il arrive que certains sujets se déplacent sur des distances relativement longues et soient repris à l'extrémité opposée du territoire de l'étude. Par conséquent, en dépit de leur sédentarité, les esturgeons effectuent quand même certains échanges entre les différents groupements du couloir fluvial.

Les différences les plus marquées observées entre les caractéristiques biologiques des trois groupements d'esturgeons du couloir fluvial du Saint-Laurent concernent les tailles, masses et âges moyens des captures du filet maillant, ainsi que le taux annuel de mortalité totale. Les tailles, masses et âges moyens inférieurs, observés au LSTP, quant aux captures du filet maillant de 200 mm de maille étirée et de la ligne dormante, de même les différences observées au niveau de la distribution de fréquence de ces variables, sont peut-être reliées à l'exploitation commerciale plus intense qui se pratique dans ce plan d'eau, et plus particulièrement dans le secteur de l'Archipel du LSTP. Dumont et al (1987) mentionnent que des taux d'exploitation de 15-20% pour l'Archipel du LSTP, et de 10-15% ailleurs dans le couloir fluvial, sont vraisemblables.

Le taux annuel de mortalité totale plus élevé au LSTP pourrait aussi être attribuable à l'exploitation commerciale plus intense. En absence d'estimés du taux d'exploitation, la courbe de capture des esturgeons du FSTL est difficile à interpréter. La similitude du tracé et des valeurs estimées de A, avec ceux des esturgeons du LSTP, semble indiquer que les individus âgés sont plutôt rares dans ce secteur du fleuve, en comparaison avec le LSTL. Payne (1987) a constaté que les tailles, masses et âges moyens des captures expérimentales étaient inférieurs chez les Esturgeons jaunes de la rivière Mattagami (Ontario), surexploités par la pêche commerciale, comparativement à ceux des rivières Abitibi et Frederick House, où l'exploitation était moins intense.

L'exploitation ne semble toutefois pas être le seul facteur susceptible d'influencer les caractéristiques biologiques des groupements. En effet, il appert, de façon inattendue, que les esturgeons du LSTP affichent une croissance en longueur et en masse légèrement plus lente que celle des autres groupements, particulièrement aux âges inférieurs à 16 ans. Payne (1987) rapporte un cas où les esturgeons réagissaient de façon plus classique à l'intensification de l'exploitation commerciale et à la réduction de la densité du peuplement. En effet, ceux de la rivière Mattagami présentaient des taux de croissance plus rapides que ceux des rivières avoisinantes, qui étaient exploités moins intensivement. Les facteurs responsables de ces légères variations de croissance entre les esturgeons des différents secteurs du couloir fluvial restent à déterminer. Signalons enfin que les différences observées au niveau du coefficient de condition de Fulton et de la relation masse-longueur sont plutôt ténues et ne semblent pas devoir constituer un critère suffisant pour permettre l'établissement d'une distinction nette entre les trois groupements du couloir fluvial.

Le statut des esturgeons du BL reste à clarifier, compte tenu de la faiblesse des effectifs disponibles, tant au point de vue de l'expérience de marquage-recapture que de la caractérisation de la récolte commerciale. Les premières indications qui se dégagent de notre étude tendent à montrer que ce plan d'eau constitue un lieu de transit ou une aire de repos au milieu du long secteur de rapides qui s'étend sur une dizaine de kilomètres. Par ailleurs, compte tenu de l'exiguité de l'habitat convenable pour l'esturgeon, il paraît peu probable que le BL puisse héberger une population locale stable d'esturgeons. En ce qui a trait aux esturgeons du FSTL, les résultats présentés ici devront être complétés par une étude des déplacements afin de mieux apprécier l'importance des différences observées entre les esturgeons de ce secteur et de ceux du LSTP et du LSTL.

Soulignons cependant que Magnin et Beaulieu (1960) rapportent les résultats du marquage de 1507 Esturgeons jaunes, la plupart de taille inférieure à 800 mm, dans une section du FSTL située en aval de notre territoire à l'étude. La répartition de 124 recaptures obtenues indique que l'Esturgeon jaune y est plutôt sédentaire et que, dans les cas où des déplacements sont notés, leur amplitude dépasse rarement 48 km et qu'elle est généralement inférieure à 16 km. Ces résultats sont similaires à ceux que nous avons obtenus pour les autres tronçons du CFSL.

Les différences observées entre les groupements sympatriques d'Esturgeon jaune du couloir fluvial du Saint-Laurent, particulièrement au niveau de la taille, de la masse et de l'âge moyens des captures, ainsi qu'au niveau du taux de mortalité totale et de la croissance en longueur, sont du même ordre de grandeur et parfois plus prononcées que celles qui existent entre les populations allopatriques de certains lacs et rivières de l'Abitibi-Témiscamingue (Fort, 1986) et de certaines rivières du nord-est de l'Ontario (Payne, 1987 ; Nowak et Jessop, 1987). Ces différences, couplées aux résultats obtenus quant aux déplacements, justifient, selon nous, l'attribution du statut de stock à chacun des quatre groupements retenus dans la présente étude. En conséquence, il est suggéré que ces différentes entités soient traitées séparément pour la formulation de modalités de gestion et d'aménagement. Compte tenu de la nature des différences relevées entre ces groupements, il ne s'agit évidemment pas ici de stocks génotypiques.

En effet, l'utilisation de frayères communes par des esturgeons du LSTL et du LSTP laisse supposer que ces poissons font partie du même pool génique. Une étude menée actuellement par une des co-auteurs du présent travail (S. Guénette) vise à distinguer les stocks d'esturgeons du LDMT et des différents secteurs du CFSL au

moyen de caractères morphométriques et méristiques et de l'analyse de l'ADN mitochondrial. Il sera intéressant de vérifier la concordance des résultats de cette étude avec ceux du présent travail.

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ANALYSE DES CAPTURES ET CARACTERISTIQUES BIOLOGIQUES DE L'ESTURGEON JAUNE (*Acipenser fulvescens*) DANS LE COULOIR FLUVIAL DU SAINT-LAURENT DE 1985 A 1989

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RESUME

Ce rapport analyse les données biologiques recueillies de 1986 à 1988 à partir des prises commerciales d'Esturgeon jaune (*Acipenser fulvescens*) entre Contrecoeur et Trois-Rivières sur le fleuve Saint-Laurent. Il dresse un bilan des statistiques récentes de pêche. La taille moyenne des esturgeons capturés à la ligne dormante diminue de mai à octobre 1987 au lac Saint-Pierre. L'abondance relative des poissons de moins de 80 centimètres à cet endroit passe de 29,3% à 45,1% de juillet 1986 à octobre 1987. En 1987, les esturgeons les plus abondants (10,5%) sont ceux de la classe d'âge de 1975. Le taux annuel de mortalité totale (A) est de 22,9% en 1987 au lac Saint-Pierre. Les captures atteignent un sommet de 216,7 tonnes en 1987. Elles demeurent supérieures à 129,8 tonnes au cours de la période de 1985 à 1989 dans le secteur entre Montréal et Trois-Rivières. Nous discutons des causes possibles des changements d'abondance des petits esturgeons et de la variation du taux de mortalité.

Mots clés : *Acipenser fulvescens*, biologie, taille, âge, mortalité, exploitation.

ABSTRACT

This report analyses biological data collected from the Lake Sturgeon (*Acipenser fulvescens*) commercial fishery between 1986 and 1988 from the Contrecoeur - Trois-Rivières portion of the St. Lawrence River. A review of recent landings is carried out. The relative abundance of fishes smaller than 80 centimeters increases from 29,3% to 45,1% between July 1986 and October 1987 in Lake Saint-Pierre. The average size of line caught sturgeons decreases from May to October 1987 in this area. The 1975 year class sturgeons is the most abundant (10,5%) in 1987. The annual total mortality rate (A) is 22,9% in 1987 for Lake Saint-Pierre. Commercial catches reach 216,7 tons in 1987 and remain above 129,8 tons between 1985 and 1989 in the Montréal - Trois-Rivières portion of the St. Lawrence River. Potential causes for changes in small sturgeons abundance and in the variations of mortality rate are discussed.

Key-words : *Acipenser fulvescens*, biology, size, age, mortality, exploitation.

INTRODUCTION

L'exploitation de l'Esturgeon jaune (*Acipenser fulvescens*) au Québec est pratiquée à peu près uniquement par les pêcheurs commerciaux. Les pêcheurs sportifs s'intéressent très peu à cette espèce. Sur le fleuve du Saint-Laurent, cette pêche remonte probablement au début de la Nouvelle-France. L'espèce est particulièrement abondante entre Montréal et Trois-Rivières où on trouve l'exploitation commerciale la plus importante en Amérique du Nord (figure 1). En aval de Trois-Rivières et jusqu'à Montmagny à 65 kilomètres en aval de Québec l'Esturgeon jaune partage son territoire avec l'Esturgeon noir (*Acipenser oxyrhynchus*).

La croissance (Cuerrier, 1951 ; Cuerrier et Roussow, 1951), les migrations (Roussow, 1955 ; Magnin et Beaulieu, 1960) et la reproduction (Cuerrier, 1966) ont déjà été étudiées pour l'espèce dans ce secteur. Les travaux récents de Dumont et al. sur les migrations, la différenciation des stocks et le taux de mortalité ont contribué

considérablement à accroître nos connaissances sur ces populations (Dumont et al., 1987).

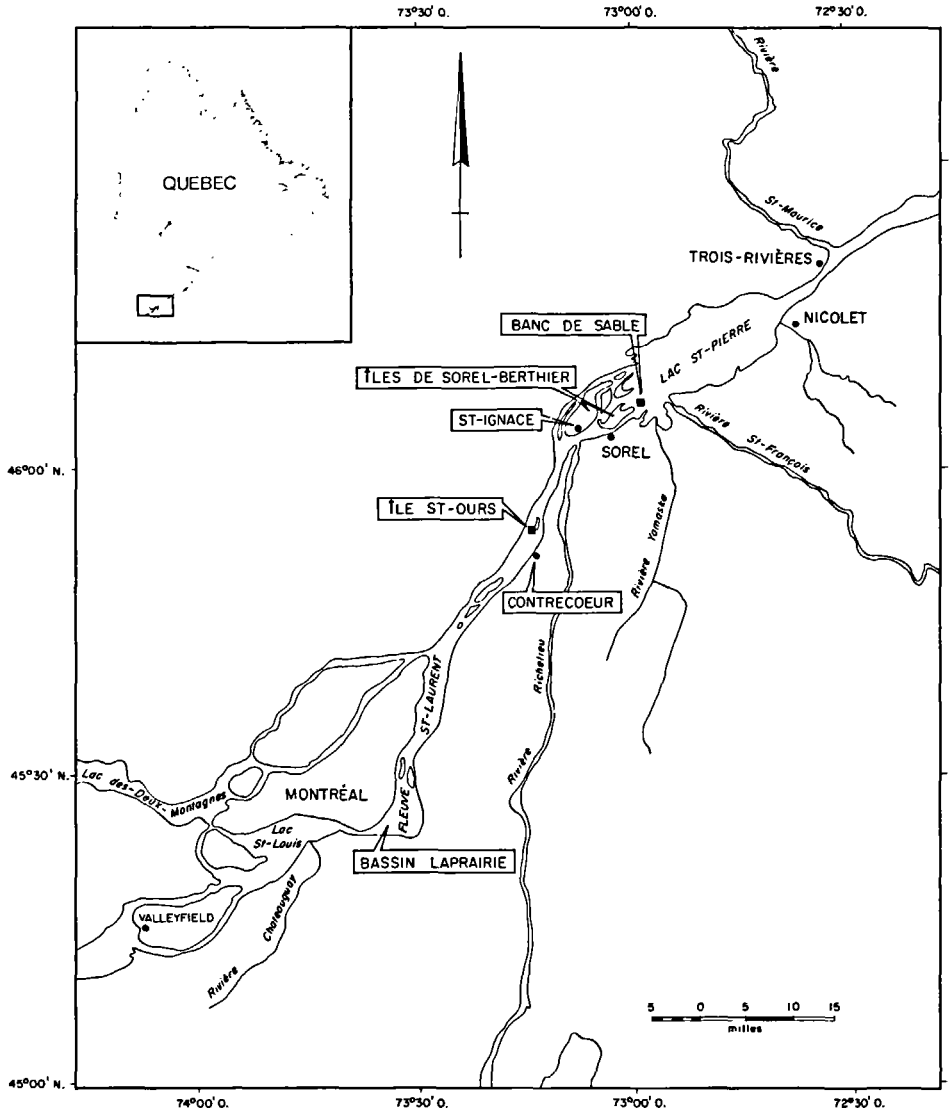


Figure 1 - Localisation du territoire utilisé pour l'étude de l'esturgeon jaune dans le couloir fluvial du Saint-Laurent de 1986 à 1988.

Dans la région de Montréal la pêche débute le 15 juin alors qu'au lac Saint-Pierre et dans les îles de Sorel-Berthier elle commence le 1er juillet. Elle prend fin le 30 octobre à ces endroits. En aval du lac Saint-Pierre, elle n'est interdite que du 1er avril au 14 juin. La ligne dormante est interdite partout au Québec depuis 1988.

La possession d'Esturgeon jaune de taille légale de moins de 45,7 centimètres[1] est interdite. La limite quotidienne pour la pêche sportive dans le couloir fluvial est de 1 poisson.

L'objectif du présent travail est d'utiliser les nouvelles données de taille et d'âge recueillies de 1986 à 1988 à partir de la pêche commerciale afin d'actualiser nos connaissances sur l'état des stocks de la région du lac Saint-Pierre. Nous voulons ainsi vérifier si cette population a subit des changements depuis les derniers travaux en 1984 (Dumont et al., 1987).

MATERIEL ET METHODES

Description de l'échantillonnage

De 1986 à 1988, 1236 esturgeons furent échantillonnés à partir des captures commerciales des filets maillants et des lignes dormantes. Afin d'éviter la perte des petits esturgeons rejetés par les pêcheurs, l'échantillonnage a eu lieu sur le site même de pêche plutôt qu'au débarquement. La longueur totale, la longueur à la fourche et le poids vif ont été recueillis pour chacun des poissons. Le sexe n'a été déterminé que chez les esturgeons débarqués. Il ne fut pas toujours possible d'obtenir un nombre suffisant de poissons, en particulier pour les captures aux filets maillants.

Des données additionnelles provenant des essais de lignes dormantes effectuées en dehors de la saison de pêche (mai-juin 1987) dans la partie ouest du lac Saint-Pierre (Banc de Sable) étant disponibles celles-ci furent ajoutées à l'analyse des captures commerciales (figure 1). Ces essais faits en collaboration avec un pêcheur commercial furent effectués afin de vérifier la sélectivité de différents types d'hameçons. Seuls les captures obtenues avec l'hameçon 6/0 furent utilisées pour la présente étude. Ces données sont comparables à celles de la pêche commerciale, le site, les méthodes et l'engin de pêche étant les mêmes.

Les filets maillants utilisés par les pêcheurs possèdent une maille étirée dont la dimension varie de 19 à 20,3 centimètres. Les lignes dormantes des pêcheurs sont habituellement munies d'hameçons 6/0.

Détermination de l'âge

On a prélevé les nageoires pectorales droites d'un lot de 606 esturgeons capturés par les pêcheurs commerciaux entre Contrecoeur et Nicolet du 3 juillet au 6 octobre 1987. De ces poissons, 399 proviennent de captures à la ligne et 207 proviennent de captures au filet. On y trouve 246 mâles, 266 femelles et 54 poissons dont le sexe n'a pas été déterminé car n'ayant pas été autopsiés. Le sexe n'a pu être déterminé chez les juvéniles.

L'âge est déterminé par le dénombrement des annuli sur l'épine de la nageoire pectorale. La coupe des nageoires pectorales, leur montage sur lame et la lecture subséquente des âges ont été confiés à la firme Gilles Schooner et Associés de Québec. La technique utilisée est décrite par Cuerrier (1951), Probst et Cooper (1954), Roussow (1957), Wilson (1987) et Goyette et al. (1988). Quinze (15) poissons ont du être rejetés à cause de difficultés d'interprétation du nombre d'annuli.

Cueillette des statistiques de pêche

Le MAPA remet des livres de pêche en main propre à chacun des pêcheurs au début de chacune des saisons de pêche. Les statistiques de l'effort, des prises et

[1] La taille légale se mesure du bord postérieur de la fente branchiale à la partie postérieure de l'attache de la nageoire dorsale. Elle correspond à une longueur totale approximative de 80 cm.

des rejets par espèce et par engin sont compilées sur une base quotidienne par les pêcheurs et expédiées mensuellement au MAPA. Après avoir été vérifiées, ces données sont entrées sur fichiers informatiques pour exploitation ultérieure.

RESULTATS

Caractéristiques biologiques

Structure de taille

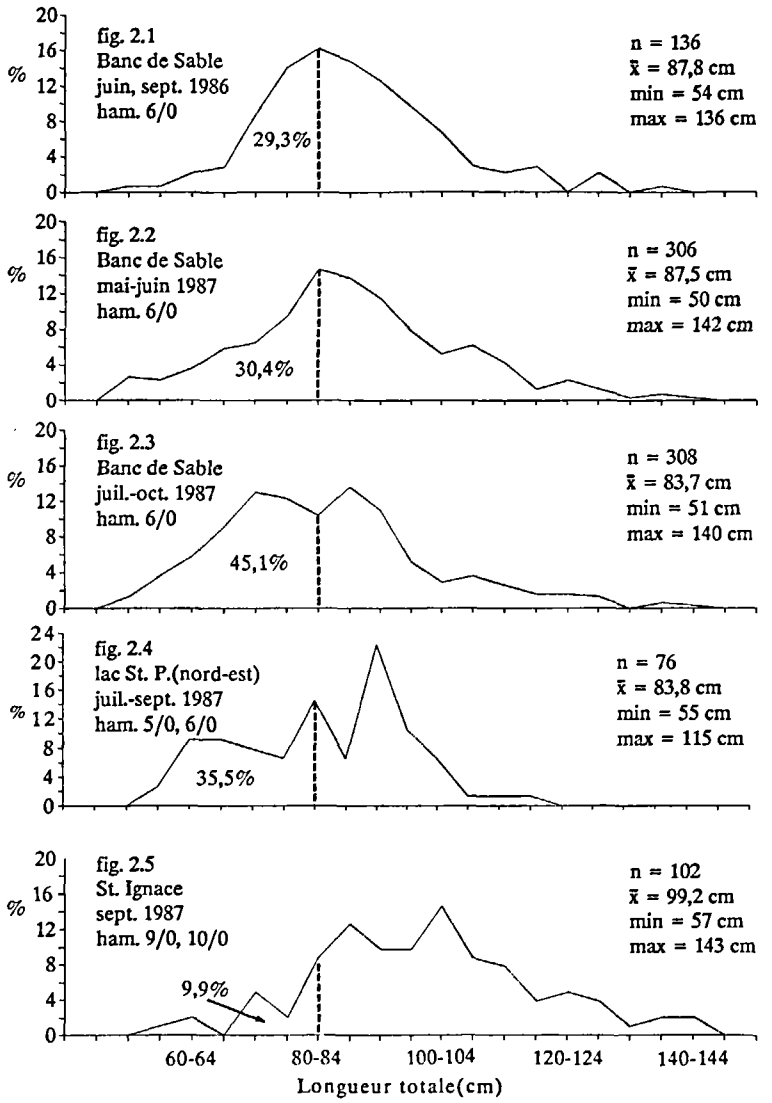


Figure 2 - Distribution de longueur des Esturgeons jaunes échantillonnés à partir des captures commerciales à la ligne dormante dans la région du lac Saint-Pierre en 1986 et 1987.

Nous illustrons les distributions de longueur des esturgeons capturés à la ligne dormante (hameçon 6/0) au Banc de Sable, à l'ouest du lac Saint-Pierre en 1986 et en 1987 (Fig. 2.1, 2.2 et 2.3). Un nombre limité de poissons capturés avec des hameçons 5/0 (90%) et 6/0 (10%) proviennent du nord-est du lac en 1987 (Fig. 2.4). La distribution de longueur des esturgeons capturés avec des hameçons plus gros (9/0 et 10/0) à Saint-Ignace à l'ouest des îles de Sorel-Berthier en 1987 est présentée à la figure 2.5.

L'allure générale de la distribution de taille des poissons capturés à la ligne au Banc de Sable en juillet et septembre 1986 et en mai et juin 1987 est similaire (Fig. 2.1 et 2.2). Leur classe modale se situe à 80-84 centimètres.

Un test de Kolmogorov-Smirnov ne révèle aucune différence significative entre ces 2 distributions (tableau 1). L'abondance relative des esturgeons de moins de 80 centimètres demeure voisine de 30% en 1986 et au printemps 1987 (Fig. 2.1 et 2.2). Cette abondance atteint 45,1% durant la période de juillet à octobre 1987.

La taille moyenne baisse à 83,7 centimètres au cours de cette dernière période. Un test de Kolmogorov-Smirnov révèle des différences significatives ($p < 0,01$) entre la distribution de taille de 1986 et celle de juillet-octobre 1987 (tableau 1).

Il y a également une différence significative entre les 2 distributions de 1987 (Fig. 2.2 et 2.3). La taille moyenne des 76 poissons échantillonnés au nord est du lac Saint-Pierre en 1987 est semblable à celle des poissons capturés à l'ouest de ce lac même si 90% des hameçons sont de calibre légèrement plus faible, soit 5/0.

Nos travaux antérieurs sur la sélectivité des hameçons indiquent qu'une différence aussi faible dans la taille des hameçons n'entraîne pas de différences au niveau de la taille des esturgeons capturés (Dumont et al., 1989).

Tableau 1 - Comparaison à l'aide du test de Kolmogorov-Smirnov des longueurs totales des échantillons d'Esturgeon jaune provenant des captures à la ligne dormante (hameçon 6/0) en 1986 et 1987 au lac Saint-Pierre (Banc de Sable).

1986 et 1987 juil.-sept. mai-juin	1986 et 1987 juil.-sept. juil.-oct.	1987 et 1987 mai-juin juil.-oct.
non significatif	$P < 0,01$	$P < 0,01$

Les poissons capturés à Saint-Ignace à l'aide d'hameçons 9/0 et 10/0 en septembre 1987 affichent une taille moyenne nettement supérieure à celle des poissons du lac Saint-Pierre. Les poissons de moins de 80 centimètres ne représentent plus que 9,9% de l'ensemble des poissons capturés (Fig. 2.5). A notre avis la dimension des hameçons explique en bonne partie cette différence.

Le nombre d'esturgeons échantillonnés à partir des captures au filet maillant est beaucoup moins considéra-ble. Nous illustrons les distributions de longueur des poissons capturés à Contrecoeur (Ile-Saint-Ours) ainsi qu'à proximité des barrages de l'île de Grâce, à l'ouest des îles de Sorel-Berthier en 1986 et 1987 (Fig. 3.1 à 3.4). Cet engin, contrairement à la ligne dormante, retient peu de poissons de moins de 80 centimètres. Etant donné le nombre limité de poissons, les variations de la taille moyenne pour des secteurs et des années différentes peuvent difficilement être interprétées pour les captures aux filets maillants.

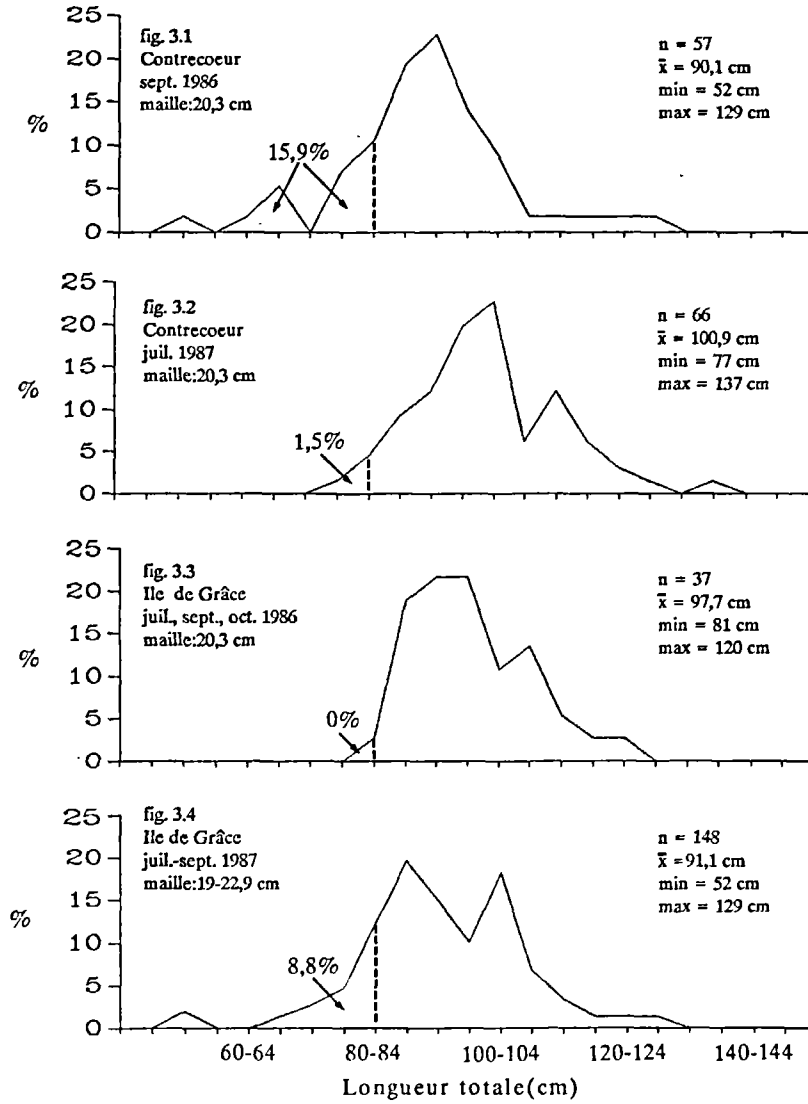


Figure 3 - Distribution de longueur des Esturgeons jaunes échantillonnés à partir des captures commerciales au filet maillant (maille étirée) dans la région de Contrecoeur et de l'île de Grâce en 1986 et 1987.

Abondance selon l'âge et mortalité

Les données de taille des 690 poissons échantillonnés à partir des captures commerciales à la ligne dormante (hameçon 6/0) en 1987 au lac Saint-Pierre ont été transformées à l'aide d'une matrice longueur-âge afin d'obtenir la distribution de fréquence des classes d'âge illustrée à la figure 4. Les données de longueur à l'âge des 591 esturgeons recueillis dans les captures commerciales et expérimentales entre Nicolet et Contrecoeur en 1987 ont servi à construire la matrice indiquée plus haut. La classe d'âge de 1975 (12 ans) est la plus abondante suivie de celles de 1974, 1978, 1976 et 1973 avec des fréquences respectives de 10,5%, 8,4%, 8,0%, 7,5% et 7,3%.

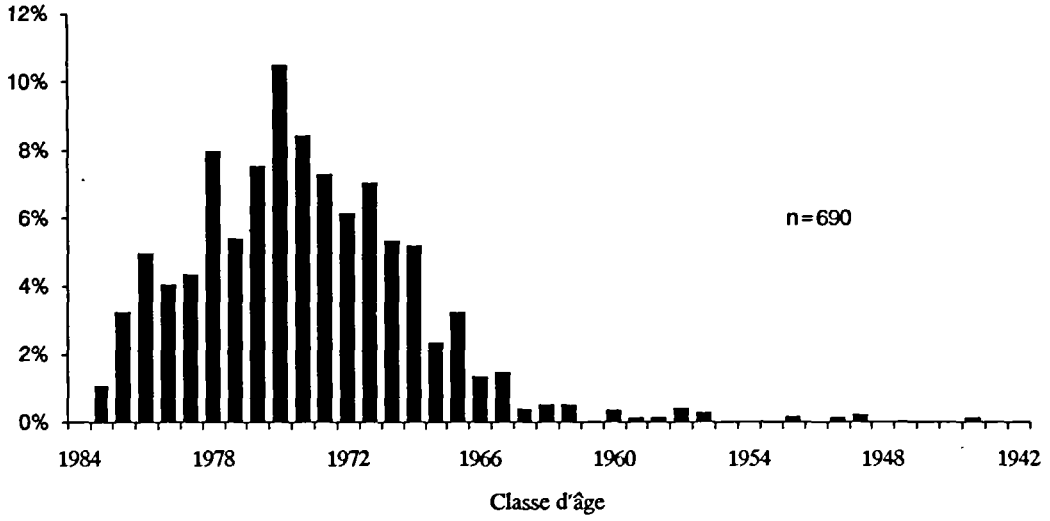


Figure 4 - Abondance (%) par classe d'âge des Esturgeons jaunes capturés à la ligne dormante (hameçon 6/0) au lac Saint-Pierre de mai à octobre 1987.

La courbe de capture calculée à partir de ces données est présentée à la figure 5 (Ricker, 1980). Les classes d'âge de 12 à 25 ans sont utilisées pour calculer le taux annuel de mortalité totale (A). Cette mortalité est de 22,9% en 1987 pour le secteur étudié. Elle correspond à un taux instantané de mortalité (Z) de 0,26 (Ricker, 1980).

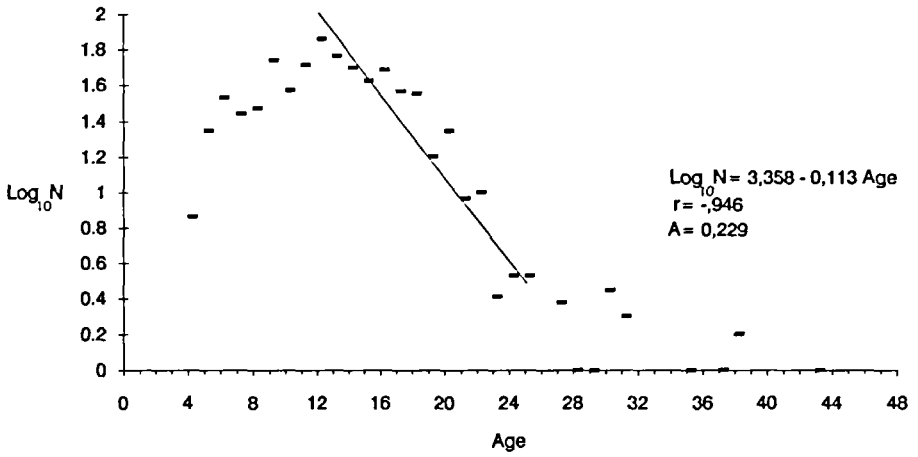


Figure 5 - Courbe de capture de l'Esturgeon jaune établie à partir des prises commerciales (n= 690) à la ligne dormante (hameçon 6/0) au lac Saint-Pierre de mai à octobre 1987.

La pêcherie

Les données recueillies par le Bureau de la Statistique du Québec (BSQ) de 1963 à 1984 pour l'ensemble du Québec indiquent que les captures n'excèdent jamais

57 tonnes durant cette période. Une enquête du ministère du Loisir, de la Chasse et de la Pêche (MLCP) auprès des pêcheurs commerciaux révèle cependant des prises de 102 tonnes pour le seul secteur de Montréal à Trois-Rivières en 1983.

La mise en place par la MAPA en 1985 d'un nouveau système de cueillette des statistiques de pêche permet de corriger en bonne partie cette situation. Ainsi, les prises enregistrées de 1985 à 1989 entre Montréal et Trois-Rivières demeurent supérieures à 129 tonnes atteignant même 216 tonnes en 1987 (tableau 2a). La demande est très forte sur les marchés internationaux au cours de cette dernière année. En 1988 et 1989 nous observons une chute importante des prises au lac Saint-Pierre (54,3 et 57,4 tonnes respectivement) et une diminution également au lac Saint-Louis en 1989. Les pêcheurs d'Esturgeon jaune éprouvent en 1988 et 1989 des difficultés importantes pour acheminer leurs prises sur le marché américain lequel demeure actuellement l'unique débouché pour cette espèce. Cette situation résulte de l'interdiction imposée par l'État de New York au transport d'Esturgeon jaune sur son territoire.

Tableau 2a - Captures commerciales annuelles (tonnes) d'Esturgeon jaune au Québec de 1985 à 1989[1].

Secteur	Nombre de Pecheurs	Année 1985[2]	1986	1987	1988	1989[4]
Couloir fluvial						
Lac Saint-Louis	5	22,8	27,9	47,4	42,1	25,8
Laprairie-Contrecoeur	3	6,7	18,6	13,2	25,5	7,7
Lac Saint-Pierre	42	97,4	93,8	123,3	54,3	57,4
Trois-Rivières	25[3]	5,8	20,9	32,5	30,9	33,5
Québec	12[3]	0,0	0,4	0,3	2,5	5,4
TOTAL		132,8	161,5	216,7	154,9	129,8
Nord-Ouest du Québec						
Abitibi			14,3	19,0	13,8	3,3
Témiscamingue		9,6	7,9	10,3	2,0	2,1
GRAND TOTAL		142,4	183,7	245,9	170,7	135,2

Tableau 2b - Captures commerciale mensuelles (tonnes) d'Esturgeon jaune au lac Saint-Pierre de 1986 à 1988.

Année	Mois								Total
	Avril	Mai	Juin	Juil.	Août	Sept.	Oct.	Nov.	
1986			22,0	26,8	19,5	19,0	6,1	0,3	93,8
1987	0,0	0,0	4,4	48,1	29,2	27,4	14,2		123,3
1988			0,7	23,3	10,2	12,3	7,9		54,3

[1] Statistiques recueillies au moyen des rapports de pêche du M.A.P.A.
[2] Il y a fort probablement sous-estimation des captures en 1985 au cours des premiers mois de la mise en place du nouveau système de cueillette des statistiques de pêche.
[3] Beaucoup de ces pêcheurs ne pêchent qu'occasionnellement ; on considère que les pêcheurs des autres secteurs sont actifs.
[4] Tiré de Johnson, G. (en préparation).

La majeure partie des captures s'effectue en début de saison. Ainsi en 1987 au delà de 66% des captures (81,6 tonnes) s'effectuent avant la fin août au lac Saint-Pierre (tableau 2b). La température plus clémente et une concentration plus importante des esturgeons en début de saison favorise probablement cette situation.

DISCUSSION ET CONCLUSION

Nous présentons au tableau 3 un sommaire des longueurs totales moyennes des esturgeons capturés à la pêche commerciale dans le secteur de Contrecoeur à Nicolet depuis 1945. La taille moyenne des poissons capturés à la ligne dormante dans les îles de Sorel-Berthier en 1984 est de 97,5 centimètres alors qu'elle demeure inférieure à 88 centimètres en 1986 et 1987. Les données de Dumont en 1984 excluent cependant les petits esturgeons rejetés par les pêcheurs avant le débarquement (Dumont et al., 1987).

Tableau 3 - Taille moyenne des Esturgeons jaunes capturés à la ligne dormante et au filet maillant dans la région de Contrecoeur dans les îles de Sorel-Berthier et au lac Saint-Pierre en 1945, 1984, 1986, 1987 et 1988.

Engin	Secteur	Année	Mois	Nombre de Poissons	Lf(cm)
Filet maillant (19-20,3cm)	Contrecoeur	1986[2]	Sept.	57	90,1
		1987[2]	Jl	66	100,9
	Iles de Sorel-Berthier (île de Grâce et autres îles)	1945[1]	J, Jl, A	684	97,6
		1984[1]	Jl, A	1063	89,4
		1986[2]	Jl, S, O	37	97,7
		1987[2]	Jl, A, S	148	91,0
Ligne dormante	Contrecoeur (île Saint-Ours) (hameçon 6/0)	1988[3]	M, J	274	94,5
	Iles de Sorel-Berthier (Banc de Sable) (hameçon 6/0)	1984[1]	Jl, A	165	97,5
		1986[2]	Jl, S	136	87,8
		1987[3]	M, J	306	87,5
		1987[2]	Jl, O	308	83,7
	Nord-est du lac Saint-Pierre (hameçon 5/0 et 6/0)	1987[2]	Jl, A, S	76	83,8

[1] Données tirées de Dumont et al. (1987). Les données de 1945 proviennent de données non-publiées de J.-P. Cuerrier utilisées par Dumont et al. (1987); ces données ont été recueillies au débarquement.
[2] Données du M.A.P.A. obtenues à partir des captures commerciales échantillonnés avant le rejet des petits poissons.
[3] Données tirées de Dumont et al. (1989) et provenant de pêches effectuées dans le cadre des essais de lignes dormantes décrits à la section 2.1 du présent document.

Les données de croissance de Dumont en 1984 indiquent que les poissons de taille égale ou inférieure à 781 millimètres sont âgés d'environ 10 ans ou moins (Dumont

et al., 1987). Ainsi, les poissons de moins de 800 millimètres constituent en 1987 jusqu'à 45% des poissons capturés à la ligne dormante (Fig. 2.1 à 2.3).

Lorsque les petits esturgeons sont très abondants ou lorsque les gros individus sont plus rares, les pêcheurs ont tendance à débarquer davantage de poissons qui se rapprochent de la taille légale. Ce phénomène aurait pu passer inaperçu dans un échantillonnage au débarquement et peut selon nous expliquer en partie les différences importantes des tailles moyennes observées de 1984 à 1986-1987. L'abondance relative plus grande de petits esturgeons en 1987 peut également résulter en partie d'un meilleur recrutement ou/et d'une diminution de l'abondance des gros esturgeons.

Par contre ceci n'explique pas la diminution de la taille moyenne observée à l'automne 1987. Nous ne pouvons à ce stade-ci fournir de véritable explication outre le fait qu'il puisse s'agir de l'arrivée de groupes de petits esturgeons en juillet. Il ne faut pas écarter cependant l'impact possible de l'exploitation.

Le taux annuel de mortalité totale (A) observé par Dumont en 1984 pour les captures au filet maillant au lac Saint-Pierre est de 27,2% pour les poissons âgés de 16 à 27 (Dumont et al., 1987). Nos données des captures à la ligne dormante pour le même secteur en 1987 indiquent un taux annuel de mortalité totale plus faible (22,9%) mais pour les poissons âgés de 12 à 25 ans. L'effort de pêche n'a certainement pas diminué au cours de cette période étant donné la forte demande d'esturgeon sur les marchés jusqu'en 1987 (tableau 2a).

Cette diminution de la mortalité totale de 1984 à 1987 peut selon nous s'expliquer de deux façons: une sélectivité différente des engins ou/et un recrutement plus grand de jeunes esturgeons (classes de 10-14 ans) à la pêche en 1984. Mentionnons d'abord que le filet maillant est beaucoup plus sélectif que la ligne dormante. Si les corrections de sélectivité faites par Dumont pour ces filets n'ont pas réussi à éliminer entièrement la sous-représentation des poissons âgés dans les prises ceci aura pour effet d'accroître le taux calculé de mortalité. Notons également qu'il y a eu arrêt de la pêche en 1970 et 1971 à cause de problèmes de contamination environnementale. L'arrêt presque total de toute forme d'exploitation à ce moment, y compris le braconnage sur les sites importants de fraye de la région de Montréal, peut avoir été très bénéfique pour la fraie et la survie subséquente de jeunes esturgeons qui se retrouvent dans la pêche en 1984. Une accumulation des jeunes esturgeons de la cohorte d'âge de 1970-71 (80-100 centimètres) aurait eu pour effet d'accentuer le taux de mortalité calculé en 1984.

Les données de Dumont et al. (1987) indiquent que nous sommes probablement en présence de sous-stocks distincts dans différents secteurs du couloir fluvial. Un effort de pêche exercé sur les sites d'alimentation semble se répercuter surtout sur la population du secteur exploité et non sur celle de l'ensemble du couloir fluvial. La concentration importante de pêcheurs au lac Saint-Pierre et dans les îles de Sorel-Berthier explique probablement en partie les tailles moyennes beaucoup plus faibles observées à cet endroit en 1986 et 1987 comparativement à ce qu'on observe à l'île Saint-Ours en 1988 (tableau 3).

Le niveau élevé des captures de 1985 à 1987 au lac Saint-Pierre est suivi d'une baisse importante de celles-ci en 1988. Il s'agit probablement d'avantage d'un ajustement de l'effort de pêche à un marché devenu temporaire inaccessible plutôt qu'une baisse réelle des stocks.

Goyette a récemment démontré que l'âge et la longueur moyenne à maturité chez les femelles du lac Saint-Louis sont estimés respectivement à 27 ans et 1330 millimètres. Ces valeurs sont similaires au lac Saint-Pierre (Goyette et al., 1988). Des 690 poissons capturés au lac Saint-Pierre en 1987, 1,7% ont plus de 26 ans comparativement à 1,8% au même endroit en 1984 (Dumont, non publié). Le maintient des stocks d'esturgeons repose donc sur une fraction réduite de la population.

Nous ne prétendons pas que l'échantillonnage effectué en 1986 et 1987 soit sans faille. Le manque de moyens ne nous a pas permis d'effectuer un échantillonnage mieux structuré. La comparaison des tailles des poissons capturés à la ligne en 1984 et en 1986-1987 est difficile car les engins et le mode d'échantillonnage diffèrent souvent. Nous sommes cependant persuadés qu'il est préférable d'échantillonner les captures avant le tri précédant le débarquement et ceci autant pour les captures à la ligne dormante qu'au filet maillant. En effet, les petits esturgeons peuvent parfois être relativement abondants dans les filets (figure 3.1). Ce type d'échantillonnage fournit une indication du recrutement futur à la pêche et constitue ainsi un outil de prédiction pour gérer les stocks.

L'analyse de la situation actuelle des stocks est complexe. Il est probable que plusieurs facteurs agissent simultanément. L'état des stocks peut résulter à la fois de l'effort actuel de pêche (mortalité par la pêche) et des conditions qui ont prévalu antérieurement dans le milieu depuis une trentaine d'années (mortalité naturelle et mortalité par la pêche). Seul un échantillonnage mieux structuré et périodique de la récolte, l'utilisation d'indices d'abondance fiables et des statistiques de pêche de bonne qualité permettront de répondre à moyen terme à ces questions. Une meilleure identification des sous-stocks est également nécessaire afin de comprendre la réaction de la population d'esturgeon aux variations spatiales et temporelles de l'effort de pêche.

Malgré les efforts pour rétablir la qualité de l'habitat sur les frayères il faudra éventuellement à notre avis recourir à la reproduction artificielle et à l'élevage de juvéniles en bassin afin de maintenir les stocks d'Esturgeons jaunes du couloir fluvial à un niveau acceptable.

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COMPARAISON DES PHENOTYPES MORPHOLOGIQUES DES ESTURGEONS JAUNES (*Acipenser fulvescens*) DU COULOIR FLUVIAL DU SAINT-LAURENT ET DU LAC DES DEUX MONTAGNES, DANS LE SUD DU QUEBEC.

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RESUME

Cette étude vise à vérifier le niveau de différenciation morphologique des Esturgeons jaunes (*Acipenser fulvescens*) du lac des Deux-Montagnes et du couloir fluvial du Saint-Laurent (lacs Saint-Pierre et Saint-Louis). Nous avons étudié 41 caractères morphométriques traditionnels et en réseau ("truss network") et deux caractères méristiques. Afin d'éviter l'effet d'allométrie de croissance, les esturgeons mâles et femelles sélectionnés pour cette étude avaient une longueur totale variant entre 890 et 1010 mm. Environ 40 esturgeons ont été récoltés à chaque lieu d'échantillonnage, soit le lac des Deux Montagnes sur la rivière des Outaouais et, de l'amont vers l'aval sur le fleuve Saint-Laurent, le lac Saint-Louis, le secteur des îles de Sorel-Berthier et celui de Nicolet au lac Saint-Pierre, et le secteur de Gentilly.

L'analyse des caractères méristiques révèle une différence moyenne de 1.5 branchicténie entre les esturgeons du lac Saint-Louis et ceux des lacs Saint- Pierre et des Deux Montagnes, malgré des intervalles de distribution égaux. L'analyse en composantes principales et les analyses discriminantes effectuées avec les descripteurs morphométriques ont montré que les esturgeons du lac des Deux Montagnes se distinguent nettement de ceux du couloir fluvial par leur tête, proportionnellement plus haute et plus longue. Les descripteurs les plus significatifs affichent des différences de 4 à 16 mm entre les esturgeons du lac des Deux Montagnes et ceux des autres secteurs du couloir fluvial. Les analyses multivariées ont également révélé l'existence de différences signifi- catives entre les groupes d'esturgeons de Nicolet, des îles de Sorel-Berthier, de Gentilly et du lac Saint-Louis. Les différences sont toutefois très ténues pour chacun des descripteurs considérés un à un; les variations de la forme sont donc très subtiles d'un groupe à l'autre.Nous pouvons ainsi confirmer que les esturgeons du lac des Deux Montagnes forment un stock phénotypique distinct.

D'autre part, nous ne pouvons conclure à l'existence de stocks phénotypiques distincts dans le couloir fluvial sur la base de ces analyses mais nos résultats ne contredisent pas les conclusions tirées par Fortin et coll. (1989) à l'effet que les esturgeons des quatre secteurs du couloir fluvial forment des stocks distincts du point de vue de la gestion.

Mots clés: *Acipenser fulvescens*, phénotypes, stocks, morphologie.

ABSTRACT

The objective of this study was to investigate the degree of morphological differentiation between lake sturgeon (*Acipenser fulvescens*) from Lake des Deux Montagnes and from the Saint-Laurent River fluvial corridor (Lakes Saint- Pierre and Saint-Louis). We studied 41 morphometric characters (traditional and truss network) and two meristic characters. In order to avoid allometric effects, sturgeon of both sexes were selected from a restricted length range (890 to 1010 mm, total length). Approximately 40 sturgeon were obtained from five sampling sites: Lake des Deux Montagnes on the Des Outaouais River and, on the Saint-Laurent River corridor, Lake Saint-Louis, the Sorel-Berthier is- lands and Nicolet sectors of Lake Saint-Pierre, and Gentilly region, downs- tream of Lake Saint-Pierre.

The study of the meristic characters showed a difference of 1.5 branchiospine between Lake Saint-Louis sturgeon and those from Lake Saint-Pierre and Lake des Deux Montagnes; however, the range was identical in all the groups. Principal components and discriminant analyses showed that Lake des Deux Montagnes fish have a proportionally higher and longer head than sturgeon from the other sites. However,the more significant descriptors show a difference ranging from 4 to 16 mm between Lake des Deux Montagnes fish and those from the fluvial corridor. The multivariate analyses also revealed significant morphometric differences between Nicolet, Sorel- Berthier islands, Gentilly and Lake Saint-Louis sturgeon. The differences are very small when each descriptor is considered separately.

The small degree of morphological differentiation between lake sturgeon groups from the fluvial corridor does not justify recognizing them as distinct phenotypic stocks on the basis of our results alone. On the other hand, we cannot contredict the conclusions drawn by Fortin and al. (1989) suggesting that sturgeon from the four sectors of the fluvial corridor should be considered as distinct stocks for the management of the commercial fishery.

key-words: *Acipenser fulvescens*, phenotypes, stocks, morphology.

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DEVELOPMENT OF A SHORTNOSE STURGEON, *Acipenser brevirostrum*, STOCK ENHANCEMENT PROGRAM IN NORTH AMERICA

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RESUME

Les populations de l'esturgeon à nez court, *Acipenser brevirostrum*, ont partout décliné en Amérique du Nord. Cela a entraîné l'élaboration d'un programme de coopération, entre le service fédéral pour le poisson et la faune sauvage et le département de la faune sauvage et des ressources marines de Caroline du Sud. Ce programme prévoit l'étude du cycle biologique, de l'écologie et des possibilités d'augmentation/restauration du stock de cette espèce grâce au repeuplement. Des progrès significatifs ont été réalisés dans toutes les directions. En particulier, la mise au point d'une technique d'élevage permet de tester les possibilités d'augmentation de stock. Les adultes sauvages sont capturés durant leur migration génésique. Les femelles reçoivent une injection d'hypophyse de carpe puis sont massées plusieurs fois quelques heures après le début de l'ovulation. L'éclosion commence après 111 heures d'incubation à 20°C puis les larves sont élevées en intensif dans des bacs jusqu'à l'obtention des juvéniles d'une longueur totale comprise entre 150 et 350 mm. La mortalité est élevée au moment du passage à l'alimentation exogène. Par la suite, elle décroit rapidement et une alimentation artificielle assure une forte croissance aux esturgeons élevés en haute intensité. Durant les quelques dernières années, des poissons d'élevages ont été marqués selon différentes techniques et lâchés en fleuve. Quelques poissons marqués ont été capturés grâce à des campagnes actives d'échantillonnage et une accélération du programme de repeuplement est prévu dès le début 1990.

Mots clés : *Acipenser brevirostrum*, reproduction artificielle, élevage, repeuplement

ABSTRACT

Shortnose sturgeon, *Acipenser brevirostrum*, populations have declined throughout North America. As a result, a cooperative program was initiated between the U.S. Fish and Wildlife Service and the S.C. Wildlife and Marine Resources Department to examine this sturgeon's life history, ecology, and potential for stock enhancement/restoration through use of cultured fish. Substantial progress has been achieved in all program aspects. In particular, a culture technology has been developed which provides the capability to test the potential for stock enhancement. Wild adults are caught during spawning migrations. Females are injected with carp pituitaries and stripped over several hours after the onset of ovulation. Eggs begin to hatch at 111 hours at 20°C and newly hatched fry are reared to advanced juvenile size (150-350 mm TL) in intensive tank systems. At initiation of exogenous feeding, mortality is high. Thereafter, mortality decreases substantially and the sturgeon grow rapidly on artificial diets at high population densities. During the past several years, cultured fish have been tagged using different techniques and released into native waters. Some tagged fish have been captured with intensive sampling efforts and expanded stocking activities scheduled to begin in early 1990.

Key-words : *Acipenser brevirostrum*, artificial breeding, farming, restocking

INTRODUCTION

The shortnose sturgeon, *Acipenser brevirostrum*, was commercially harvested in North America, but today it is listed as an endangered species (Miller, 1972) and considered rare or threatened in Canada (McAllister, 1970). This species inhabits Atlantic coast waters from the St. John River in New Brunswick, Canada to the St. Johns River on the east coast of Florida (Scott and Crossman, 1973). Relative to the sympatric Atlantic sturgeon, *A. oxyrhynchus*, the shortnose sturgeon is relatively small, rarely exceeding a size of 1.4 m in total length (TL) (Gorham and McAllister, 1974).

Age at spawning varies with latitude, ranging from about 6-15 years for females (Dadswell et al., 1984). During recent years, studies have examined the life history and ecology of shortnose sturgeon (Dadswell, 1979; Taubert, 1980; Dadswell et al., 1984). In general, this species inhabits primarily large freshwater rivers which flow to the sea. Adults are not infrequently captured in estuaries as well as in seawater near the river mouths (Vladykov and Greeley, 1963) and appear to spend considerable time in freshwater/seawater interface areas. During late winter/early spring, adults migrate upriver to freshwater sites for spawning. Limited tagging studies suggest that shortnose sturgeon do not exhibit strong migratory behavior.

In 1984, the South Carolina Wildlife and Marine Resources Department (SCWMRD) initiated a cooperative program with the U.S. Fish and Wildlife Service (USFWS) focused on shortnose sturgeon. To date, information has been obtained on certain life history aspects (migratory behavior, size at spawning, adult sex ratios, etc.) and ecological considerations (suspected spawning and nursery areas). Such studies are continuing in order to further define the habitats and requirements of shortnose sturgeon in the southeastern U.S.

Research has also focused on developing culture techniques for use in a program to examine the potential for stock enhancement/restoration of this species (Smith and Dingley, 1984). This manuscript briefly describes the current hatchery and nursery technology, as well as presents information on the status of the stock enhancement activities.

METHODS AND DISCUSSION

Hatchery Procedures

Mature adults are captured in freshwater during their spawning migration in February-March by commercial shad (*Alosa sapidissima*) fishermen fishing 14 cm stretch mesh gill nets on the Savannah River (Smith et al., 1985). Nets are checked daily and the incidentally-captured shortnose sturgeon are placed in floating cylindrical net pens (1.4 m diameter x 1.5 m deep x 6 mm mesh) near the capture sites. These fish are transported to the Orangeburg National Fish Hatchery (USFWS) where they are held in 1.5 to 3.0 m diameter x 0.7 m deep cylindrical tanks which receive flow-through freshwater. Upon arrival, fish are usually treated with a fungicide (malachite green) to prevent the fungal infections typically observed after net capture and transfer to hatchery tanks.

Fish are held in the tanks for up to 5 weeks, during which they are inspected as to stage of maturity. Attempts to feed the tank-held sturgeon have been unsuccessful. Sex discrimination among mature fish is difficult unless the fish are in ripe condition (containing mature eggs or free flowing sperm). The abdomen of males are laterally compressed and checked for extrusion of sperm, or the fish are cathetered. The urogenital area of females becomes reddish and swollen as ovulation time approaches. Once the female appear ripe, ovulation is induced by injection of carp pituitaries at dosages of about 3-8 mg/kg body weight. Time to ovulation varies and ranged from 23 to 47 during 1988-1989 (Table 1). Males receive about 1.5-2.0 mg/kg either to induce or maintain sperm production (Table 1). About 50-100 adult fish are captured annually, however, not all these fish are suitable for spawning purposes. For example, during 1988, 17 females were captured of which only 7 were induced to spawn. In 1989, only 6 females and 15 males of the 96 fish captured could be spawned.

Once several eggs are observed on the tank bottom, the egg stripping process is begun (Smith et al., 1980; Doroshov et al., 1983). Eggs are usually stripped every 1/2 - 1 hour up to a 24 hour period and fertilized with milt from several males if possible (Table 1). The highly adhesive eggs are allowed to adhere to the mesh floor of the egg tray incubators. In 1988-1989, eggs incubated at 20°C began to hatch at a mean time of 111 hours (range 83-131 hours). At hatching, fry are about 9.6 mm TL and weigh 11 mg. Newly hatched fry are transferred to either aluminum troughs (2.4 x 0.5 x 0.2 m deep)

Table 1 - Hatchery data for shortnose sturgeon spawned during 1988-1989.

Spawn		1	2	3	4	5	6	7
Female								
Date of spawning	A[1]	3/8	3/8	3/8	3/8	3/10	3/16	3/31
	B[2]	2/20	2/22	3/02	3/08	3/08	3/08	
TL (cm)	A	99	100	91	106	101	96	88
	B	78	75	66	81	83	89	
Captivity time (days)	A	7	16	2	4	37	3	31
	B	3	5	8	17	2	17	
Pituitary dosage (mg/kg)	A	3,0	b	b	4,4	8,0	6,5	b
	B	2,5	3,0	2,3	c	5,0	4,0	
time of ovulation (hr)	A	26,5	28,5	24,5	24,5	23,5	25,0	23,0
	B	47,0	29,0	31,0	-	27,0	27,0	
duration of egg removal (hr)	A	10	9	24	13	14	23	12
	B	12	13	9	12	12	19	
Male								
captivity time (days)	A	0-35	1-35	1-35	1-35	0-3	2	35
	B	b	2-3	2-3	2-17	2-17	3-17	
pituitary dosage (mg/kg)	A	a	a	1,5	1,5	1,2-1,5	1,9	a
	B	b	2,0	2,0	2,0	2,0	2,0	
No. of males	A	6	4	9	3	4	6	1
	B	b	5	4	5	7	7	
Production x 10^3								
No. of Eggs	A	30,0	40,0	30,0	40,0	35,0	55,0	40,0
	B	10,6	19,5	11,0	30,0	60,0	30,0	
No. of Fry	A	12,4	19,6	12,5	21,1	9,6	45,3	10^{-2}
	B	2,0	3,3	7,3	23,5	54,5	22,5	

A[1] = 1988 B[2] = 1989

a - Males were running ripe, no pituitary injection needed
b - Data not recorded
c - Fish not injected

or cylindrical fiberglass tanks (2 x 1 m deep) which receive flow-through or partially recirculated water.

Nursery Techniques

Depending on temperature, newly hatched fry begin exogenous feeding on about day 8-13. As optimum diet is not known, a variety of feeds are provided which include live *Artemia* nauplii, finely ground beef liver, and soft-moist pelleted salmonid feeds (Smith et al., 1985). Tanks must be regularly cleaned to minimize disease problems as small sturgeon appear highly susceptible to bacterial infections of *Aeromonas* sp., *Pseudomonas* sp. and *Flexibacter columaris*.

Most mortality of young sturgeon occurs during day 13-25 which corresponds to the onset of external feeding. In previous years, survival has been 10%, however, the 1989 nursery techniques were modified which resulted in an average survival of 57% on day 40. Technique changes included the addition of oyster meat (*Crassostrea virginica*) as a supplemental feed, frequent tank cleaning, and improved water quality maintenance. After this initial 40 day period, the small juveniles can be tank-reared at densities of 576/m^2 for 45 days with 98% survival rates. Attempts to rear small sturgeon in ponds have been generally unsuccessful with survival rates of about 4% (Smith et al., 1981, 1985, 1986) and now all small juveniles are reared in tanks. It is believed that small sturgeon are not tolerant of the wide variation in water quality parameters (especially dissolved oxygen) which occur in ponds in late spring in the southern U.S. Recent studies conducted at SCWMRD confirm this observation as it was demonstrated that 71 day-old fish exhibited 84% mortality when exposed to a 3.0 ppm dissolved oxygen level while 73 day-old fish had a 68% mortality at 3.5 ppm dissolved oxygen level (Winn et al., in press).

Nursery studies examining the effect of stocking density have been conducted in tank systems. In 1985, fish were stocked at 8.9 g and at densities to 118 sturgeon/m^2 tank bottom area. After 146 days, fish had grown to a mean size of 148g (333 mm TL) and had a survival rate of 58% at the highest density (Smith et al., 1986). Standing crop was 17.2 kg/m^3 tank volume and feed conversion was 1.5 using a soft-moist ration. During 1988, 85 day-old fish weighing 9.9g (144 mm TL) were stocked at densities from 20.6 to 148 fish/m^2 and reared for 126 days. Fish grew to a mean size of 141 g (336 mm TL) and exhibited an average survival rate of 94.5% (Table 2).

Table 2 - Data for shortnose sturgeon stocked at a mean weight of 9.9 g and reared in indoor tank nursery systems during 1988 at a mean temperature of 23.1°C. Data followed by the same letters were not statistically different ($p \leq .05$)

		Harvest Data					
Density (fish/m^2)	Duration (Days)	WT. (g)	TL. (mm)	Growth rate	Specific[1] Survival (%)	Biomass (kg/m^3)	Feed conversion
20,6	126	153^A	345^A	$2,17^A$	93^A	$4,8^A$	$1,4^A$
49,2	126	144^A	338^A	$2,13^A$	93^A	$10,6^B$	$1,1^B$
98,5	126	144^A	339^A	$2,13^A$	95^A	$22,5^C$	$1,1^B$
148,1	126	123^A	322^A	$2,00^A$	97^A	$29,3^D$	$1,1^B$

[1] S.G.R. % = $\frac{[\text{log final Wt. (g)} - \text{log initial Wt. (g)}] \times 100}{\text{Time (days)}}$

At the highest density of 148.1 fish/m^2, harvest biomass was 29.3 kg/m^3 and feed conversion was 1.1 using a soft-moist ration. At the densities tested, no differences were detected among survival or specific growth rates. Thus, juvenile shortnose sturgeon can be produced at relatively high densities in indoor intensive tank nursery systems.

Marking and Tagging Research

Substantial research has focused on trying to identify a suitable tag for use with shortnose sturgeon in order to evaluate a stock enhancement program (Smith et al., 1988). During 1985-1988, marking and tagging techniques tested included tattooing with India ink, injection of acrylic paints, dart tags, Carlin tags, and abdominal anchor tags. Carlin tags attached to base of the dorsal fin had a long retention time and caused little damage. However, tagged fish were highly susceptible to entanglement in commercial and recreational gill nets. Abdominal anchor tags avoided this problem but tagged fish developed large lesions when placed in brackish water (Smith et al., 1988). Also, tag materials tested were not of long-term durable quality.

More recently, studies have been expanded to include PIT tags (Passive Integrated Transponder), removal of a barbel and binary coded micro-wire tags (Figure 1).

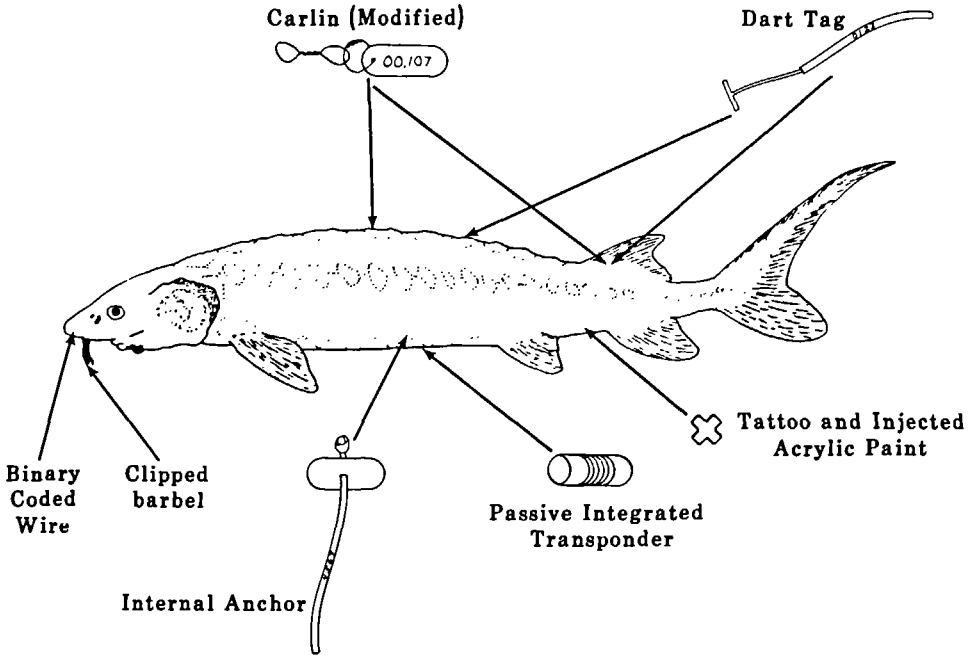


Figure 1 - Schematic diagram showing tagging techniques tested on juvenile shortnose sturgeon.

PIT tags have high retention rates but these internal tags provide no external evidence of origin of the fish. Also, specialized detection equipment is needed and tags are expensive. These tags are being used with all released wild broodstock and in 1989, three previously tagged fish were recaptured after one year at large. During 1988-1989, 651 juveniles (mean 340 mm TL) were tagged using PIT tags and released in the Savannah River as part of the stock enhancement effort (Table 3). Amputation of

barbels has been tested with white sturgeon, *A. transmontanus*, and appears suitable for at least 6 months (Bordner et al., 1988). However, amputated barbels of juvenile shortnose sturgeon begin to regrow within two months. Coded wire tags are being tested with sturgeon as small as 115 mm TL. These tags have been implanted in the snout and after 70 days, retention has been 100% and growth is similar to control fish. Although results are preliminary, this tag appears promising. However, implantation and detection equipment cost is high, and individual batches of fish can only be read by physically removing the tag and reading it with a microscope. Typically, fish need to be sacrificed in order to recover the tag.

Table 3 - Data for the stocking of tagged cultured juvenile shortnose sturgeon in the Savannah River. Total number of fish stocked tagged or not was 77178. Fish less than 100 g were not tagged

Year	Date (Month)	No. of Fish	Mean total length (mm)	Mean weight (g)	Age (days)	Tag type
1986	Jan	478[1]	351,0	180	≈260	Carlin (to scute)
	Oct	156[1]	422,0	215	≈300	Carlin (to base of dorsal fin)
1988	Oct	800[1]	336,5	143,5	217	All barbel clipped with 97 PIT3-tagged
	Oct	800[2]	211,0	-	≈180	800 with barbel clipped
	Nov	405[1]	333,7	130,5	234	All barbel clipped with 373 PIT-tagged
1989	Jan	269[1]	351,1	146,1	307	All barbel clipped with 181 PIT-tagged
Total 1984-1989		2908				

[1] Stocked by S.C. Wildlife and Marine Resources Department
[2] Stocked by U.S. Fish and Wildlife Service
[3] PIT - Passive Integrated Transponder tags

In summary, several tags appear suitable but each has certain limitations as to cost, detection, entangling susceptibility, readability, etc. Research in this area is continuing but for the present time, a combination of an internal tag (eg. PIT) and an external tag (eg. modified abdominal anchor tag) will be used to tag released juveniles.

Current Stock Enhancement Activities

Although a fully suitable mark/tag has not been identified, a program has been established to examine the feasibility of enhancing natural populations of shortnose sturgeon through stocking of cultured juveniles (Smith, 1986). This program is focused on the Savannah River as there is already a substantial data base on shortnose sturgeon in this river and there are several major concurrent sampling and monitoring programs underway which will provide information useful for evaluating the stock enhancement efforts. During 1984-1985, 6,000 small juveniles (50-60 days old) were

released primarily as a by-product of our spawning and hatchery development activities. In 1986, 634 large juveniles (351-422 mm TL, 260-300 days old) were successfully produced in our nursery systems and tagged using Carlin tags and released (Table 3). Although no sampling efforts were conducted by government personnel, a total of 11 fish (1.3%) were reported captured in commercial and recreational gill nets within a ten month period. The earliest captured fish was 21 days after release and all captured fish tended to move downriver towards suspected nursery grounds with one fish travelling 111 km over a 21-day period.

To date, no sampling for these stocked fish has occurred, however, intensive sampling efforts and expanded stocking activities are planned for early 1990.

The efforts underway in South Carolina are designed to examine the potential for success of stocking programs for Atlantic coast sturgeons. Besides the development of suitable hatchery and nursery systems, a successful stocking program requires information on life history, ecology, homing behavior, movements, nursery habitats, etc. For shortnose sturgeon in the southern U.S., such basic data are the focus of current studies and are only beginning to be identified. In addition to the direct stocking efforts, other studies are addressing questions of optimum size for stocking, seasonal movements, migration, identification of natural food habits, etc.

In summary, the stocking efforts are being conducted as a test case in the Savannah River. Within 3-5 years, substantial information should be collected and a preliminary evaluation of the potential for stock enhancement performed. Depending on the outcome of this study, stocking efforts may be expanded to other rivers historically inhabited by sturgeon and to other sturgeon species (especially the Atlantic sturgeon).

CONCLUSION

The spawning and nursery technology is sufficiently developed to allow initiation of a stocking program for shortnose sturgeon. However, there are a number of stocking issues which need to be considered, including size of fish to stock, homing behavior, appropriate marking technique, etc. Further, the shortnose sturgeon is a long lived species which matures at an advanced age. Thus, it will be necessary to have a long term commitment to fully assess the impacts of a stock enhancement program. This will require foresight by state and federal agencies to insure that needed funding will be available to conduct such programs. Through cooperative programs such as that reported herein, it may be possible to enhance/restore populations of shortnose sturgeon, as well as other North American sturgeons.

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STURGEON (*Acipenser sturio* L., 1758) IN SPAIN. THE POPULATION OF THE RIVER GUADALQUIVIR: A CASE HISTORY AND A CLAIM FOR A RESTORATION PROGRAMME

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RESUME

L'esturgeon (*Acipenser sturio* L., 1758) a été signalé en Espagne sur les côtes Cantabriques, Atlantiques ouest et sud-ouest et Mediterranéennes. Ainsi il a été présent dans les rivières Urumea, Miño, Guadiana, Guadalquivir, Júcar, Turia et Ebro. Il remontait le Guadalquivir jusqu'à Cordoue, soit à 230 km de l'embouchure. En 1930, un barrage a été construit à Rio d'Alcala (à 100 km de l'embouchure) interdisant toute migration vers l'amont. Le barrage a également réduit les aires de ponte à une section de 15 km de long située juste au dessus du niveau supérieur des marées. En 1930 un atelier de production de viande et de caviar a été installé en aval et dès lors, la population d'esturgeon a été exploitée. Nous avons examiné les statistiques de capture disponibles depuis 1932. Durant les premières 12 années de pêche (1932-1943), 1832 poissons (1484 femelles) représentant 76 600 kg (69 680 kg de femelles) ont été capturés. Le maximum a été atteint en 1935 avec 385 poissons (342 femelles). Au total, sur 23 ans (1932-1954) c'est 3186 esturgeons qui ont été pêchés dont 2544 femelles. Depuis les années 1940, on observe une réduction progressive et significative du nombre, des taille et poids moyens des poissons. De plus le rapport gonado-somatique décroît également. Cette réduction amène une raréfaction des captures dans les années 1960. Les pêcheurs s'orientèrent sur des espèces plus abondantes et l'atelier de transformation a fermé. Finalement en 1974 et 1975, seulement une femelle et un mâle ont respectivement été capturés. Depuis lors aucun esturgeon n'a été signalé. Une loi de 1983 interdit la pêche en Espagne, et 3 ans plus tard, l'esturgeon était inscrit dans la liste rouge espagnole des espèces menacées.

Nous proposons une possibilité de restauration de la population par la mise en oeuvre d'un programme comportant les points suivants : 1. Meilleure application des lois actuelles sur la pêche, 2. Amélioration de la qualité de l'eau, au moins dans les parties avales, 3. Construction de passe à poisson dans les barrages, 4. Reproduction artificielle et stockage de juveniles en estuaire.

Mots clés : *Acipenser sturio*, statistiques de capture, biométrie, rapport gonadosomatique, barrage, espèce menacée, repeuplement.

ABSTRACT

The sturgeon (*Acipenser sturio* L., 1758) has been reported in Spain from the Cantabrian, north west and south west Atlantic, and Mediterranean coastes. Likewise, it has been recorded from the rivers Urumea, Miño, Guadiana, Guadalquivir, Júcar, Turia and Ebro. Sturgeon migrated by the River Guadalquivir upstream to Córdoba at about 230 km from the river mouth. In 1930 a dam was built in Alcalá del Río (c. 100 km from the mouth) cutting down their upstream migration. This dam also reduced the spawning area to a 15 Km long section situated just above the uppermost level of the tides. Likewise in 1930, a factory for caviar and flesh production was established downstream and thus, the population of sturgeon began to be exploited for these purposes. We have examined the fisheries statistics available for the period 1932 onwards. On the first twelve years of fishery (1932-1943), a total of 1832 specimens (1484 females), accounting for 76600 Kg in weight (69680 Kg of females), were captured. The maximum was reached in the year 1935 with 385 specimens (342 females). As a whole, 3186 sturgeon (2544 females) were caught in 23 years (1932-1954). From the 1940s onwards, a progressive and significant reduction in numbers, mean size and weight of fish is noted. In addition the mean gonadosomatic index decreases as well. This reduction in numbers resulted in scattered captures by the 1960s. Therefore, the fishermen moved to other, most profitable species and the factory closed down. Finally, in 1974 and 1975 only a female and a male were respectively caught. Since then, no more sturgeon has been recorded. Fishing for sturgeon became forbidden by law in Spain in 1983, and three years later sturgeon was considered "endangered" in the Spanish Read List of Threatened Species.

We suggest the possibility of recovering this population and we purpose a restoration programme based on the following: 1. A further application of the actual fishing laws, 2. Improvement of the water quality in, at least, the lower reaches, 3. Construction of fish passes in dams, 4. Induced spawning and stocking of young specimens in the estuary.

Key-words : *Acipenser sturio*, catches, biometry, gonadosomatic index, dam, endangered species, restocking.

INTRODUCTION

Sturgeon (*Acipenser sturio* L., 1758) was never an abundant species in Spain, although it was commonly captured in the rivers Ebro (until the 1950s) and Guadalquivir (until the 1960s). The figure 1 presents the known distribution area in the Iberian peninsula after data reported by Almaça (1988) and ours.

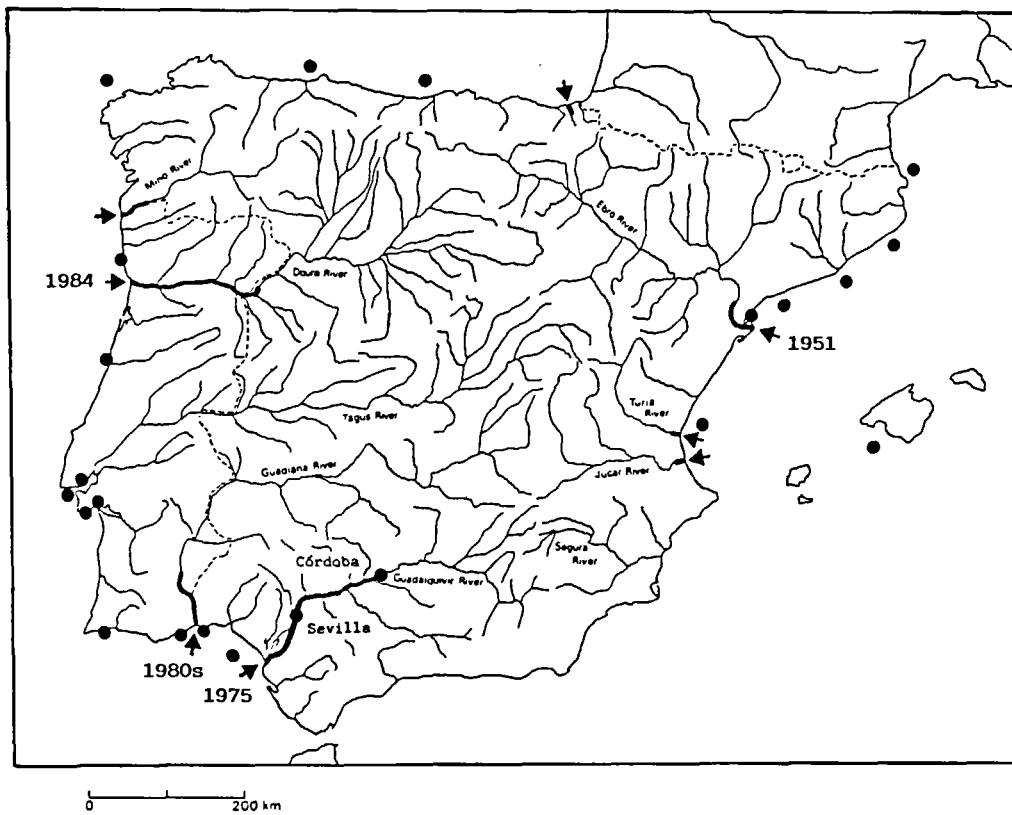


Figure 1 - Recorded distribution of sturgeon in the Iberian peninsula (after Almaça 1988 and our data). Dates refer to the last known fished sturgeon in those rivers.

In the River Guadalquivir, it was a common species, exhibiting upstream migrations far away from the river mouth. Steindachner (1866) reports sturgeons in Córdoba (Fig. 1) situated 230 km far from the river mouth, where they spawned.

Early this century, its abundance and regular fishery was big enough as to promote an interest into the fishery industry that resulted in the stablishment, in 1930, of a caviar and flesh factory in Coria del Río (Fig. 2). In the same year, a dam was built in Alcalá del Río (c. 100 km from the river mouth). A fish pass was constructed, however it was not properly designed and sturgeon was never able to pass away.

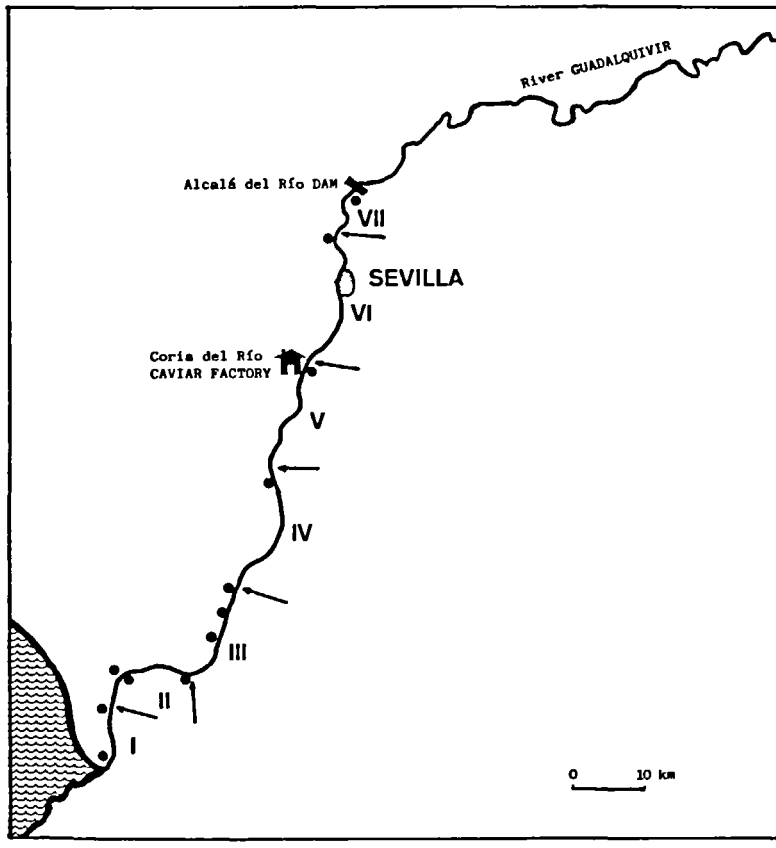


Figure 2 - Map of the river Guadalquivir with the fishing areas (black points) and the seven river sections (I-VII) used in the analysis of catches.

METHODS

The most and best data actually available on Guadalquivir sturgeon come from the studies by Classen (1936, 1944). Fisheries reports and statistics are found in Classen (1944), Rada (1954) and Muñoz-Goyanes (1959), however, these data are scattered and mainly aimed to female sturgeon, while data on male are scarce or none. Further information on reproductive and growth tactics of this population are given in Classen (1936, 1943, 1944), Lozano-Rey (1936) and Gutiérrez (1962).

The fishing method mostly used by fishermen was the "palangre", a succession of lines and hooks without bait, laid down in the river bottom. The fishing areas where the "palangres" were settled are shown in Fig. 2. In order to analyse the sturgeon catches, we have divided the river into seven sections (I-VII) from down to upstream (Fig. 2).

A two sample t test was used for comparison of samples. Simple linear regression equations were calculated by the method of the least squares. Fitness of regression was tested by an analysis of variance procedure (Zar, 1984).
Gonadosomatic index (GSI) is defined as follows:

$$GSI\ \% = (\text{gonad weight/total fish weight})\ 100$$

RESULTS

The total number of sturgeon captured in the period 1932-1954 was 3186 (2544 females). The annual variation in the numbers caught in this period is shown in the Fig. 3.

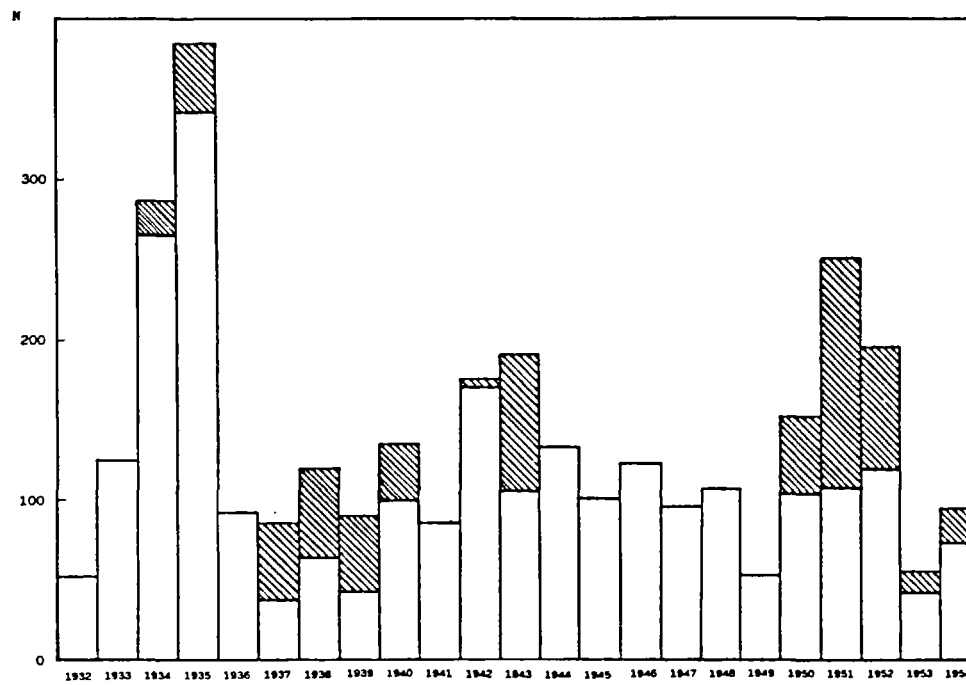


Figure 3 - Annual variation in the numbers of catches (period 1932-1954) (white: females, striped: males).

In the early 1930s, fishermen got rather important amounts of sturgeon. An early increase in the numbers is evidenced with a maximum value in 1935, when 385 sturgeons (342 females) were captured (Fig. 3). From this year the annual catches decrease and later remain rather constant for the females (about 100 specimens per year).

The sex-ratio of the captured sturgeon (period 1932-1943) according to these river sections is shown in Fig. 4. Females contribute with a significant proportion to the total catches. It is a result of the selectivity of the fishing methods. Besides, 63.2 % of all the fish were captured in the lower sections (I and II), that is the last 25 km of the river course.

In 1932, most of the fish were caught in the uppest reaches (Fig. 5), where sturgeons accumulated since their migration was stopped by the dam. In Fig. 5, it is also put forward that as the time passed the maximum percentage of annual catches moved towards downstream. A trend to stabilization is noted by the mid-sections and finally, the higher catches remain stabilized in the lowest sections

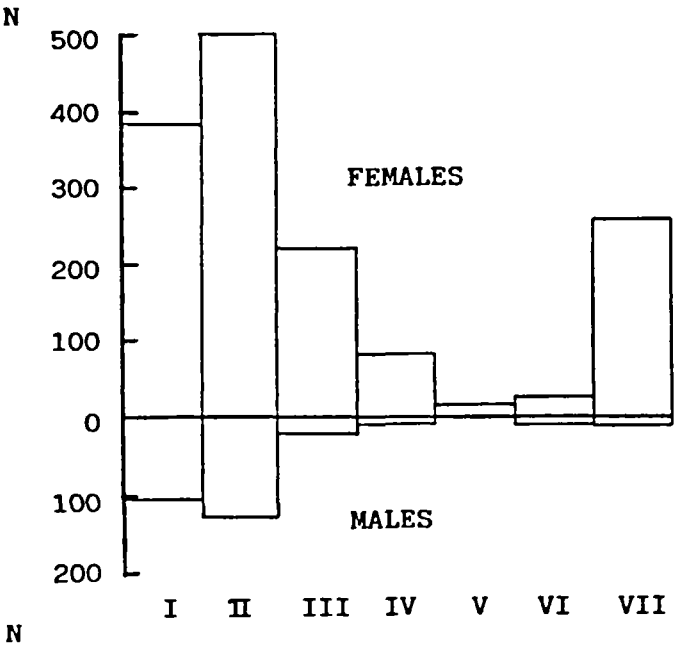


Figure 4 - Sex-ratio of catches (period 1932-1943).

This is very peculiar for a caviar industry, since as the female sturgeons come into the river, and reach these lowest sections they have not finished the development of their gonads (Classen 1944, Gutiérrez 1962). In Fig. 6 are shown the catches of sturgeon in terms of weight. These data couple with the numbers, with a maximum value in 1935 (17320 kg, 16500 kg for females) and reduce to about 4000 kg of females in the 1950s. Perhaps the clearest event in the decline of sturgeon fisheries is shown by the reduction of the gonads production (Fig. 7). In the early 1930s as the number and weight of captured females were the largest, the fisheries rendered 2300 kg in 1934 and 3260 kg in 1935 of female gonads, getting down to about 500 kg in the 1950s (Fig. 7)

The decline in the numbers along this time is further associated with a reduction of the mean size (Fig. 8 and 9). A comparison (t test) between the length of female sturgeons captured in the years 1932-1934 versus those captured in 1941-1943 shows significant differences ($p \leq 0.05$). These results are further supported by a comparison between the mean weights ($p \leq 0.05$). Very surprisingly, the size of females decreased in a really short period of time. A similar trend continues along the 1950s. Nevertheless, the available data for these years is limited to mean and extreme values, and no comparison with the former data is possible. However, a significant correlation ($p \leq 0.05$) and a regression between the mean lengths and mean weights versus the time (from 1932 to 1954) have been also found:

$$\text{length(cm)} = 1176.18 - 0.509\ \text{time(year)}$$

$$\text{weight(kg)} = 735.633 - 0.355\ \text{time(year)}$$

These facts, reduction in length and weight in a short time, can be sign of overfishing, which would be one of the causes for the decline of sturgeon populations in the River Guadalquivir.

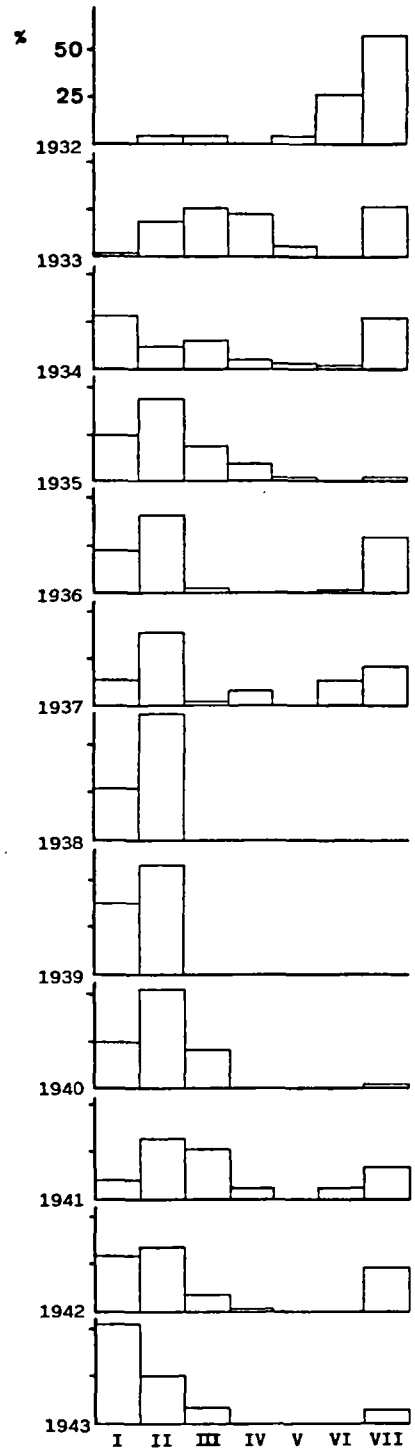


Figure 5 - Annual percentage of catches (period 1932-1943) along seven river sections.

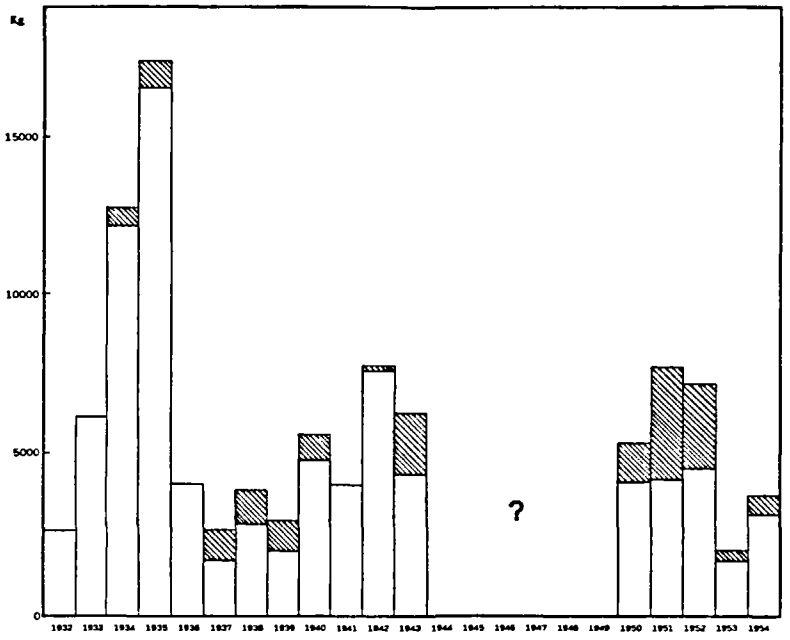


Figure 6 - Annual variation in weights of catches (period 1932-1954, no available data from 1944 to 1949) (white: females, striped: males)

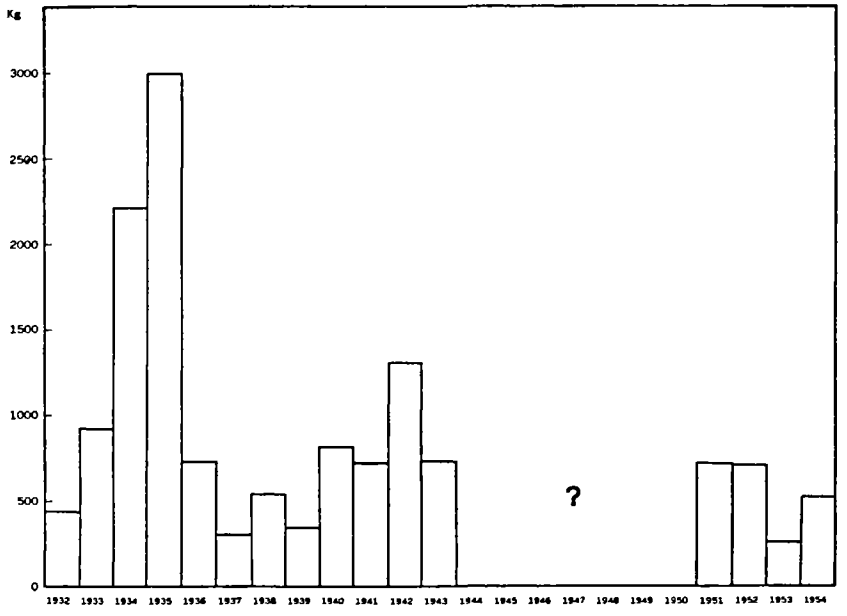


Figure 7 - Annual variation of total weight of female gonads (period 1932-1954, no available data from 1944 to 1950).

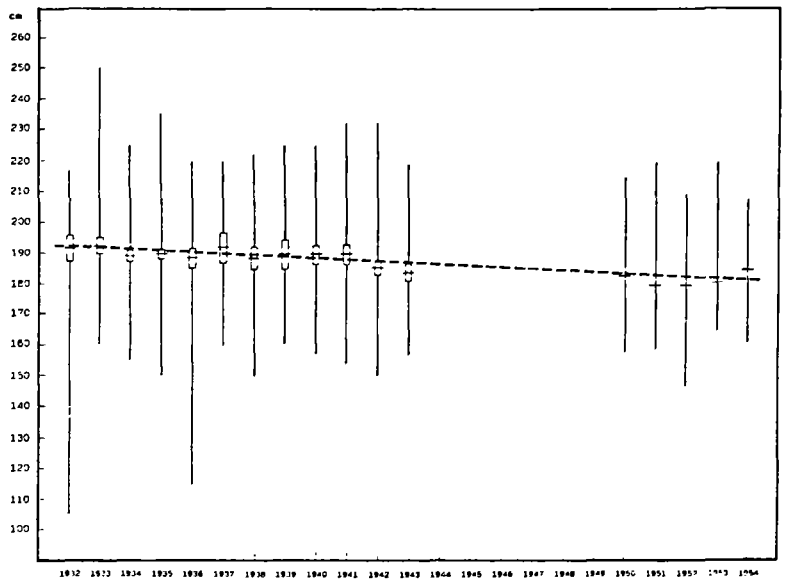


Figure 8 - Annual variation in length of females (period 1932-1954, no available data from 1944 to 1949) expressed by interval of variation, mean and 95% confident limits. Broken line represents the regression line calculated for annual mean values(see text).

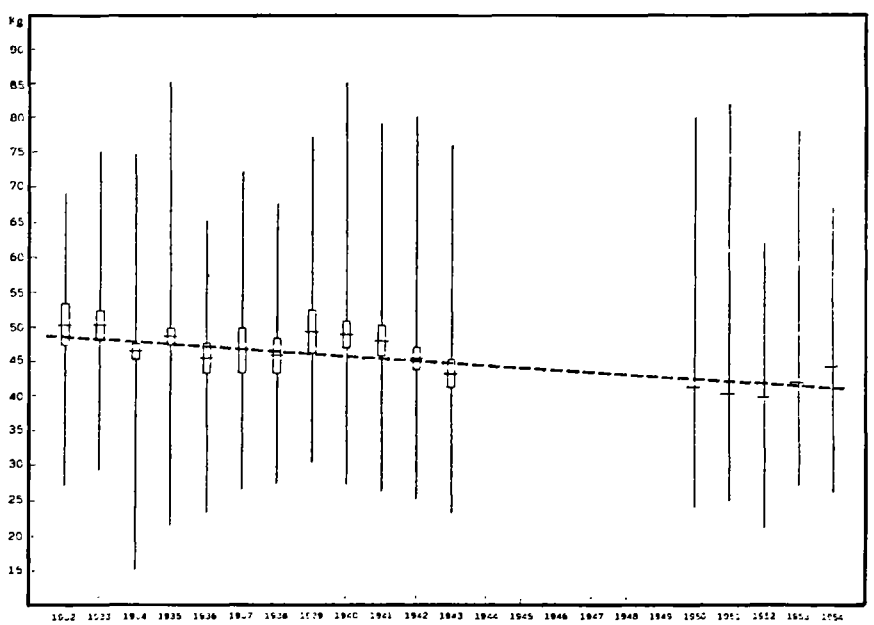


Figure 9 - Annual variation in weight of females (period 1932-1954, no available data from 1944 to 1949) expressed by interval of variation, mean and 95% confident limits. Broken line represents the regression line calculated for annual mean values (see text).

A similar decline is valid for the gonadosomatic index (GSI) (Fig. 10). For this parameter and for the same periods of time (1932-1934 and 1941-1943) a significant difference is also noted ($p \leq 0.05$).

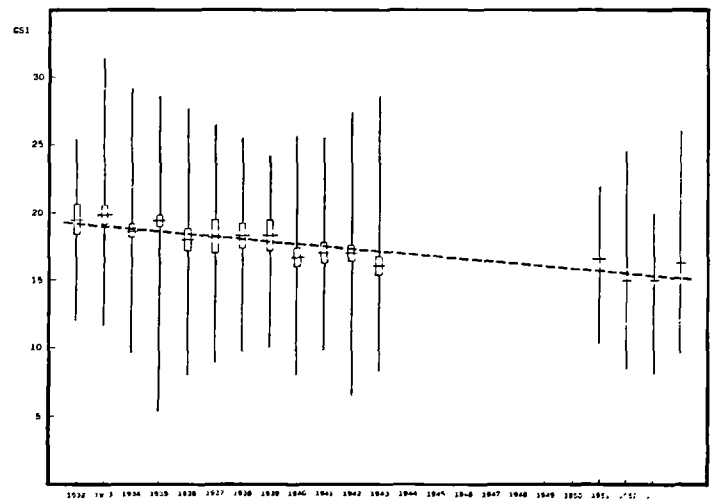


Figure 10 - Annual variation in gonadosomatic index of females (period 1932-1954, no available data from 1944 to 1950) expressed by interval of variation, mean and 95% confident limits. Broken line represents the regression line calculated for annual mean values (see text).

Likewise, a significant correlation ($p \leq 0.05$) between the mean values and years (1932-1954) was also found:

$$GSI = 362.192 - 0.177 \text{ time(year)}$$

The decrease of the gonadosomatic index along the time is due to both, the reduction in females size and to the fact of fisheries were changing from upstream to the river mouth. Here the females were captured just when they came into the river and the gonadal development was not finished yet (Classen 1944, Gutiérrez 1962).

Finally, during the 1960s the captures were more and more scattered (Gutiérrez 1962), the fish for sturgeon became of no interest and the factory closed down. Later, in the 1970s only two fishes are known to be captured: a female in 1974 and a male in 1975 (Hernando 1975).

DISCUSSION

The human activities possibly influencing the decline of sturgeon in the River Guadalquivir can be summarized as follows: 1. The construction of a dam in Alcalá del Río, 2. The overfishing, 3. The loss of spawning areas due to the sand and gravel extractions, 4. Reduction in water quality due to pollution from industries and city wastes.

Several authors (Classen 1936, 1943, 1947, Vélaz de Medrano 1940, Vélez-Soto 1951, Lozano 1956) have claimed for the decline of this population. Everybody agree in blaming the Alcalá del Río dam as the only responsible, since reproduction upstream the dam was no possible after its construction (Gutiérrez 1962).

Although they also consider a damage the capture of young specimens in the estuary, nobody mention the overfishing of reproductive females. Only Gutiérrez (1962) reported the negative effects of the pollution, the sand extraction and fisheries. Nevertheless, incidence of these factors was never tested.

CONCLUSION

After the revision of known data we conclude that the initial and main reasons conducing to the decline of the sturgeon population were the construction of the dam and, in part, the overfishing. At the beginning of the 1930s the sturgeons were quite abundant, as shown by the initial fisheries numbers. When the presence of the dam stopped the migration of reproductive sturgeons, they accumulated downstream and they were captured in a high number. After all, some of them were able to find new spawning grounds as prove the captures made some years later (Gutiérrez 1962). Perhaps, after the construction of the dam the sturgeons would have been able to survive, but the intensive fishery they supported had to be a negative effect for their survival. The sand extractions near the spawning sites and the water pollution could help to the sturgeon decline, but anyway in a less proportion than the two former facts.

Since we infer the possible reasons for the decline, we can propose the solutions. Thus, the restoration programme would be as follows:

1. A further application of the actual fishing laws.

2. Control and improvement of the water quality. The present level of water quality can be insufficient, but appropriate efforts are currently doing (Toja 1985). The future is hopeful since a programme for water depuration of the river is now in progress and will finish on the early 1990s.

3. Construction of fish passes in dams. It is technically possible to carry on in the Alcalá del Río dam. The project of a fish pass available for sturgeon was done in 1940 (Vélaz de Medrano 1940) but never enabled. Construction of fish passes in upstream dams would be consider in a second step.

4. Inducing spawning and stocking of young specimens in the estuary. Adult specimens could be obtained from the same river if possible, from the neighbouring River Guadiana, or perhaps from other European fish farm.

Several Spanish institutions can contribute to this programme with support or funds: the National Institute for Nature Conservancy (ICONA), the Environment Agency of the Autonomic Region of Andalucía, other local or regional authorities, etc. In any case, the programme would have to coordinate the efforts of different technical and scientific groups. Nevertheless, an international cooperation would be of a great interest.

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ELEVAGE

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INDUCED OOCYTE MATURATION, OVULATION AND SPERMIATION IN STURGEONS (ACIPENSERIDAE) USING SYNTHETIC ANALOGUE OF GONADOTROPIN-RELEASING HORMONE

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RESUME

La possibilité d'utiliser l'analogue synthétique du GnRH des mammifères, le GnRHa (Gonadotropin-Releasing Hormone Ethylamide - des Gly^{10}, $D\text{-}Ala^{6}$) pour provoquer l'ovulation et la spermiation chez les esturgeons a été étudiée entre 1983 et 1988. Des doses aussi faibles que 0.15 - 0.6 µg/kg poids vif sont capables d'induire l'ovulation chez l'esturgeon étoilé (*A. stellatus*), l'esturgeon russe (*A. gueldenstaedti*), le beluga (*H. huso*) et le sterlet (*A. ruthenus*). Les pourcentages de femelles et de mâles des trois premières espèces donnant des produits sexuels de bonne qualité sont équivalents qu'ils soient traités avec du GnRHa ou un extrait hypophysaire d'esturgeon (EHE) Par contre chez le sterlet, une seule injection de GnRHa entraîne des ovulations et des spermiations médiocres, ces résultats peuvent être améliorés avec deux injections. La qualité des oeufs mûrs était la même quelque soit la préparation hormonale appliquée (GnRHa ou EHE). Le délai entre l'injection et l'ovulation est identique chez le beluga quelque soit le traitement hormonal, alors que des différences significatives sont observées chez l'esturgeon étoilé et l'esturgeon russe. Les résultats obtenus montrent que le remplacement des hypophyses par le GnRHa est très prometteur pour l'obtention des gamètes chez les esturgeons.

Mots clés : *Acipenseridae*, GnRH, ovulation, spermiation

ABSTRACT

The application of synthetic analogue of mammalian gonadotropin-releasing hormone (GnRHa), des Gly^{10} [$D\text{-}Ala^{6}$] - GnRH ethylamide, for inducing ovulation and spermiation in sturgeon fish was investigated in 1983-1988. The dosages of GnRHa as low as 0.15 - 0.6 µg/kg body weight were shown to be capable of inducing ovulation in the starred sturgeon (*A. stellatus*), in the Russian sturgeon (*A. gueldenstaedti*), in the beluga (*H. huso*) and in the sterlet (*A. ruthenus*). The percentage of females and males of the first three species, that produced mature gametes of high quality was practically the same upon administration of GnRHa or sturgeon pituitary extract (SPE), while the sterlet exposed to a single low dosage of GnRHa showed a worse ovulation and spermiation responses compared with the ones following SPE injection. These responses could be increased by administering two spaced dosages of GnRHa. The developmental potential of mature eggs was shown to be equal, whatever the hormonal preparation (GnRHa or SPE) used for induction of their maturation. A comparison of the time of ovulation, induced by GnRHa or SPE revealed some significant differences in the cases of starred sturgeon and Russian sturgeon, but not in the case of beluga. The results demonstrate that replacement of conventional gamete maturation inducing technique by a strategy of GnRHa application on sturgeon fish farms holds much promise.

Key-words : *Acipenseridae*, GnRH, ovulation, spermiation

INTRODUCTION

Propagation of sturgeons at the USSR state fish farms is based on an induction of ovulation and spermiation by administering sturgeon pituitary extract (SPE). This method has long been developed (Gerbilsky, 1941) and remains parctically unchanged to date. The disadvantages of the use of pituitary preparations are as follows : hand-made extirpation of pituitaries and their separation from surrounding tissue, partial loss of commercial value of fish after pituitary extirpation, problems in standardization of

pituitary gonadotropic activity, and, finally, the high cost of SPE injection. The earlier attempts to replace SPE by progesterone (Goncharov, 1969) or human chorionic gonadotropin (Barannikova et al., 1975) have not been successful. A similar problem of replacing piscine pituitary by a low-cost hormonal preparation remains topical for many other artificially bred fish.

Recent studies have shown that mammalian gonadotropin-releasing hormone (GnRH) and its superactive analogues (GnRHa) are capable of inducing ovulation and spermiation in fish, but their efficacy varies widely between the species (see Lam, 1982 ; Donaldson, Hunter, 1983 ; Sherwood, 1987).

To check the GnRHa capacity of inducing ovulation and spermiation in sturgeons, experiments were conducted in 1979-1982 with two species : the starred sturgeon (*Acipenser stellatus Pallas*) and the Russian sturgeon (*Acipenser gueldenstaedti Brandt*). The lowest of the used dosage of GnRHa (1 µg/kg body weight) administered in a single intramuscular injection, proved to be effective (Goncharov, 1984). However, more studies were required to develop a new standard method of inducing ovulation and spermiation in sturgeons.

In this report we examine :

1) the lowest dosage of GnRHa effective in inducing ovulation in the starred sturgeon ;

2) the quality of mature eggs and sperm, obtained from GnRHa and SPE-treated sturgeons in comparative experiments, carried out on different fish farms during several years ;

3) the effectiveness of GnRHa in the induction of ovulation and spermiation in several sturgeon species ;

4) the time of ovulation as induced by GnRHa and SPE.

MATERIAL AND METHODS

Experiments were performed in 1983-1988 on sturgeon fish farms on the Volga and Kuban riverside. The fish were captured during their anadromous migration or near the spawning sites and maintained in outdoor artificial reservoirs before and during the experiments. The holding facilities were equipped with running river water.

GnRH analogue, des Gly^{10} [D-Ala^{6}]-GnRH ethylamide was purchased from the USSR Cardiology Research Centre, Acad. Med. Sci. USSR (Moscow) as lyophilized compound, and from "Biolar" Inc. (Olaine) in saline (0.9 % NaCl) solution form - 20 µg/ml. The lyophilized preparation was dissolved in Ringer solution just prior to use. GnRHa and SPE were administered via intramuscular injection. The animals for one or another treatment were chosen randomly. All routine manipulations with fish and mature gametes were undertaken as earlier described (Charlon et Williot, 1978 ; Detlaff et al., 1981). Temperature was recorded during induction experiments, normally four times a day. The females were weighed before the injection of hormonal preparation or before sacrifice.

Egg quality was estimated as a percentage of the fertilization rate and/or as a percentage of normal embryos at one or two developmental stages up to hatching. The samples were taken from the fish farm incubators. Each sample comprised 200-300 embryos.

Milt volume was only scored in individual males during the first collection. Sperm quality was estimated using a subjective five-class scale (Persov, 1941 ; see also Ginzburg et al., 1989). Only high quality sperm (4-5 points) was used for egg insemination.

The chi-quare test was used to evaluate the differences among qualitative responses to GnRHa and SPE treatments. The median values of different quantitative characteristics, obtained in experiments were tested by the Student criterion (Urbach, 1964).

Minimal effective GnRHa dosage for inducing ovulation in the starred sturgeon.

The effective dosage of des Gly^{10} [D-Ala^6]-GnRH ethylamide for inducing ovulation in the starred sturgeon being first unknown, in our initial experiments we used those dosages that were shown to be effective for des Gly^{10} [D-Leu^6]-GnRH ethylamide (Goncharov, 1984). Administration of a single injection of GnRHa in doses of 40 and 10 µg/fish produced an ovulatory response in most of the treated females (Table 1). Thereafter, each consecutive group of ten females was injected with a two-times lower dosage of GnRHa until a significant difference ($P<0.05$) in the response was obtained. The results presented in Table 1 show that the minimal effective dosage of GnRHa was 1.25 µg/fish. Due to variability of female body weight, the dosages per weight unit varied within each group. Among the females treated with a dosage of 2.5 µg/fish, response was recorded in those which received 0.16, 0.20, 0.23 and 0.24 (3 females) µg/kg. Another four females receiving 0.19, 0.22 , 0.26 and 0.28 µg/kg of GnRHa did not ovulate. Thus, no relationship between the positive response and the dosage of GnRHa calculated per kilogram body weight was recorded in this group. In the next group (1.25 µg/fish) only a single female, treated with 0.15 µg/kg of GnRH, ovulated. By contrast, the other females, treated with 0.15, 0.14 and less than 0.1 µg/kg (9 females in this and 8 females in the following low dosage groups), displayed no ovulatory response. It follows that the minimal effective dosage of des Gly^{10} [D-Ala^6]-GnRH ethylamide inducing starred sturgeon females to ovulate is about 0.15 - 0.20 µg/kg body weight.

Table 1 - Effect of different doses[1] of gonadotropin-releasing hormone analogue (GnRHa) on ovulation in the starred sturgeon (*A. stellatus*)

Dose(µg)		No. of	No. of fish
per fish	per kg	fish	ovulated
40	3-5	10	5
10	0,7-1,3	10	8
5	0,4-0,7	10	6
2,5	0,16-0,28	10	6
1,25	0,10-0,15	10	1
0,63	0,04-0,07	5	0
0,31	0,02-0,03	3	0

[1] *The absence of saline control in our experiments is due to the fact that the long experience of sturgeon* fish farming indicates that sturgeon females do not ovulate spontaneously in captivity. In addition the 17 females which did not ovulate in response to very low dosages of GnRHa can considered to be a saline control.

Comparison of the effects of GnRHa and SPE on ovulation and egg quality in Acipenserid fish

Acipenser stellatus. Table 2 summerizes the results of administering of GnRHa and SPE to starred sturgeons. In this table, data are included that were obtained in 1981-1982, using des Gly^{10} [D-Leu^6]-GnRH ethylamide (Goncharov, 1984) to pool all our results of ovulation induction with GnRHa's.
It can be seen that the effectiveness of GnRHa's application varies between years. This is made particularly clear by comparison of the percentage of treated females from which the high quality eggs were obtained. By contrast, comparison of the effectiveness of GnRHa and SPE during each experimental season reveals no significant differences. This implies that the physiological state of females in a local population at

Table 2 - Effects of sturgeon pituitary extract (SPE) and gonadotropin-releasing hormone analogue (GnRHa) on ovulation and egg quality in the starred sturgeon (*Acipenser stellatus*).

Experiment	Treatment	Dose	No. of fish	Fish ovulated		Fish ovulated (high quality eggs)		Fertilization rate, %(M ± SE)	Normal embryos according to the stage, % (M ± SE)	
				No.	%	No.	%		17*	35*
1981 Ural river	SPE	25 mg/fish	7	7	100	7	100	82.4 ± 13.3	79.2 ± 6.4	66.0 ± 25.8
	GnRHa	4-73 µg/kg	11	11	100	11	100	85.2 ± 6.0	81.1 ± 6.1	74.0 ± 11.0
1982 Volga river	SPE	25-35 mg/fish	51	46	90	38	75	82.8 ± 12.8	76.9 ± 15.5	71.3 ± 19.3
	GnRHa	1-13 µg/kg	38	34	89	29	76	86.6 ± 8.5	81.7 ± 13.8	78.3 ± 16.2
1983 Volga river	SPE	25-35 mg/fish	54	50	93	31	57	86.5 ± 8.5	80.3 ± 11.1	81.4 ± 11.7
	GnRHa	0.2-5 µg/kg	40	34	85	25	63	85.6 ± 10.4	82.1 ± 11.0	78.2 ± 15.0
1984 Volga river	SPE	25-35 mg/fish	56	39	70	33	59	85.3 ± 9.6	76.7 ± 13.0	77.4 ± 15.4
	GnRHa	0.6-1.7 µg/kg	56	41	73	33	59	87.4 ± 9.8	80.5 ± 15.8	78.8 ± 16.9
1987 Kuban river	SPE	25-30 mg/fish	61	52	85	38	62		82.7 ± 2.0	
	GnRHa	0.5-1.0 µg/kg	45	43	96	38	84		88.2 ± 1.1	

* stages are given according to Dettlaff et al., 1981

any one moment is of greater importance in the stimulating of ovulation, than the nature of the stimulants compared. It should be noted that in the case of the Kuban population of the starred sturgeon GnRHa tends to be even more effective than SPE.

The developmental potential of eggs obtained from GnRHa- and SPE-treated females was compared, but only in case their fertilization rate was over 50 %, since eggs of lower initial quality normally are not incubated on stugeon fish farms. During most seasons no significant differences were obtained, or else the quality of the eggs obtained from GnRHa-treated females was slightly higher (Table 2).

Acipenser gueldenstaedti. Table 3 shows the results of the induction of ovulation in the Russian sturgeon with GnRHa's and SPE obtained in different years. Again the 1982 experiments were performed with des Gly^{10} [$D\text{-}Leu^{6}$]-GnRH ethylamide (Goncharov, 1984), while des Gly^{10} [$D\text{-}Ala^{6}$]-GnRH ethylamide was used in all subsequent studies. No special experiments were conducted to define the lowest effective dosage of GnRHa inducing ovulation in the Russian sturgeon. In 1982 the used doses of GnRHa , ranging from 25 to 4 μg/kg, were shown to be effective. Taking into account the high sensitivity of the starred sturgeon, a standard dose of GnRHa (20 μg/kg) was administered in the following experiments with the Russian sturgeon. Again no significant differences in the percentage of ovulated females, in the percentage of females with eggs of high quality, or in the developmental potentials of the eggs obtained upon GnRHa or SPE treatment (Table 3).

Huso huso. The study of the effect of GnRHa on ovulation in the beluga, biggest sturgeon of the Caspian Sea, was carried out in 1984 and 1986. Since the body weight of beluga females varies to a greater extent than those of the starred sturgeon and the Russian sturgeon, the GnRHa standard dosage was not used. Beluga females were administered with GnRHa doses that corresponded in microgram to their weight (in kilogram) determined visually. The actual dosage per kilogram was determined after the ovulated females were sacrificed and weighed.

Experiments with beluga females necessitated application of lyophilized GnRHa preparation, since the concentration of "Biolar" GnRHa solution (20 μg/ml) was not suitable for injection into such a huge species. To diminish the injection volume and, hence, the leakage of the solution, a more concentrated GnRHa solution was used - 50 μg/ml. Moreover, when injecting the females whose weight was over 150 kg, the total dosage was divided into two which were injected into both sides of the dorsal musculature.

Like the previous two sturgeon species of the genus *Acipenser*, the beluga, a member of another genus *Huso*, was shown to be highly sensitive to GnRHa. The lowest tested dosage of 0.6 μg/kg precipitated ovulation. During the two years of experiments only a limited number of beluga females could be stimulated but the uniformity of their reaction provides evidence of the equal effectiveness of hormonal treatments compared and equal quality of the eggs obtained as a result of these stimulations (Table 4).

Acipenser ruthenus. By 1987 the sterlet was the fourth species of sturgeons whose capacity to ovulate in response to GnRHa treatment was examined. Unexpectedly, administration of 1 - 3 μg/kg of GnRHa resulted in a significantly poorer ovulation response as compared to those induced by SPE injection (Table 5). In this experiment all females were injected simultaneously. Fifty seven hours later, when mature eggs clearly could be no longer expected, the 16 unovulated females from GnRHa group were repeatedly injected with two GnRHa doses of 1 or 10 μg/kg. The second treatment resulted in 50 % ovulation response independently of the dosage used. The quality of the eggs obtained from GnRHa- and SPE-treated females was roughly equal, as shown by records of their early development (Table 5). For technical reasons the embryos were transferred to another farm, and we could no longer follow their development.

In 1988 an experiment to elucidate the dose-response relationship in the sterlet was performed. Three GnRHa dosages differing fivefold were administered in single or double (spaced by 13 h) injections. The results of this experiment, presented in Table 5,

Table 3 - Effects of sturgeon pituitary extract (SPE) and gonadotropin-releasing hormone analogue (GnRHa) on ovulation and egg quality in the Russian sturgeon (*Acipenser gueldenstaedti*)

Experiment	Treatment	Dose	No. of fish	Fish ovulated		Fish ovulated (high quality eggs)		Fertilization rate, % (M ± SE)	Normal embryos according to the stage,% (M± SE).	
				No.	%	No.	%		17*	35*
1982 Volga river	SPE	30 mg/fish	35	33	94	29	83	78.8 ± 15.9	74.7 ± 17.2	84.0 ± 14.5
	GnRHa	4-25 µg/kg	17	16	94	14	82	76.8 ± 15.2	69.9 ± 19.2	80.0 ± 17.6
1984 Volga river	SPE	30-40 mg/fish	55	47	85	38	69	82.9 ± 11.4	76.9 ± 13.6	72.0 ± 18.8
	GnRHa	0.5-1.7 µg/kg	55	47	85	39	71	80.6 ± 12.9	74.8 ± 14.1	70.0 ± 19.3
1987 Kuban river	SPE	60 mg/fish	5	5	100	5	100		83.4 ± 9.0	
	GnRHa	0.6-0.9 µg/kg	5	5	100	5	100		90.2 ± 4.6	

* stages are given according to Dettlaff et al., 1981

Table 4 - Effects of sturgeon pituitary extract (SPE) and gonadotropin-releasing hormone analogue (GnRHa) on ovulation and egg quality in the beluga (*Huso huso*).

Experiment	Treatment	Dose	No. of fish	Fish ovulated		Fish ovulated (high quality eggs)		Fertilization rate, % (M ± SE)	Normal embryos according to the stages, % (M ± SE)	
				No.	%	No.	%		17*	35*
1984 Volga river	SPE	250-350 mg/fish	8	8	100	7	88	91.6 ± 5.8	78.8 ± 19.3	90.3 ± 4.9
	GnRHa	0.6-1.1 µg/kg	8	8	100	8	100	92.4 ± 2.0	77.0 ± 13.1	72.0 ± 10.9
1986 Volga river	SPE	250-350 mg/fish	21	21	100	21	100	90.0 ± 7.5	83.7 ± 10.3	86.0 ± 9.3
	GnRHa	0.9-1.8 µg/kg	21	21	100	21	100	93.4 ± 3.6	85.7 ± 5.1	89.5 ± 7.1

* stages are given according to Dettlaff et al., 1981

Table 5 - Effects of sturgeon pituitary extract (SPE) and gonadotropin-releasing hormone analogue (GnRHa) on ovulation and egg quality in the sterlet (*A. ruthenus*)

Experiment	Treatment	Dose	No. of fish	Fish ovulated		Fish ovulated (high quality eggs)		Fertilization rate, % (M ± SE)	Normal embryos at stage 17 % (M ± SE)
				No.	%	No.	%		
1987 Volga river	SPE	4 mg/fish	32	29	91	27	84	92.1 ± 3.5	89.6 ± 4.8
	GnRHa	1-3 µg/kg	32	14	44	9	28	94.5 ± 2.5	95.0 ± 2.3
	GnRHa	1 µg/kg	8[1]	4	50	4	50	88.6 ± 7.6	
	GnRHa	10 µg/kg	8[1]	4	50	3	38		
1988 Volga river	GnRHa	0.2 µg/kg	10	1	10				
	GnRHa	0.2 + 0.2[2] µg/kg	10	3	33				
	GnRHa	1 µg/kg	10	1	10				
	GnRHa	1 + 1 µg/kg[2]	10	7	70				
	GnRHa	5 µg/kg	9	4	44				
	GnRHa	5 + 5 µg/kg[2]	9	6	67				
	saline control		7	0	0				

(1) The unovulated females, injected 57h earlier with 1-3 µg/kg GnRHa, were used in this experiment
(2) Double injections were spaced by 13 h.

show that the population of the sterlet under study was highly heterogenous in terms of GnRHa sensitivity. One individual ovulated after administration of 0.2 µg/kg GnRHa, while others failed to respond to a dose of 5 µg/kg. Double injections were more effective than single ones, even if the total dosage was lower. The proportion of females that ovulated in response to double treatment, did not, however, reach those observed in a 1987 experiment. Since no SPE treatment was made in 1988, it remains unknown whether the ovulation potential of this population was lower than in 1987, or wether we have not yet developed the optimal of GnRHa application.

The timing of ovulation induced by SPE or GnRHa was analyzed in 1983 and 1984 in the starred sturgeon and the Russian sturgeon, and in 1984 and 1986 in the beluga. To compare the results obtained at different temperatures, we expressed the interval between a stimulant injection and ovulation as the number of τ_0 (duration of a single mitotic division of the early period of egg cleavage at a given temperature for details of this method see Dettlaff et al., 1981, 1987). Only females from which eggs of high quality were obtained, were considered. Fig 1 shows the duration of the latent period of ovulation as induced by two hormonal preparations in three sturgeon species.

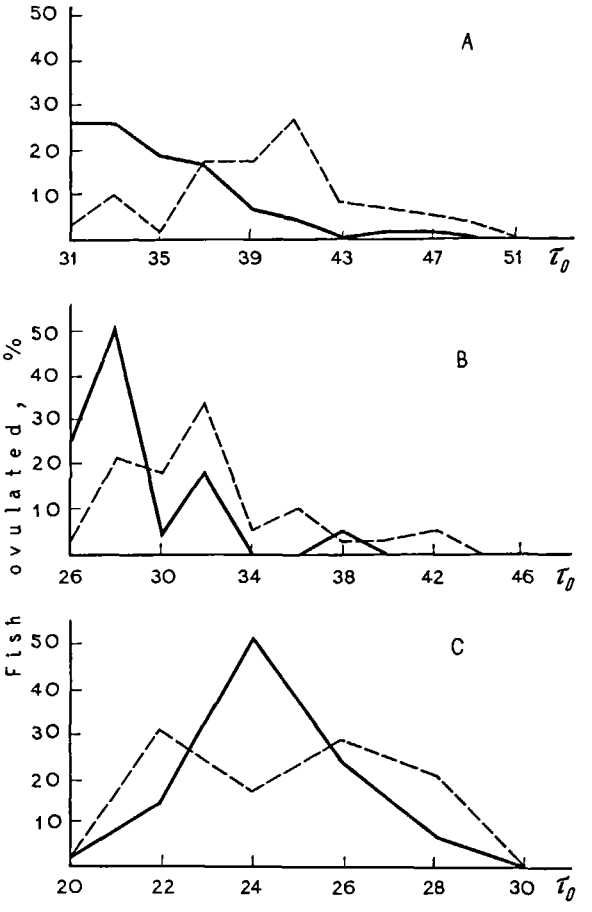


Figure 1 - Time of ovulation induced by sturgeon pituitary extract (solid line) and gonadotropin-releasing hormone analogue (broken line) in the starred sturgeon (A), Russian sturgeon (B) and Beluga (C). Only females with mature, ovulated eggs of high quality are included

It can be seen (Fig. 1A and 1B) that starred sturgeon and Russian sturgeon females ovulated on the average later under the influence of GnRHa as compared with the time of ovulation induced by SPE. The mean time of ovulation calculated for the starred sturgeon was 28.8 τ_0 (for 70 females in the SPE group) and 32.0 τ_0 (for 59 females in the GnRHa group), that for the Russian sturgeon was 34.7 τ_0 (for 38 females in the SPE group) and 39.5 τ_0 (for 39 females in the GnRHa group). The differences are significant for both species ($P<0.01$).

It should be emphasized that the different patterns of distribution of ovulated females in the time scale upon administration of SPE vs GnRHa is related to the fact that in accordance with the fish farm's operational schedule the first inspection on the presence of free ova usually occured when a considerable part of SPE-treated females had already ovulated. The reasonable delay of insemination of ovulated eggs does not influence their developmental potential, but reduces the number of inspections. The latter prevented us from determining precisely the mean time of ovulation in the case of SPE injection. Thus, the actual differences in the mean time of ovulation, induced by GnRHa and SPE are somewhat greater.

Contrary to the starred sturgeon and the Russian sturgeon, beluga females treated with SPE or GnRHa ovulated practically within the same time interval (Fig. 1C). The distribution of ovulated females in the time scale was slightly different, but the mean time of ovulation was virtually the same, i.e. 24.3 and 24.4 τ_0 respectively, for the SPE- and GnRHa-treated fish. Each experimental group included 29 females.

Comparison of the effects of GnRHa and SPE on spermiation and sperm quality in Acipenserid fish

Acipenser stellatus. The effect of GnRHa on spermiation in the starred sturgeon was reported earlier (Goncharov, 1984), but a careful comparison of stimulation techniques was made as late as 1984. Starred sturgeon males were injected with a dosage of 10 µg/fish, which proved highly effective in inducing ovulation, or with SPE. Since the milt was used for egg insemination, we were obliged to examine the males for the presence of free sperm as females ovulated. The number of males inspected at any one moment was determined by the number of ovulated females. The conditions of inspecting maturing males were similar in the GnRHa and the SPE groups, thus permitting the comparison of the effectiveness of administered hormonal preparations.

The results presented in Table 6 show that there was no difference either in the percentage of males that responded to the treatment with the hormonal preparations compared, or in the percentage of males producing sperm of high quality, or in the mean volume of milt obtained during first collection.

Acipenser gueldenstaedti. The experiments were carried out in 1984. All details concerning the conditions of induction experiments discussed in the previous section are applicable to the Russian sturgeon. Table 6 shows that in this species too the effectiveness of GnRHa is equal to that of SPE.

Acipenser ruthenus. No comparison with SPE was made in 1988, when the effects of two doses of GnRHa (1 and 5 µg/fish) on spermiation in the sterlet were studied. Like females, the males of this species were found to be less sensitive to GnRHa as compared to the reaction of starred sturgeon and Russian sturgeon males. None of the 30 males responded to the low dosage of GnRHa. The milt was obtained only from 14 of 29 males injected with 5 µg/fish of preparation. Clearly, it is a significantly less-pronounced reaction, than those normally observed after SPE administration. When the same males were injected repeatedly with 10 µg/fish of GnRHa two days after the first injection, most of them produced voluminous milt, and their sperm was of high quality.

Table 6 - Effects of sturgeon pituitary extract (SPE) and gonadotropin-releasing hormone analogue (GnRHa) on spermiation, sperm quality and milt volume in sturgeon fishes (*Acipenser stellatus* and *Acipenser gueldenstaedti*).

Species	Treatment	Dose	No. of fish	Fish spermiating		Fish spermiating (high quality sperm)		Milt volume (first collection) ml
				No.	%	No.	%	M ± SE
Acipenser stellatus	SPE	10-15 mg/fish	47	42	89	33	70	65 ± 34
	GnRHa	10 µg/fish	51	43	84	36	71	73 ± 31
Acipenser gueldenstaedti	SPE	15-20 mg/fish	36	35	97	28	78	132 ± 64
	GnRHa	20 µg/fish	32	30	94	26	81	153 ± 77

DISCUSSION

The results of this study along with those published previously (Goncharov, 1984) clearly show that synthetic analogue of mammalian GnRH are highly effective in stimulating oocyte maturation, ovulation and spermiation in Acipenserid fish. To our knowledge, 0.15 μg/kg body weight of des Gly^{10} [D-Ala^{6}]-GnRH ethylamide is the lowest effective dosage ever reported to induce ovulation in fish. Such a high effectiveness of GnRHa has been revealed in dose-response experiments on the starred sturgeon. The lowest tested dosages of GnRHa (0.5 and 0.6 μg/kg) were also effective in the Russian sturgeon and the beluga. Since in propagation practice it is not reasonable to use dosages approaching minimal ones, in most experiments the dosages in a range of 1 μg/kg were used.

With special reference to the problem of replacing the traditionally used sturgeon pituitaries by a more advantageous preparation to induce gamete maturation, an extensive comparative study was performed. No differences were found in the effectiveness of SPE and GnRHa. The percentage of females and males that responded to both hormonal treatments by the production of mature gametes of high quality was practically the same in different local populations and in different seasons. The ovulation was complete in these females after both treatments. Equal developmental potentials of mature eggs were also demonstrated. When possible in most experiments we monitored the development of sturgeon embryos up to hatching. Members of the Volgograd sturgeon fish farm compared the progeny of the beluga obtained from SPE- and GnRHa-treated females on releasing the young fish into the Volga river. They observed no significant differences in fish body weight or their survival in rearing ponds. Thus, treatment of Acipenserid fish with GnRHa appears to affect normal physiological process without causing any abnormalities in the reproductive systems. The coincidence in the effectiveness of gonadotropic and gonadotropin-releasing preparations indicates that the failure in obtaining high quality gametes or even mature gametes in some individuales is most probably due not to inadequate hormonal treatment, but rather to incomplete readiness of the gonads to respond to gonadotropin surge, or to the pathological state of the gonads.

In contrast to three migrant species of sturgeons under study, the sterlet that permanently inhabits fresh water demonstrated a great diversity in sensitivity to GnRHa. Administration of a single dosage of 0.2 or 1 μg/kg proved effective in some females, while in others even two spaced injections of 5 μg/kg of GnRHa failed to precipitate an ovulatory response. Problems were also faced in induction of spermiation in this species by a single injection of GnRHa.

It is suggested that the great diversity in the sensitivity of different fish species can be attributed to the evolution of GnRH molecules and their receptors, as well as to the evolution of mechanisms of hormonal regulation of final gamete maturation (Donaldson et al., 1983). It would be very attractive to speculate on the high potency of mammalian GnRH and its analogues in sturgeons, taking into account the possible identity or close homology in the structure of their gonadotropin-releasing hormones (Sherwood, 1987). Another possible explanation of the high sensitivity of sturgeons to GnRHa may be the absence or low level of the inhibitory regulation of gonadotropin secretion by dopamine, that is characteristic for many teleost fish (Peter et al., 1986). More comparative studies of the species differing by their taxonomy and life histories are called for to fully understand the nature of this problem.

The preliminary experiments carried out on the freshwater sturgeons, *Acipenser baeri Brandt*, reared up to maturity in captivity (Goncharov et al., unpublished), and *Pseudoscaphirhynchus kaufmanni* Bogdanow, maintained in captivity (Goncharov et al., unpublished), have shown that these species are also capable of responding to low doses of mammalian GnRHa. However no conclusion about the variability in sensitivity to this preparation can yet be made for these species. Des Gly^{10} [D-Ala^{6}]-GnRH ethylamide has also been successfully used to induce ovulation in the white sturgeon (*Acipenser transmontanus* Richardson) (Doroshov et al.,1984).

The single dosage of 100 µg/kg was applied. It seems to be injustifiably high, taking into account the marked sensitivity of other sturgeon species to this GnRHa, as has been shown in the present study.

It has been reported that synthetic mammalian GnRH can also be effective in inducing ovulation in the starred sturgeon and the Russian sturgeon, but only when two injections spaced by 5-6 h were administered. The reported effective dosages of GnRH were 100-200 + 200 µg/fish for the starred sturgeon, and 200 + 200-400 µg/fish for the Russian sturgeon (Barannikova et al., 1984, 1985 ; Barannikova et al., 1984). A single dose of synthetic GnRH (50 µg/fish) proved effective in inducing ovulatiion in the starred sturgeon (Goncharov, 1984). In the sterlet the mature eggs were obtained after a single administration of 100 and 300 µg/fish to females weighing from 0.35 to 1.5 kg (Horvath et al., 1986). Spermiation was induced by a single application of GnRH (100-200 µg/fish) in the starred sturgeon or by two spaced injections (100-200 + 200-300 µg/fish) in the Russian sturgeon (Barannikova et al., 1984 ; Barannikova et al., 1984). Since neither dose-response experiments with GnRH, nor a correct intra-populational comparison of specific activities of GnRH and GnRHa has been undertaken, no final conclusion can be made on the superactivity of the GnRH analogues in sturgeons. Nevertheless, the available data suggest that this is actually the case.

In this study we have found that GnRHa-induced ovulation occurs on the average later than those induced by SPE in the starred sturgeon and in the Russian sturgeon. This phenomenon has also been noted in the GnRH experiments on the starred sturgeon (Barannikova et al., 1982), while no differences in the time of ovulation induced by gonadotropic or gonadotropin-releasing preparations have been found neither in the Russian sturgeon (Barannikova et al., 1984), nor in our experiments on the beluga. The discrepancy of the results can be attributed to the differences in the physiological state of fish, or to an insufficient size of the sample.

CONCLUSION

A superactive analogue of mammalian gonadotropin-releasing hormone, des Gly^{10} [D-Ala^{6}]-GnRH ethylamide, is shown to be a highly effective compound inducing oocyte maturation, ovulation and spermiation in sturgeon fish. The effectiveness of GnRHa action administered in very low dosages via a single intramuscular injection as well as the quality of the obtained mature gametes are quite comparable with those of the conventional sturgeon pituitary extract injection. This is true for three sturgeon species propagated in the USSR, i.e. the starred sturgeon, the Russian sturgeon, and the beluga. Further research is called for to develop a reliable technique for GnRHa application in the sterlet and other sturgeon fish. An increased latent period of ovulation appears to be a specific feature of GnRHa action on some sturgeons.

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MANAGEMENT OF FEMALE SPAWNERS OF THE SIBERIAN STURGEON, *AcipenSer Baeri* BRANDT : FIRST RESULTS

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RESUME

L'essentiel des résultats présentés a été obtenu sur 2 cohortes d'esturgeons élevés en pisciculture. Une vernalisation annuelle semble nécessaire au moins pour synchroniser l'ovogenèse qui est bisannuelle pour 50 % des femelles et ceci au mieux à partir de 7 ans, âge correspondant aux pubertés les plus précoces. La compétence à la maturation des follicules in vitro est un meilleur critère de choix des femelles que la position de la vésicule germinale. L'ovulation est obtenue après une stimulation hormonale avec de la poudre hypophysaire de carpe qui entraîne un délai de réponse, fonction de la température, plus long qu'avec des hypophyses d'esturgeon. Les ovules sont récupérés par plusieurs pressions abdominales espacées. Les meilleurs taux de fécondation sont obtenus dans un délai compris entre 6 heures et 16 heures après le début de l'ovulation. La quantité d'ovules obtenus semble mieux corrélée à l'âge des femelles qu'à leur poids. Le poids moyen des ovules (p = 20,6 mg) a permis d'estimer la fécondité relative à $7{,}4.10^3$ oeufs/kg. Par rapport au nombre de femelles disponibles, seulement 15 % en moyenne ovulent chaque année avec des écarts importants. La durée d'incubation entre la fécondation et les premières éclosions est fonction de la température. Enfin quelques caractéristiques biologiques sont comparées à celles des esturgeons sauvages.

Mots clés : *Acipenser baeri*, élevage, choix des géniteurs, reproduction artificielle, ovulation, maturation *in vitro*.

ABSTRACT

Work was carried out with two generations of raised sturgeons. An annual vernalization appears to be necessary at least for synchronizing oogenesis which is biennial in 50 % of broodfish. First puberties are observed in 7 year old females. The in vitro maturation competence of follicles is a better criterion of female selection than germinal vesicle position. Ovulation was observed after hormonal stimulation with carp hypophysis powder, and observed delay in ovulatory response is compared with stimulation with sturgeon pituitary extracts. The ovulatory response is temperature - dependant. The ova were collected mainly by strippings. Best fertilization success was observed between 6 and 16 hours after beginning ovulation. Quantity of ova seems to be more correlated with female age rather than with their weight. Ovum mean weight, 20,6 mg, allowed estimation of the mean relative fecundity $7{,}4.10^3$ eggs/kg. Only 15 % of all females spawn each year, fluctuations are important. The duration of eggs incubation from fertilization to hatching is temperature dependant. Some biological caracteristics are compared with those of wild sturgeons.

Key-words : *Acipenser baeri*, farming, breeder selection, artificial breeding, ovulation, *in vitro* maturation

INTRODUCTION

The Siberian sturgeon, *Acipenser baeri*, was introduced into France on a strictly experimental basis. However, the species fairly quickly showed a marked potential for growth, which seemed interesting to exploit (Barrucand et al., 1979). The impossibillity of basing any production on a regular supply of fry from abroad made it necessary to master the production of the species. Success was first obtained in 1981 (Williot et al., 1982 b) and was confirmed the following year (Williot et al., 1982 c). We then possessed arguments to justify the development of a programme of research on the farming of the Siberian sturgeon.

The aim of this work is to develop a means of managing a generation of female breeders. We wanted to produce precise information for this species as concerns :

puberty, selection of fish, response to hormonal injections, collection of gamete samples, quantity of ova obtained, frequency of laying.

We came upon four types of difficulty :

- It seems that in a natural environment, laying of females in this family of fish never takes place yearly. According to Makarov (1970), this is the case for *Acipenser gueldenstaedti, Acipenser stallatus* and *Huso huso*. Priegel and Wirth (1971) make the same observation for *Acipenser fulvescens*. As far as the *Acipenser baeri* is concerned, we have no precise information, although Votinov and Kas'yanov (1974) indicate that this species in the Lena river present a long sexual cycle. This results in more complex procedures and longer experiments.
- The small number of fish prevented us from producing strict experimental plans and obliged us to use methods which protected the fish.
- Although we had the data collected by Charlon et Williot (1978) concerning breeding techniques, this concerned other species than the Siberian sturgeon, and, apart from a few indications about the bester, no information was available on the rearing of sturgeon spawners.
- Finally, the initial inexistance, later relative inadaptation of experimental structures meant that we lost fish and were obliged to retain a particular type of management.

Although the work presented deals with females, a minimum of information will be given about males in order to insure a better understanding of the work as a whole.

MATERIAL AND METHODS

Fish and farming

The Siberian sturgeon which constitute the object of the present experiments belong to two generations. The first is made up of fish born in the USSR in 1974 and which arrived in France the next year. They come from spawners in the Lena river. The second generation used in experiments corresponds to progeniture obtained in France in 1981. In one case we will present results obtained on several females from 1982.

These fish are reared in the Donzacq pisciculture (Landes) which is supplied by water from a resurgence, the temperature of which is virtually constant all year round 16 ± 2° C.

At the beginning of our research work, we had no information concerning the influence of temperature on oogenesis in *Acipenser baeri*. However, two items of information about other sturgeons was to help us retain a means of managing this factor. On the one hand, we knew (Charlon et Williot, 1978) that the Soviets obtained reproduction of besters reared in ponds with large annual variations in water temperature. On other hand, Detlaff and Davydova (1979) showed that with *Acipenser stellatus* it was no longer possible to stimulate in vitro maturation of follicles nor to obtain the ovulation of females if they were kept more than four days at a temperature favourable for reproduction. By generalizing these results to include our experimental species, we concluded that this could mean that breeders, females in particular, should undergo a drop in temperature to obtain laying. We retained this hypothesis and changed breeders into colder water from November to February inclusive.

We arranged this successively in two ways : the first consisted in taking the fish to ponds in other piscicultures (Winters 1980-1981 and 1981-1982), the second, since 1983-1984, in stopping the renewal of water in the Donzacq ponds containing the fish. During the winter of 1982-1983 we coukd not do this for practical reasons. In all cases the water temperature is quickly balanced with that of the air and we noted minima of around 7 ± 2° C depending on the year. This vernalization is used for puberal fish of both sexes stocked together. For immature fish, vernalization takes place for the first time during the winter before the first probable lawing.

The increase in biomass has made it necessary since 1986 to install aerators allowing the degree of oxygen to be maintained at a level of at least 80 % of saturation.

The maximum biomass was 12 kg/m^2 during the summer period and was often less than 5 kg/m^2 during vernalization. The ponds have a natural base (clayey soil) and the depth of the water is between 0,80 m and 1 m.

The fish are fed twice a day and 6 days a week with commercial granules for trout (Humidity 11 %, gross protein 45 %, fats 8 %, mineral substances 14 %). The rate of daily distribution according to live weight is close to or slightly less than 1 % when the temperature is high (16 ± 2°C) and between 0,3 and 0,5 % during the vernalization period. In fact, according Akimova et al., (1979), *Acipenser baeri* is probably one of the few species of sturgeon which continues to feed in the winter and during reproduction migration.

Checking of fish - selection

In the autumn, before vernalization, the gonads are checked by making a small abdominal cut (< 1cm) which allows a few follicles to be removed. The observation concerns homogeneity, size, colour and shape. Colour especially will be important in determining over-maturity (Ginzburg, 1968).

Then the position of the nucleus is determined according to the method described by Kazanskij et al., (1978). This allows us to calculate a polarization index corresponding to the ratio of the distance separating the nucleus from the animal pole to the largest dimension of the oocyte. The nearer this number is to zero, the more the maturation is advanced. This is therefore an important element in the selection of females.

At the end of winter (beginning of March) the fish are taken out of the vernalization ponds and brought back inside the hatchery where we try to maintain the same water temperature as outside. We carry out the same operation as in autumn, plus a competence test for the maturation of follicles according to the same principle as that described by Detlaff and Skoblina (1969).

For each female, 4 series of 25 follicles are removed. The first is used for the observations described above. Two other series are incubated separately in 5 ml of solution (table 1), the first complemented with progesterone at 10 µg-ml^{-1}, the second since 1988 with 17 α, 20 dihydroxy progesterone (17 α, 20 OH.P) from Sigma at the rate of 1 µg-ml^{-1} knowing that this steroid induces in vitro maturation of *Acipenser transmontanus* follicles (Lutes, 1985). The last series is incubated in just 5 ml of solution as a reference.

Table 1 - Characteristics of solution used for in vitro incubation of A. *baeri* follicles

Constituant	m.mole . l^{-1}
$CaCl_2, 2H_2O$	1,95
$MgCl_2, 6H_2O$	0,85
KCl	2,7
Na_2SO_4	0,7
NaCl	127,5
HePes	20
Sterilized and toxin free water	
pH (20°C) brought to ~ 7,5 - 7,8 (Na OH) Osmotic pressure = 270 ± 10 m.osmoles	

The incubation temperature retained is the same as that of the water in which the fish are kept. The length of incubation is 48 hours.

The disparition of the germinal vesicle, i.e. the bursting of the nucleus shell, shows maturation competence and constitutes the test of selection for the hormonal

injection. The result is expressed as the ratio between the number of follicles where the germinal vesicle has disappeared, and the total number (25) incubated. This ratio which varies from 0 to 1 constitutes the maturation index.

As far as the males are concerned, the presence of sperm in a fluted probe pushed into one of the gonads through a small opening is used as a criterion for selection.

Abdominal cuts made on fish are sewn up with one or two stitches of chrome catgut.

To limit loss of information we now use double tagging, one type of which is a ring tag from Hall Print put at the base of the dorsal and the other is a Tip-Tag for livestock at one of the pectorals.

Hormonal injection, collection of gametes, fertilization and incubation

- Hormonal stimulation is nearly always carried out with carp hypophysis powder at a rate of de 4 to 6 $mg.kg^{-1}$ for females and 2 $mg.kg^{-1}$ for males. It is finely pounded with a pestle in a mortar in NaCl at 9 ‰. The solution obtained is administered as an intramuscular injection.

Just once we used an analogue of LH.RH (des - Gly^{10} -[D - Ala^{6}] - Ethylamide from SIGMA) on several fish in the same year (Williot, 1986).

Ovulation is marked by the presence of ova in the tanks where females are stocked individually.

- Approximately 2 hours before the anticipated time of ovulation, sperm is removed from the males by massage. Observation under microscope allows us to retain those which present the best motility during two minutes.

Collection of ova is usually done by massage (Williot et al., 1982a) at a rate of every two hours approximately. Much more rarely we carry out a caesarean (Williot et al., 1982a).

- Fertilization is obtained by adding per kilo of ova 10 ml (Charlon et Williot, 1978) of a mixture of 3 sperm diluted at 1/200 (Ginzburg, 1968) in water.

For 7 females, we estimated the average weight of ova by weighing 40 to 60. This result and the total weight of the ova allowed us to calculate relative fecundity :

$$rf = \frac{\text{number of ova}}{\text{total live weight - weight of ova}}$$

After fertilization (2 to 3 minutes) the eggs are treated for 45 minutes in a watery suspension of fine clay to neutralize the sticky envelop secreted by the eggs. The clay and water are sterilized before use.

After being rinsed, the eggs are incubated. The fertilization rate is estimated by counting 200 to 300 eggs between stages 6 and 10 (4th and 7th segmentations) according to the description made by Ginzburg et al., (1969). In order to evaluate the influence of the delay between the beginning of ovulation and each collection on the rate of fertilization, series of eggs from 5 females were incubated separately.

The numbers and characteristics of the females which ovulated are grouped together in table 2.

Statistical analysis

Spearmann's coefficient (r_s) allowed us to establish possible correlations. The probabilities correspond to bilateral tests (Sherer, 1984).

Table 2 - Number and characteristics of available, selected *A. baeri* females having ovulated

Year of experiment	Year of birth of fish	Total number of fish	Number of fish selected / number of fish having ovulated	Weight and standard error (kg) of females having ovulated
1981	1974	40	5/9	5,1 ± 0,3
1982	1974	32	4/8	5,2 ± 0,5
1983	1974	32	0/1	No ovulation
1984	1974	25	4,5/15[1]	March 5,1 ± 0,4
			3,5/4[1]	December 5,6 ± 0,9
1985	1974	19	1/1	5,5
1986	1974	19	3/4	8,3 ± 1,5
1987	1974	19	0/1	No ovulation
1988	1974	18	5/5	9,7 ± 0,9
1988	1981	12	1/2	11,2
1989	1974	16	6/7	9,4 ± 1,1
1989	1981	10	6/6	9,7 ± 1,0

[1] 0,5 = partial ovulation

RESULTS

Influence of type of hormone on maturation index

A test was carried out in March 1989 on a group of fish reared identically and having undergone vernalization (table 3). We found that the two steroids produce equivalent results. All fish, except female number 10, ovulated after stimulation.

Table 3 - Comparison of maturation indexes (%) in March 1989 of follicles incubated at 15 °C during 48 hours

Females	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Pro.[1]	100	100	100	100	96	92	96	96	100	54	100	100	100	100
17α 20ß[2]	100	100	96	96	92	96	88	100	96	42	100	100	92	96

[1] Progesterone
[2] 17α. 20 ß dihydroxy progesterone

Efficacity of mode of selection of females

Taking 22 females of the 1982 generation having undergone vernalization, we compared the indexes of polarization and maturation obtained with the 17 α, 20 ß dihydroxy progesterone. We observed that a polarization index of more than 0,1 always corresponds to a maturation index of < 40 %. On the other hand, for polarization indexes of ≤0,05, showing a very advanced migration of the germinal vesicle, we found very variable maturation indexes (0,12 à 1), thus revealing the limitations of the polarization index alone (Fig. 1).

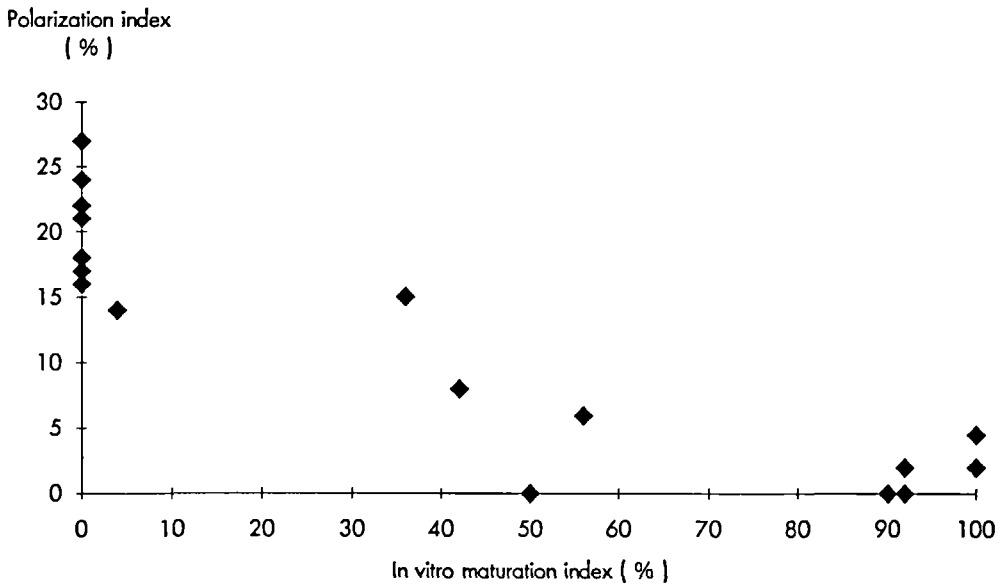


Figure 1 - Relation between polarization and in vitro maturation indexes (17 α - 20 DiOHP)

Retaining the maturation index as a criterion of selection, we expressed as a percentage the number of females having totally or partially ovulated compared to the number of fish selected, i.e. those having received a hormonal injection (table 2).

As far as the 1974 generation is concerned, we can make several remarks. For the three years, 1983, 1985, 1987, only one female was selected, whereas for all the other years the number selected was greater.

In 1983 and 1987 neither of the two females selected ovulated. Finally in 1984 layings were obtained in December.

For the 1981 generation, one of the two females selected in 1988 died a few hours after the hormonal injection. A blood sample taken just before the injection showed very pronounced anaemia (hematocrit ~ 5 %) which could account for this death.

Taking this observation into account, and, with reserve as regards the results of 1983 and 1987, we can note an improvement in the efficacity of the selection of females as years go by which is near to 80 %.

Beyond this global aspect, from a practical point of view it is interesting to know if this technique allows a precise choice as regards timing. To this end, we dispose of certain observations made in 1986 concerning the evolution of the results of the test for in vitro maturation on several fish (table 4). We noticed that the results evolve remarkably from one week to the next. At the first observation (12th - 13th March) all the females except the first one were apparently at the same physiological stage which would constitute subsequent confirmation of the management schedule retained.

Table 4 - Evolution of the in vitro maturation index (%) of follicles of 10 females.
(Figures rounded off to the nearest ten)

Females	Dates			
	12-13/03	18-19/03	27-28/03	9/04
1	80 [1][2]			
2	0		90 [1]	
3	0	[3]	10	[3]
4	10	70	80 [1]	
5	0	[3]	90 [1]	
6	10	60	50	0
7	10	40	40	[3]
8	0	40	30	100 [1]
9	0	[3]	50	[3]
10	0	70	30	[3]

[1] females having been injected
[2] only fish amongst preceding ones not having ovulated
[3] test impossible owing to errors in manipulation

4 out of 5 females presenting a maturation index of $\geq$ 0,8 and having received a hormonal injection, ovulated.

Delay between injection and ovulation

The experimental points corresponding to the number of hours separating the hormonal injection and the beginning of ovulation are shown in fig. 2.

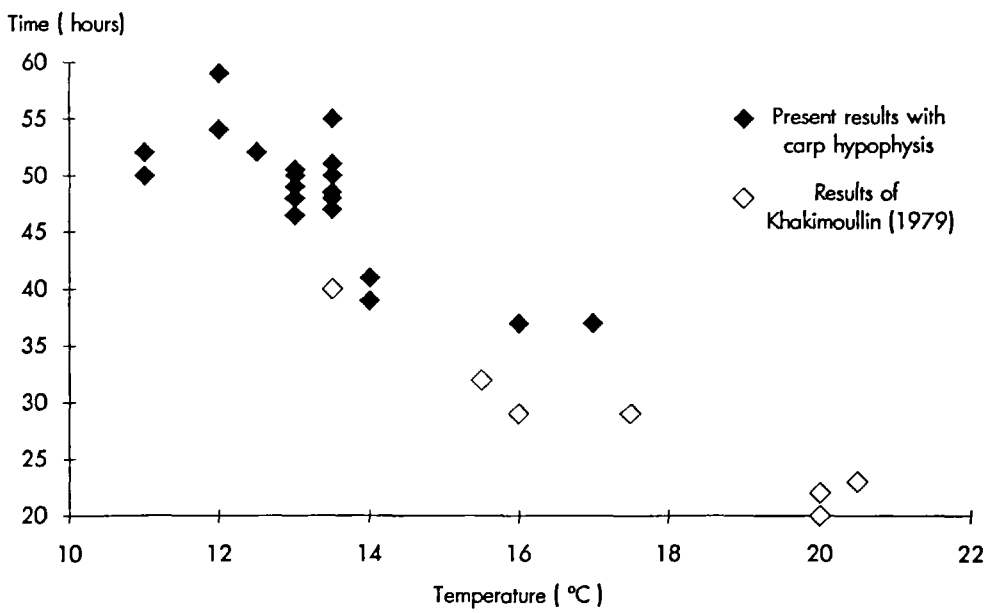


Figure 2 - Delay between injection and beginning of ovulation

We notice that this delay decreases if the temperature is increased (r_s = -0,85 , p<0,001).

In spite of this strong correlation, we observed a variability which must be taken into account as it can amount to 6 hours.

Elements of management with regard to available stock

Percentage of ovulation :

For each year, we noted the percentage of females ovulating compared to the total number of fish available for each generation (fig. 3).

From the results obtained with that of 1974 we observed that :

- in 1983 et 1987, the results were nil as already - mentioned above ;
- a biennial cycle seems to appear ;
- the average is around 15 %, but fluctuations are important.

We obtained the best results in 1989 with approximately 35 % of the stock of the 1974 generation and 60 % of that of 1981.

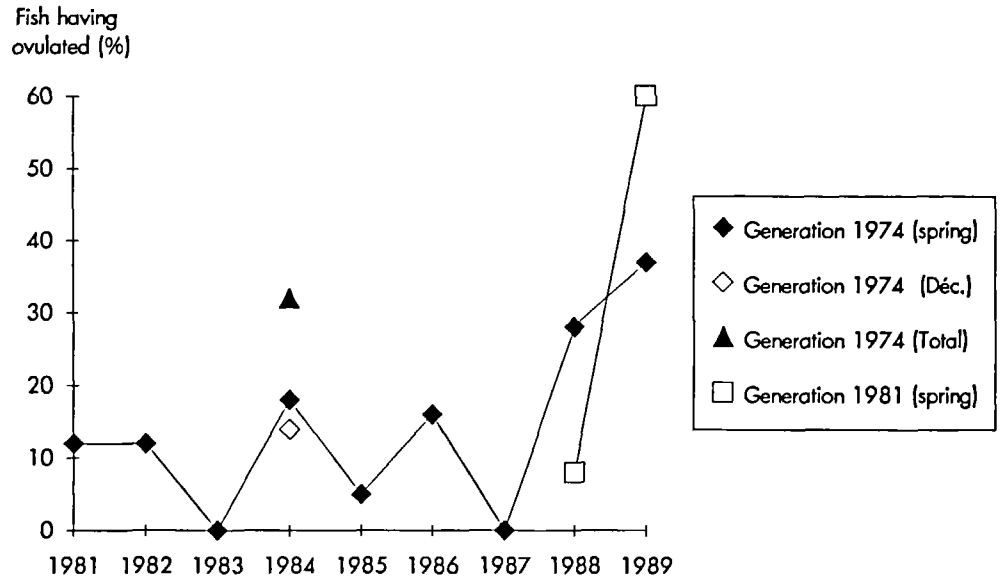


Figure 3 - Annual percentage of females having ovulated according to their total number for each generation

Probable frequency of laying :

This observation is based on the aforementioned real layings to which were added the observations of fish over-maturity. Some of them present an asynchronous oogenesis compared to the average of their generation, which means that if we had carried out more frequent checks, we would have been able to procure laying. These over-mature fish gradually resorb their oocytes and a new oogenesis develops. Thus, for a few marked fish, we were able to observe the interval between two probable layings from 17 cases (table 5). Although the total number of cases is low, we can retain two elements.

Table 5 - Probability of intervals between 2 layings

Interval between 2 Layings (years)	Probability %
1	12
2	47
3	17,5
4	23,5

The rate of return of biennial layings is the most probable (almost 50 % of cases).

Finally, there are possibilities of annual layings and also four-yearly ones.

Puberty :

With the management conditions described, the first sexual maturity was obtained at 7 years old for the 2 generations of females (table 2). This observation is confirmed for the 1982 generation (Williot, not published).

Males are sexually mature one year earlier.

For both sexes, these puberties only concern a part of the stock.

Samples of ova : number, quantity, average weight

The number of samples needed to obtain the totality of the ova is indicated in table 6 according to the age of the females. From 12 years onwards, this number increases up to 9 to 10 collections.

Table 6 - Evolution of number of ova collections according to the generation and age of females

Generation	Age (years)	Number of samples collected / female
1981	7	3
	8	7,10,7,6,5,6
1974	8	6
	10	7,7,7,6,8,7,6
	12	9,10,11
	14	8,9
	15	10,10,8,10

The total quantity of ova varies between 0,2 and 2 kg per fish (Fig. 4 a and b).

The quantity of ova is more correlated to age (r_s = 0,68, $p<0,001$) than to weight (r_s = 0,43, $p<0,05$), (fig. 4 a and b).

The average weight of ova is 20,6 mg and leads to an average relative fertility of $7,4.10^3$ eggs/kg.

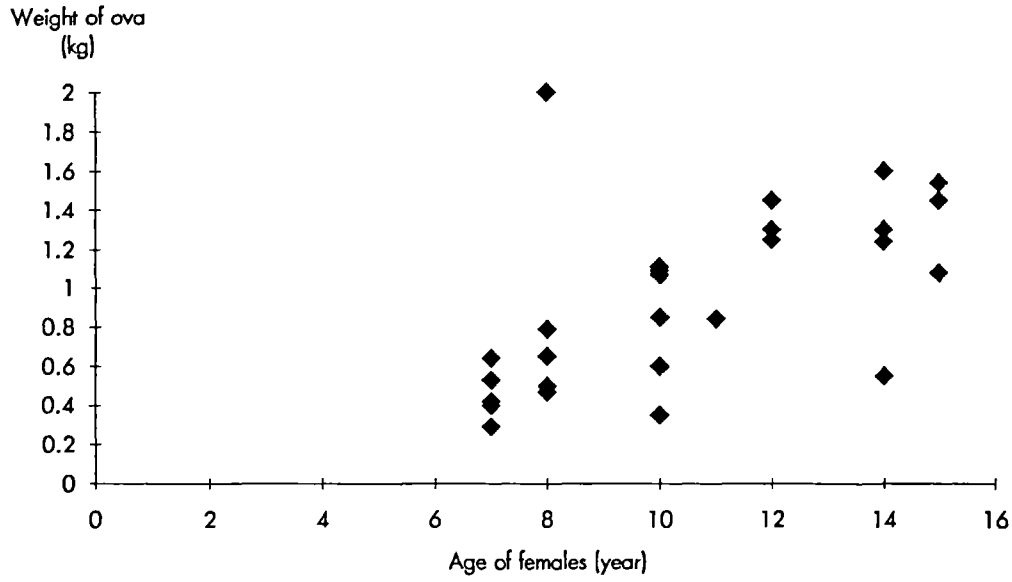


Figure 4a - Quantity of ova according to the age of the females

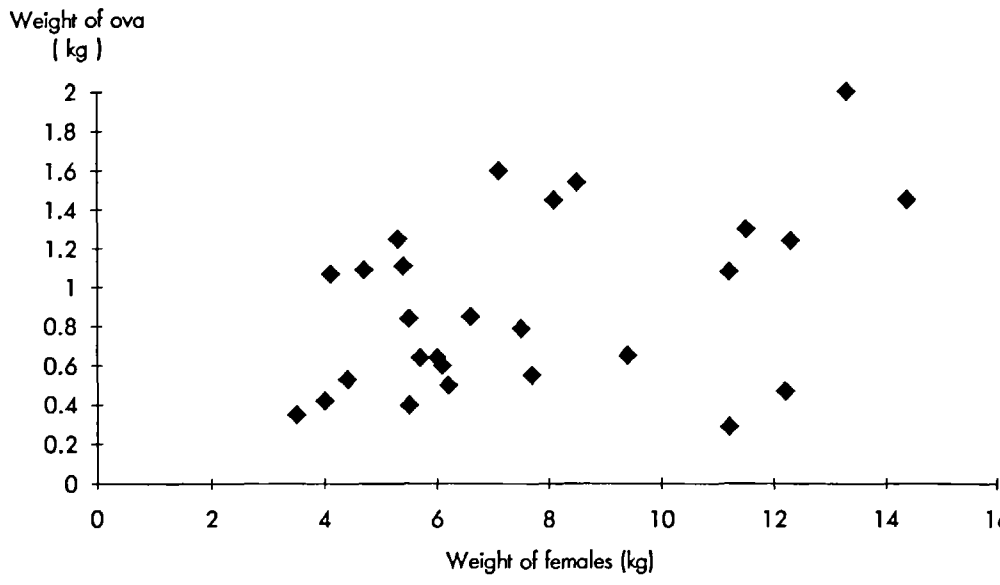


Figure 4b - Quantity of ova according to the weight of the females

Fertilization rate

In 1989, the fertilization rate was determined for each of the samples taken from 5 females ; the results are shown as a function of number of hours between the beginning of ovulation and the collecting of the sample (fig. 5).

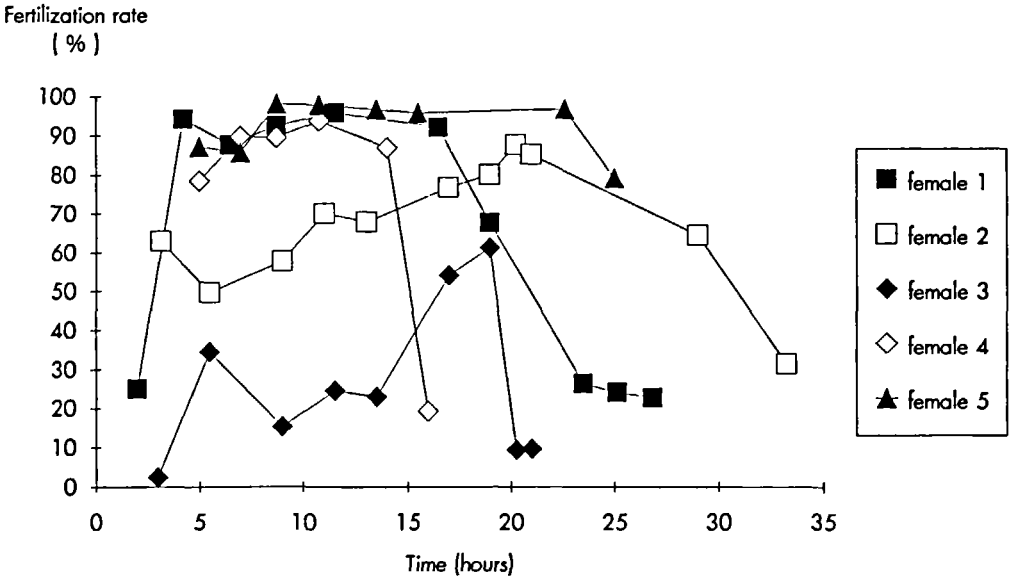


Figure 5 - Fertilization rate according to the delay between the beginning of ovulation and the collecting of each sample of ova

The poor general result for female number 3 is most probably due to the fact that vernalization did not take place.

The general aspect of the curves allows us to make several remarks. The poorest fertilization rates are observed in ova samples collected shortly after the beginning of ovulation or beyond a certain length of time, 16 to 26 hours according to the fish.

"If we leave aside the case of female number 3 already mentioned, we can hope to obtain in the majority of cases fertilization rates superior or equal to 80 %.

Length of incubation

This data (Fig. 6) concerns the beginning of hatching, which can take under our conditions up to 48 hours. We noticed that the duration of embryogenesis diminishes if the temperature is increased (r_s = -0,87, p¼0,02).

DISCUSSION

Selection of females

From our results obtained under precise conditions, it is possible to indicate that the first criterion to be taken into account is age, as the first layings are obtained at 7 years (table 2), whereas they are observed at 11 or 12 years in the case of wild sturgeons (Sokolov and Malyutin, 1977).

For a population of spawners regrouping several age groups of puberal fish, it seems that the quantity of ova is more correlated to the age of the females than their weight (fig.4 a et b). This would mean that the weight of the females should not be retained as a criterion for selection. On the other hand, however useful these indications may be, they are not precise enough to allow us to choose females capable of laying after hormonal stimulation.

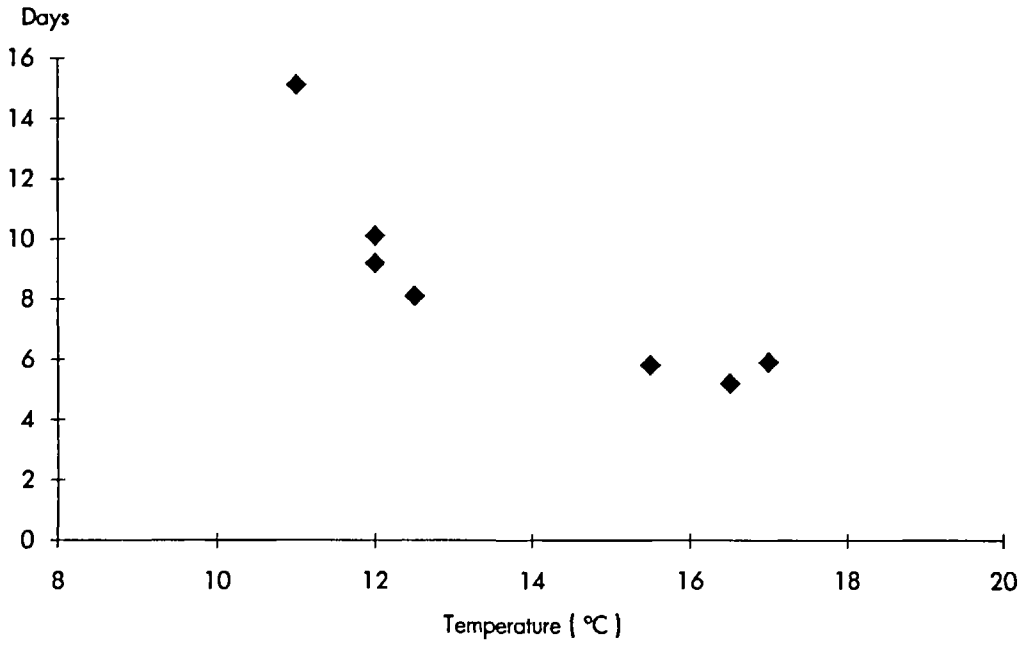


Figure 6 - Lenght of incubation from fertilization to beginning of hatching

It is for this reason that we have indicated that the maturation index is a better criterion for choice than the polarization index. It leads to a success rate around 80 %. This confirms the data published by Lutes et al. (1987) on wild *Acipenser transmontanus* females.

If we take as a maximum limit a value of 0,07 polarization index like Kazanskij et al. (1978), this would lead in our case to failure for 5 fish out of 11 (fig.1). An improvement in the efficacity of the selection of females could be brought about by a better understanding of endocrine phenomena of which the follicle is the centre and this to be linked with fish management. Indeed, if we generalized for all sturgeons the diagram proposed for *Acipenser stellatus* by Detlaff and Skoblina (1969) and better known in teleosts (Nagahama 1983, Nagahama and Yamashita 1989), follicle cells would play an essential role in the elaboration of Maturation Inducing Steroid (M.I.S) under the action of gonadotropin hormones. The use of the latter would then be preferable to induce in vitro maturation. This would avoid the short-circuiting of follicle cells which we know lose their efficacity in certain cases, according to the work of Detlaff and Davydova (1979) on *Acipenser stellatus*.

Another element of improvement could be to use hormonal stimulation on females for a maturation index of $\geq$ 90 % instead of 80 %. We noticed (female 1, table 4) that the gonads of this fish were still greasy and therefore oogenesis was insufficiently advanced.

Indeed biopsies carried out through a very small cut in the abdominal wall do not permit a general examination of gonads, and can lead to an incorrect estimation of the stage of gametogenesis. On the other hand more frequent biopsies of this kind, which traumatise the fish only to a very small extent, have allowed us to show that the discriminating power of the in vitro maturation test is a week at the most.

Elements of management of a generation of females owners

The yearly vernalization of *Acipenser baeri* spawners as from the last winter before probable laying (i.e. at 6,5 years old) has allowed us to obtain laying each year except in 1983 and 1987 (Fig. 3) i.e. failure 2 years out of 9, the explanation for which is probably not vernalization (or lack of it) even though it did not take place in 1982-1983.

This practice is partially confirmed by Kazanskij (1981) who indicates the possibility of procuring layings in *Acipenser gueldenstaedti* by stocking these fish for several months at a temperature of about 4 °C lower than that of laying. According to Giovannini (pers. comm.) the first artificial reproduction of *Acipenser naccarii* in 1988 (Arlati et al., 1988) was carried out on fish bred for 10 years in a fishfarm where the water presented a pronounced annual thermic cycle.

Finally the fact that the majority of the fish, 9 out of 10 (table 4), were in a very similar physiological state shows that vernalization has the advantage of synchronizing oogenesis.

The laying period takes place at present from March to mid-April whereas in the Lena river it would probably take place at the earliest at the end of June, the period which corresponds to the peak of migration (Sokolov and Malyutin, 1977).

In 1984 we obtained layings in December (Fig. 3) from spawners not having undergone any decrease in temperature during the preceding 9 months. The fry from these layings did not survive long after weaning. Trusov (1964) reports the obtaining of *Acipenser gueldentstaedti* in September from late migrants thanks to the possibility of managing temperatures. It therefore seems possible to envisage for the future not only a staggering of the laying period, but also perhaps modifications through appropriate selection and fish management.

Almost all layings were obtained after hormonal stimulation with carp hypophysis powder. The recent reduction in efficient doses of analogues of LH-RH, 35 µg/kg on *Acipenser naccarii* (Arlati et al., 1988) instead of the 100 µg/kg on *Acipenser transmontanus* (Doroshov and Lutes, 1984) and the 150 µg/kg on *Acipenser stellatus* (Horvath et al., 1986), opens up the way to new and interesting possibilities with this type of hormone.

Delay in response of females :

Khakimoullin (1979) established the same type of curve as in figure 2. Although the range of temperature was not the same, it seems that the delays in response we observed were longer. The probable explanation of this prolongation could be the use of hormones from other species in our case while, although he did not specify, the previously cited author probably used sturgeon pituitary extracts.

Global results on ovulations, consequences :

We have shown (Fig. 3) that the percentage of ovulation within a given stock fluctuates considerably from one year to the next and that in 50 % of cases oogenesis is biennial (table 5). According to Sokolov (1965), the length of time between two spawning-season of Siberian sturgeons in the Lena river is three to six years, thus much longer than in our findings. This strong biennial frequency applied to a small number of fish probably explains the failures of 1983 et 1987.

This observations are of considerable importance for an industrial hatchery, because they mean that a better regularity of production will be obtained if two generations separated by one year are used simultaneously.

Samples of ova, fertilization rate :

The obtaining of ova by abdominal massage is not often used for sturgeons. Apart from our results we can mention those of Arlati et al. (1988) on *Acipenser naccarii*. The caesarean is used much more often, Smith et al. (1980) on *Acipenser oxyrhynchus*, Williot et al. (1982 b) on *Acipenser baeri*, Doroshov et al. (1983) on

Acipenser transmontanus and Horvath et al. (1986) on *Acipenser ruthenus*. In all these cases survival data is not given and only Williot et al. (1982 a) indicate a survival rate of three fish out of four. Although this result is satisfactory for a first attempt, due to low stocks we have moved towards a generalization of abdominal massage which has the advantage of preserving the fish. On the other hand, in the future the caesarean will be used again to facilitate work on females. For this we have a supplementary element which is the fertilization rate from samples (Fig. 5). In fact we obtained the best fertilization results between 6 and 16 hours after the beginning of ovulation. Thus, abdominal massages should be stepped up and a caesarean carried out in this lapse of time.

Relative fecundity :

For wild Siberian sturgeon, Sokolov and Malyutin (1977) report a relative fecundity (compared to kg of body weight) of between 10 and $37{,}5.10^3$ and note that this decreases with the weight of the females. These results, which are superior to ours, can be explained in two ways. Firstly, wild brood fish are lighter in weight, 1 to 5 kg compared with 5 to 10 kg in our case (Table 2) and the average weight of the ova seems inferior to those we dealt with, 11 to 25 mg compared with 17 to 25 mg.

The second reason stems from the fact that when we collect samples through massage, we sometimes lose ova during manipulation.

Doroshov and al. (1983) report a relative fecundity of $7{,}6\text{-}10^3$ to $10{,}9\text{-}10^3$ for wild *Acipenser transmontanus* females in artificial breeding conditions. These results are just only higher than ours.

CONCLUSION

Some of the objectives this work aimed to fulfil at least provisionally had been attained either on the Siberian sturgeon, as for example the delay in response to hormonal stimulation, or on other species of sturgeons as the method of selecting females.

The originality of these results lies in the fact that they have been obtained together on farm fish and that they provide supplementary elements of information on methods of managing spawners, number of females ovulating as a percentage of available stock, the relationship between the method of collecting samples and fertilization rates. So, we have very little comparable information available to enable us to estimate the reliability of these results.

What is more, we have shown that criteria such as delays in response to hormonal stimulation, number of females ovulating or fertilization rates present quite strong individual fluctuations. The use of this data for prospective purposes would entail a certain amount of uncertainty of which it has been taken into account when elaborating the development plans. It is probable that, if we could make use of this heterogeneity in the future, the performances would be improved.

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USE OF VITELLOGENIN LEVEL AS A MATURATIONAL INDICATOR FOR ARTIFICIAL SPAWNING OF CULTURED HYBRID STURGEON, *Huso huso* x *Acipenser ruthenus*

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RESUME

La relation entre le développement ovarien et la concentration en vitellogénine dans le sérum a été envisagée chez l'esturgeon hybride issu du croisement entre, *Huso huso* et *Acipenser ruthenus*. Une ponte artificielle de l'esturgeon était réalisée en utilisant de la LH-RHa. Les stades de maturation des ovaires étaient déterminés par mesure du diamètre des oocytes les plus développés. Les concentrations en vitellogénine étaient mesurées par méthode immunologique. Des implants d'um mélange de cholestérol et de beurre de cacao contenant de la LH-RHa (des-Gly^{10}[D-Ala^{6}]-LH-RH éthylamide) étaient utilisées à une dose de 1 mg par poisson pour l'induction de la ponte. Les concentrations de vitellogénine dans le sérum augmentaient avec le développement des oocytes, et elles atteignaient une valeur maximale de 10 mg/ml. Après ce pic, elles diminuaient rapidement. D'autre part, une protéine semblable à la vitellogénine était observée dans le sérum de femelle pondant des ovules sur-matures. Elle pourrait être dûe à la réabsorption des protéines de l'oeuf. Trois des cinq poissons ayant une faible concentration en vitellogénine (inférieure à 1 mg/ml) pondaient des oeufs normaux 2 à 3 jours après le traitement hormonal. En même temps, cinq des huit poissons ayant une forte concentration en vitellogénine (supérieure à 10 mg/ml) pondaient des oeufs sur-matures. Les autres ne réagissaient pas au traitement LH-RHa. La F2 était obtenue artificiellement à partir d'oeuf de femelle ayant peu de vitellogénine et de sperme traité avec des injections d'HCG ou d'hormones pituitaires de saumon. Ces résultats suggèrent que (1) la vitellogénine est un bon indicateur du développement ovarien de l'esturgeon, (2) pour une ponte artificielle, l'esturgeon peut être sélectionné selon son taux de vitellogénine, et (3) la LH-RHa est efficace pour l'induction de la ponte chez l'esturgeon.

Mots clés : esturgeon, vitellogénine, maturation, LH-RH, reproduction artificielle.

ABSTRACT

The relationship between ovarian development and serum vitellogenin levels was examined in the hybrid sturgeon, *Huso huso* x *Acipenser ruthenus*. Artificial spawning in the sturgeon was also done using des-Gly^{10}[D-Ala^{6}] luteinizing hormone releasing hormone ethylamide (LH-RHa). Maturational stages of ovaries were determined by measurement of diameter of the most developing oocytes. Vitellogenin levels were measured by immunological method. Cholesterol pellets of LH-RHa were used at a dose of 1 mg per fish for the induction of spawning. Serum vitellogenin concentrations increased as the oocytes developed, reached peak levels of 10 mg/ml, followed by a prompt decrease. On the other hand, a vitellogenin related protein was observed in serum of female with over-ripened eggs. This protein may be released into the blood during the reabsorption of egg yolk proteins. Three out of 5 experimental fish with low serum levels of vitellogenin (lower than 1 mg/ml) spawned 2-3 days following the LH-RHa treatment. In contrast, 5 out of 8 fish with high serum vitellogenin levels (higher than 10 mg/ml) discharged over-ripened eggs. The others could not spawn with LH-RHa. The F2 were reproduced artificially from eggs of females having low vitellogenin with sperm of males treated with human chorionic gonadotropin or salmon pituitary extracts. These results suggested that (1) vitellogenin is a good indicator of ovarian development in the sturgeon, (2) the sturgeon for artificial spawning can be selected according to serum vitellogenin level and (3) LH-RHa is effective for induction of spawning of the sturgeon.

key-words : sturgeon, vitellogenin, maturation, LH-RH, artificial spawning.

INTRODUCTION

Natural populations of the sturgeon have decreased and have been almost exterminated in Japan. Few during one year are caught by the coastal fishery, but recently there have been no reports confirming their spawning in the rivers. Few sturgeon can be seen in aquariums or research institutes. Sturgeon farmers would like to propagate them, and some trials have been undertaken to artificially induce spawning. All the trials in Japan never succeeded. There are some papers concerning the artificial spawning of sturgeon (Doroshov et al., 1983, Czeskleba 1985), but in most of these studies sturgeon from wild stocks or hybrids were used (reviewed by Doroshov 1985). It seems that artificial spawning from cultured sturgeon is very difficult. The present paper describes the relationship between ovarian development and serum vitellogenin concentrations, and the successful induction of artificial spawning with LH-RHa cholesterol pellets for the first time in Japan.

MATERIAL AND METHODS

Fish

Fertilized eggs of hybrid sturgeon, *Huso huso* x *Acipenser ruthenus* , were introduced from the Soviet Union in 1980, hatched and reared at the Inland Station, National Research Institute of Aquaculture, Mie, Japan. Fifty females weighing about 9.5 kg (mean) with vitellogenin in their sera were used for investigating the relationship between ovarian development and serum vitellogenin concentrations in March and June, 1988. These fish were killed for collecting ovaries after collecting blood samples. Fifteen females weighing about 10.0 kg (mean) with various concentrations of serum vitellogenin and 12 males weighing about 4.9 kg (mean) were used for artificial spawning in August, 1988. Fish were reared in fresh water (well water) which temperature was 12-16° C from March to August, 1988.

Preparation of antisera

The polyvalent antiserum against serum proteins of estradiol-17 beta treated sturgeon was produced by immunizing rabbit with a mixture of an equal volume of serum and Freund's complete adjuvant (Fujii et al., 1987). The antiserum against serum proteins of male sturgeon was produced similarly. Specific antiserum to vitellogenin was prepared by absorbing the antiserum against estradiol-17 beta treated fish with an equal volume of male serum.

Properties of the antisera were examined by the double immunodiffusion technique (Ouchterlony 1953) using 1.0% agarose gels.

Preparation of egg extract and vitellogenin

Egg extract was prepared as follows. A piece of ovary was removed, homogenized with same weight of cold 0.9% NaCl and centrifuged at 15,400 g for 60 min at 4° C to obtain a clear middle layer. The clear solution was used as an egg extract. Properties of the egg extract were examined by immunoelectrophoresis using a 1.0% agarose gel (Fujii et al., 1987).

Vitellogenin was purified as follows. Serum of female at vitellogenesis stage was dialysed against distilled water. The precipitate was separated by centrifugation at 2,000 g for 15 min at 4° C. The pellet was suspended in distilled water, recentrifuged and dissolved in 0.5 M NaCl. The solution was applied to a gel filtration column containing Sephadex G-200 (2.6 X 90 cm) equilibrated with 0.01M phosphate buffer, 0.5 M NaCl, pH 7.5. Protein contents of each fraction were measured by the method of Lowry et al., (1951). The markers used in the gel filtration were bovine thyroglobulin (mol. wt 670,000), bovine gamma globulin (mol. wt 158,000), chicken ovalbumin (mol. wt 44,000), horse myoglobin (mol. wt 17,000) and cyanocobalamin (mol. wt 1,350). The main

peak at a position of a molecular weight of about 400,000 was used as a purified vitellogenin. The purified vitellogenin was tested by the double immunodiffusion technique and SDS-PAGE (sodium-dodecyl-sulphate polyacrylamide gel electrophoresis). SDS-PAGE was caried out according to the procedure of Laemmli (1970) in slab gels of 7.5% polyacrylamide and stained with Coomassie Brilliant Blue R-250. The marker proteins used in the SDS-PAGE were rabbit skeletal muscle myosin (mol. wt 200,000), E. coli beta-galactosidase (mol. wt 116,250), rabbit muscle phosphorylase b (mol.wt 97,400), bovine serum albumin (mol. wt 66,200) and hen egg white ovalbumin (mol. wt 42,699).

Determination of maturational stage and vitellogenin concentration

Maturational stages of 50 females were determined by measurement of oocyte diameters. Concentrations of vitellogenin were measured by the single radial immunodiffusion technique (Mancini et al., 1965) using 1.0% agarose gels containing 2.0% specific antiserum to the vitellogenin. The absolute levels were quantitated using a standard curve of purified vitellogenin serially diluted with male serum.

Artificial spawning

a/ Preparation of LH-RHa cholesterol Pellet

Fifteen mg of synthetic luteinizing hormone-releasing hormone analogue (des-Gly^{10}[D-Ala^{6}]-LH-RH ethylamide, LH-RHa: Sigma) was dissolved in 0.4 ml of 50% ethanol and mixed with 250 mg of cholesterol (99%, Sigma) to form a paste-like substance. The paste was dried at 35-40° C to form a powder. The powder was mixed with 50 mg of melted cocoa butter. The pellet was produced by placing 20 mg of the mixture into a stainless steel tube (ID 2.0 mm) and removing it with a stainless steel stick (OD 2.0 mm). Each of the pellets contained 1.0 mg of LH-RHa.

b/ Hormone treatment

Each female was implanted intramuscularly with a cholesterol pellet of LH-RHa at a dose of 1.0 mg per fish. Each group of 4 males was injected intramuscularly with 5,000 units of human chorionic gonadotropin (HCG) dissolved in 0.9% NaCl or with salmon pituitary extract prepared by homogenizing 5 mature salmon (Oncorhynchus keta) pituitary glands or 5 mature silver carp (Hypophthalmichthys molitrix) pituitary glands in 0.9% NaCl.

RESULTS

Properties of antisera, purified vitellogenin and egg extract

Specific antiserum against vitellogenin formed a precipitin line with serum of female at vitellogenesis stage in the double immunodiffusion. It did not react with male serum (Fig. 1). This result indicates that the specific antiserum was absorbed with male serum sufficiently. The purified vitellogenin formed a precipitin line with the antiserum to vitellogenin, and did not react with the antiserum to male serum in the double immunodiffusion (Fig. 1). It displayed a single band in SDS-PAGE migrated at a position corresponding to a molecular weight of about 200,000 (Fig. 2). Similar band was observed in serum of female at vitellogenesis stage but not male serum. It is assumed that the sturgeon vitellogenin (mol. wt 400,000) consists of two identical polypeptide chains (mol. wt 200,000). These results indicate that vitellogenin was purified sufficiently. The egg extract showed three precipitin lines with antiserum to vitellogenin in immunoelectrophoresis (Fig. 3). A marked difference of the pattern from that of serum of female at vitellogenesis stage was the existence of a precipitin line extended toward the anode.

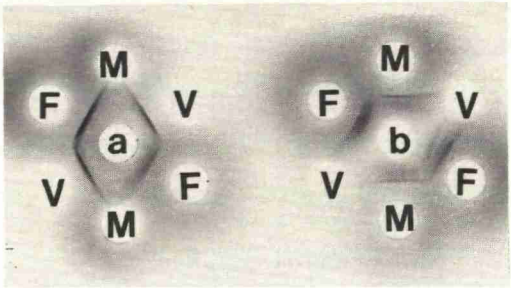


Figure 1 - Immunodiffusion of sturgeon sera and purified vitellogenin.
M : male serum,
F : serum of female at vitellogenesis stage,
V : purified vitellogenin,
a : antiserum against vitellogenin,
b : antiserum against male serum proteins

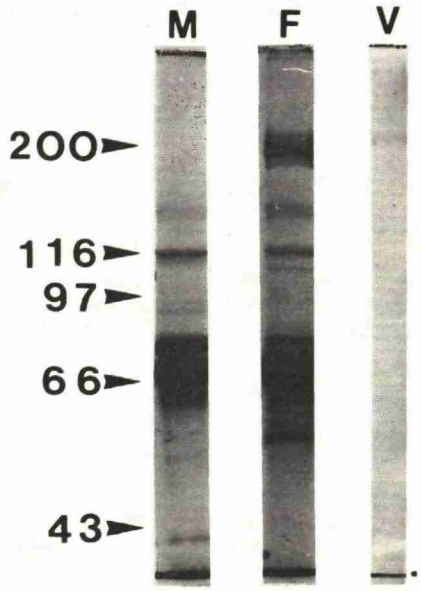


Figure 2 - SDS polyacrylamide gel electrophoresis on 7.5 % gel of sturgeon sera and purified vitellogenin. Molecular mass values are indicated in kDa on the left
M : male serum
F : serum of female at vitellogenesis stage,
V : purified vitellogenin,
arrow : band observed in F and V but not M

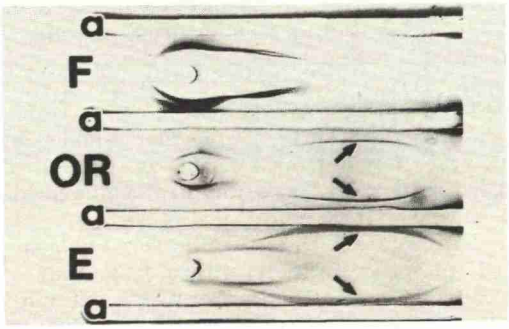


Figure 3 - Immunoelectrophoresis of sturgeon sera and egg extract
F : serum of female at vitellogenesis stage,
OR : serum of female with over-ripened eggs,
E : egg extract,
a : antiserum against vitellogenin,
arrow : precipitin line observed in E and OR but not F

Relationship between ovarian development and vitellogenin concentration

Figure 4 represents the relationship between oocyte growth and serum vitellogenin concentrations. The standards of purified vitellogenin more than 0.01 mg/ml could be distinguished from the blank (male serum) by the Mancini method. All females with serum vitellogenin had oocytes over 1.0 mm in diameter except one (0.9 mm). Serum vitellogenin tended to increase on oocyte growth. When the oocytes grew to about 2.6-2.8 mm, vitellogenin attained maximum levels (mean: 9.8 mg/ml). Subsequently, serum vitellogenin decreased to about 1/100th when the oocytes reached maximum size (about 3.0 mm). On the other hand, The serum vitellogenin levels of the fish with over-ripened eggs showed a wide range (2.3-28.0 mg/ml), and its mean level (13.6 mg/ml) was higher than that of the maximum level of females at vitellogenesis stage. The sera of females with over-ripened eggs formed precipitin lines with the antiserum to vitellogenin which were similar to those of the egg extract in immunoelectrophoresis (Fig. 3).

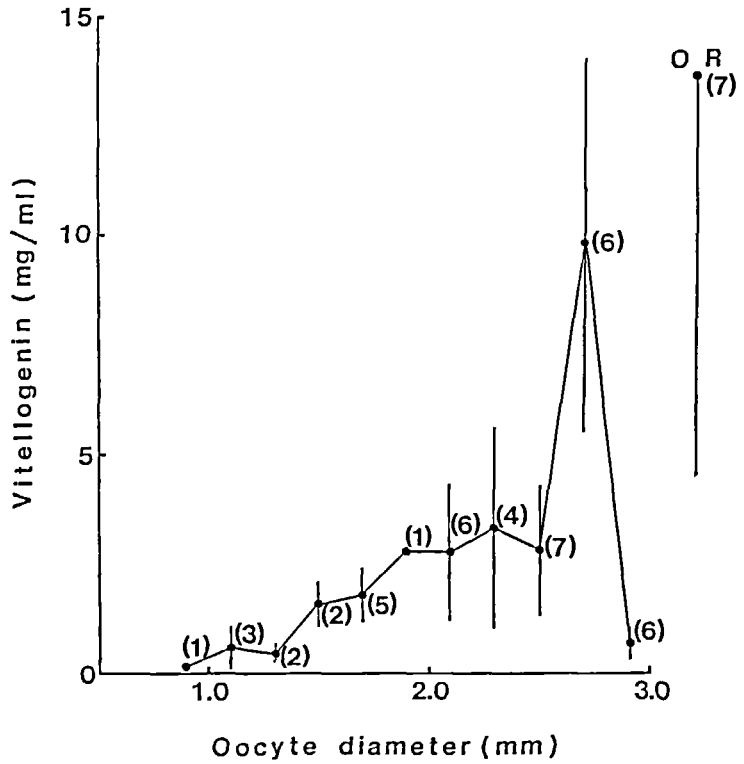


Figure 4 - Relationship between ovarian development and serum vitellogenin levels. Vertical bars represent the standard deviation. Numbers in parentheses represent numbers of fish. OR : concentrations of vitellogenin (or reabsorbed egg yolk protein) of females with overripened eggs

Artificial spawning

Fifteen females used for artificial spawning showed various serum vitellogenin levels (Table 1). Three out of 5 fish having low levels of vitellogenin (lower than 1.0 mg/ml) spawned 2-3 days following the LH-RHa treatment. Remaining two fish had

oocytes at the early stage of vitellogenesis (1.3 mm in diameter) or oocytes degenerated to the last degree. In contrast, 5 out of 8 fish having high levels of vitellogenin (higher than 10.0 mg/ml) discharged over-ripened eggs. The sera of these 5 females formed precipitin lines with the specific antiserum to the vitellogenin which were similar to those of the egg extract in immunoelectrophoresis. Two out of the 8 fish had oocytes which diameter were 2.6-2.7 mm. Maturational stage of the ovary of the remaining 1 out of the 8 fish was not observed. Maturational stages of the ovaries of 2 fish having middle levels of vitellogenin (1.0-10.0 mg/ml) were not observed too. Sperm was obtained at 2 days after the treatments from 2 out of 4 males treated with HCG and 1 out of 4 males treated with the salmon pituitary extract. The other males did not spermiate. Artificial fertilization was done using eggs and sperm from the mature fish. F2 larvae developed from the eggs of the 2 females which had low serum vitellogenin levels.

Table 1 - Summary of artificial spawning in the sturgeon using cholesterol pellet of LH-RHa (1mg) in August 1988

Fish no.	Total length (cm)	Body weight (kg)	Vg level *1 (mg/ml)	Results	Remarks
1	117.6	9.65	27.9	OR*2	
2	121.2	9.88	27.6	OR	
3	114.3	9.06	14.9	Not spawned	OD*3 : 2.6 mm
4	119.4	9.95	14.0	Not spawned	OD : 2.7 mm
5	125.6	9.70	13.2	OR	
6	121.2	9.90	12.5	OR	
7	119.0	7.80	11.5	OR	
8	116.4	9.35	10.3	Not spawned	NO*4
9	131.0	13.72	5.5	Not spawned	NO
10	115.0	7.91	3.5	Not spawned	NO
11	129.5	12.96	1.0	Spawned	
12	123.0	9.88	0.5	Not spawned	Over-ripe
13	130.0	11.72	0.3	Not spawned	Immature = OD:1.3mm
14	120.0	9.60	0.2	Spawned	
15	122.0	9.20	0.1	Spawned	

*1 Concentration of vitellogenin (or reabsorbed egg yolk protein) in serum
*2 Discharged over-ripened eggs
*3 Oocyte diameter
*4 Maturity of ovary was not observed

DISCUSSION

Since 1985 artificial spawning of sturgeon has been studied in our institute but never succeeded. Failures of these experiments were estimated to be due to lack of confirming the maturational stage of the fish before hormonal treatment. Sturgeon of similar age show various stages of ovarian development when reared under identical conditions (Table 1, Fig. 4). Understanding the maturational stages of sturgeon is important for artificial spawning, since the timing of hormone treatment is critical. It was necessary to find some indicator of ovarian maturational stage. Serum vitellogenin levels are a useful predictor of ovarian development.

Vitellogenin is a yolk precursor protein that appears in the serum of females during vitellogenesis and is synthesized by the liver as a direct response to circulating estrogen (reviewed by Wiegand 1982, Wallace et al., 1987). Other works have clearly demonstrated that changes in serum vitellogenin levels correspond to development of

the gonad in several piscine species (Crim and Idler 1978, Ueda et al., 1984, Fukayama et al., 1986). However, there is no report describing those of sturgeon.

The present study documented changes in serum vitellogenin levels during the course of sexual maturation. Serum vitellogenin isn't detected in serum of female with oocytes under 0.8 mm in diameter by the Mancini method (K. Fujii, unpublished data). Vitellogenin levels increased concurrently with ovarian development and reached maximum levels during the final stages of vitellogenesis. A decrease in serum vitellogenin was observed at the end of vitellogenesis. Similar results have been observed in several piscine species (Scott and Sumpter 1983, So et al., 1985, Fukayama et al., 1986). A specific protein which appeared to be egg yolk protein was observed in the serum of female sturgeon with over-ripened eggs. It is difficult to distinguish the specific protein from vitellogenin by the Mancini method. In this study, sturgeon with over-ripened eggs were discriminated from females at vitellogenesis by the existence of a third precipitin line in immunoelectrophoresis using antiserum to vitellogenin. It seems to be released into the blood from over-ripened eggs. Many of over-ripe females showed high concentrations of vitellogenin. They might be higher than real concentrations due to the reabsorbed egg yolk protein which made the third precipitin line in immunoelectrophoresis. And vitellogenin or the reabsorbed egg yolk protein levels of fish with over-ripened eggs showed a wide range. These levels might indicate degrees of reabsorption and degeneration of eggs.

The results of serum vitellogenin levels were correlated to the success of artificial spawning of sturgeon using LH-RHa cholesterol pellets. Only females with low level vitellogenin at the end of vitellogenesis spawned fertilizable eggs. They will be distinguished from fish at the early vitellogenesis stage and over-ripe fish with similar levels of vitellogenin (or reabsorbed egg yolk protein) by quantitative analysis at regular intervals or qualitative analysis. These analysis of vitellogenin can be used to select fish for artificial spawning and to predict egg quality.

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EFFECTS OF VARIOUS HORMONE IMPLANTS ON VITELLOGENIN SYNTHESIS AND OVARIAN DEVELOPMENT IN CULTURED WHITE STURGEON, *Acipenser transmontanus*

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RESUME

En élevage la femelle de l'esturgeon blanc (*Acipenser transmontanus*) n'arrive pas à atteindre la maturité sexuelle vers l'âge de 8 ans, bien que le mâle l'atteigne vers l'âge de 3-4 ans.

Le développement sexuel, chez les femelles s'arrête au stage previtellogénique avec un développement folliculaire insuffisant pour initier la synthèse des oestrogènes. Nous avons trouvé qu'un traitement avec de l'estradiol exogène, peut induire la synthèse de la vitellogénèse chez ces poissons, mais même lorsque la concentration plasmatique de vitellogénine est élevée, le follicule n'est pas capable de l'incorporer. L'administration de l'hormone synthétique de libération de l'hormone luteinisante (LHRHa), seule ou combinée avec l'estradiol n'a pas d'effet sur l'incorporation de la vitellogénine par le follicule. Par ailleurs, l'administration d'extraits hypophysaires de carpe, en présence d'estradiol, n'a pas d'influence sur l'incorporation de la vitellogénine. Nous avons noté que le traitement combiné, estradiol et LHRHa, entraîne une augmentation significative de la concentration plasmatique d'oestrogénes. Il reste à déterminer, chez l'esturgeon femelle en élevage, si un traitement prolongé avec la LHRHa et l'estradiol peut induire une différenciation folliculaire suffisante permettant un développement sexuel plus avancé.

Mots clés : oestrogène, maturité sexuelle, vitellogénine, LHRH, *Acipenser transmontanus*.

ABSTRACT

Cultured female white sturgeon (*Acipenser transmontanus*) fail to reach sexual maturity be eight years of age even though cultured males reach sexual maturity by 3-4 years of age.

Sexual development in the cultured females is arrested at the previtellogenic stage with insufficient follicular development to initiate estrogen synthesis. We have found that exogenous estradiol treatment will induce vitellogenin synthesis in these fish, but even when the plasma concentrations of vitellogenin are elevated, the follicle is not capable of incorporating vitellogenin. Administration of synthetic gonadotropin releasing hormone (LHRHa) alone or in combination with estradiol did not induce uptake of vitellogenin by the follicle. Likewise, administration of carp pituitary extract in combination with estradiol did not influence vitellogenin uptake. We did note that the combined treatment of estradiol and LHRHa resulted in significantly higher circulating concentrations of estrogens than the administration of estradiol alone, suggesting that the combined treatment may be the result of induced estrogen synthesis. It remains to be determined if prolonged treatment with LHRHa and estradiol may induce follicular differentiation sufficient to support further sexual development in cultured female sturgeon.

Key-words : estrogen, sexual maturity, vitellogenin, LHRH, *Acipenser transmontanus*.

INTRODUCTION

At present, the rearing of white sturgeon (*Acipenser transmontanus*) by the aquaculture industry in the United States is dependent upon fingerlings produced entirely by the induced spawning of wild-caught broodstock. Cultured males that are raised from fingerlings reach sexual maturity by 3-4 years of age, but the cultured females do not exhibit sexual maturation by 8 years of age. The failure of these females to mature is characterized by their ovarian maturation being arrested at the

previtellogenic stage. Because the cultured females fail to reach sexual maturity, the industry is dependent upon wild-caught females as a sole source of eggs.

The limited availability of wild females makes such collections impractical for the long-term development of the industry, necessitating that methods be developed to induce sexual maturity in cultured females.

There may be several reasons why female sturgeon fail to reach sexual maturity in culture. One is that they may not be old enough. In the San Francisco Bay, white sturgeon females reach maturity around 15 to 20 years of age at a body weight of 20- 49 kg (Chapman, 1989). Under the favorable conditions of culture, females attained a body weight of 20-30 kg by 7-8 years of age, well within the range of body size of sexually mature wild females. Thus, the cultured females should be of sufficient size to sustain reproduction. Furthermore, in culture, males reach sexual maturity by four years of age, considerably earlier than for wild males whose estimated age of sexual maturity is 10 to 15 years (Chapman, 1989). These observations suggests that some factors other than age may be responsible for the failure of the cultured females to reach sexual maturity.

Because the survival of the sturgeon culture industry depends upon inducing the females to reach sexual maturity, we have begun a series of endocrine manipulations of females in order to induce sexual maturity. Since the sexual maturation of these cultured females is arrested at the previtellogenic stage, we have investigated whether the implantation of estrogen or estrogen in combination with an analog of gonadotropin-releasing hormone (LHRHa) or carp pituitary (CP) extract will induce vitellogenesis and ovarian maturation in female sturgeon.

MATERIALS AND METHODS

Four to seven year-old white sturgeon females (first generation of wild broodstock spawned at UC Davis hatchery in 1980-1983) were raised in 6.5 m diameter, 1 m deep outdoor freshwater tanks with an annual water temperature range of 10°C in the winter to 18°C in the summer. Fish were fed commercial trout broodstock diet (Murray Elevators, Utah), at a rate of 0.2-0.7% body weight/day, depending on water temperature and body size.

During the experimental period, selected females were moved to smaller (4.5 m diameter, 0.6 m deep), but otherwise identical tanks. Prior to sampling the fish were fasted for 48 h, with feeding commenced again 24 h after sampling. For sampling, animals were removed from the holding tank, anesthetized (MS-222 ; 80-100 ppm bath), and placed on a stretcher while freshwater was supplied orally. Five ml of blood were obtained by caudal puncture, using heparinized vacutainers. Small fragments (2 mm) of the ovarian tissue were removed surgically, through 1 cm abdominal incision. To obtain representative sample of gametocytes, fragments were removed from the medial portion of the ovigerous fold situated on the lateral side of sturgeon ovary.

Ovarian tissue was biopsied on day 0 and the final day of each experiment, while blood was sampled at 1 or 2-week intervals. Each animal was marked by a tattoo, permitting the correlation between changes in the ovarian conditions, plasma constituents and body weight of individual fish. After each experiment, which lasted approximately two months, experimental animals were returned to their respective age group cohorts for further rearing and all were sampled again 7-9 months later, during October 1988.

Serum samples were assayed for total estrogens using radioimmunoassay procedures (Parkin and Hendrickx, 1975) that utilized G.D. Niswender's estradiol antibody #244. The standard used was 17 -estradiol (Sigma Chemical Co.), and ^{3}H-estradiol (90-115 Ci/mmol, New England Nuclear) was used as the tracer. The minimum detectable estrogen level was 40 pg/ml. The interassay variability was 6.39% and the intraassay variability was 8.90%.

Plasma concentrations of vitellogenin were determined by the quantification of alkaline-labile protein phosphorus (ALPP). The procedure was similar to that described by Wallace and Jared (1968) and de Vlaming et al (1984). Plasma protein was

precipitated with 24% TCA, and phospholipids were removed by washing the pellet in 3 : 1 ethanol-ether mixture and then with 1 ml of ether. Protein-bound phosphate was liberated by resuspension of the pellet in 0.25 ml of 1N NaOH and placing the pellet into a dry heating block (102°C) for 15 min. Concentration of phosphorus was measured by color reaction with ammonium molybdate (Lowry and Lopez, 1946), using the autoanalyzer (660 nm, 96°C). Intra-assay variation was determined by measuring ALPP of a high µg plasma pool. Repeated freezing, thawing, and measurement produced insignificant changes in ALPP values (variation 3.1%). The reliability of the ALPP assay is based on the demonstration that plasma vitellogenin protein contains nearly all the ALPP present in serum (Wallace and Jared, 1968 ; Emmersen and Petersen, 1976 ; Hori et al 1979 ; Nath and Sundararaj, 1981). Validation of the ALPP method for the white sturgeon was accomplished by using gel filtration (Sepharose 6B-100), as described by Hara and Hirai (1978), Hamazaki et al (1987), and Norberg and Haux (1985) to isolate the plasma protein vitellogenic fraction (molecular weight approximately 360 KD) from the wild vitellogenic females, and estrogenized domestic females. Protein phosphate was determined in each plasma constituent in immature females and vitellogenic females. The vitellogenin protein purified by gel filtration and ion exchange chromatography contained >95% of the plasma phosphorus (Kroll and Doroshov, unpublished).

The ovarian development was examined by a light microscopy. To determine ovarian differentiation, biopsied fragments were preserved in 10% phosphate-buffered formalin, dehydrated and embedded in paraffin. Sections 4-5 µm were stained with hematoxylin and eosin, retic and periodic acid Schiff stains to establish differentiation of granulosa cells, secretion of glycoprotein zona radiata, and deposition of yolk. Sections were examined under compound scope (Leitz, Dialux) at magnification 630X, and photographed at magnification 390X.

Stage of the ovarian follicle was evaluated by score for the most advanced oocytes in each section. Presence or absence of capillary bed in the thecal layer, differentiation of the granulosa cell layer, the presence of the zona radiata, and yolk platelets in peripheral cytoplasm were recorded (1 or 0) and weighted by factors 1x, 2x, 3x, and 4x, respectively. Individual score for each fish was a sum of weighted points for each structure.

Experiment 1 - Estradiol Dose Response

This experiment was conducted from January to March (total of 56 days) at water temperatures of 10.5°-13°C. Twenty fish (4 and 5 years of age with a body weight range of 4.9-11.8 kg) were divided into 5 equal groups and implanted with cellulose pellets containing various doses of estradiol. The cellulose-estradiol pellets were prepared (Innovative Research of America, Toledo, Ohio) for the specific doses and water temperature ranges required for the experiment and were implanted intraperitoneally. The five groups were implanted with pellets to deliver 0,4,8.8,17.2 or 33.6 mg/kg of estradiol. Following implantation, each fish was bled weekly and the plasma was assayed for estrogens and vitellogenin. This bleeding frequency did not result in any detectable deterioration in the condition of the fish, but individual hematocrit values decreased from 35-40 to 20-27% by the end of the experiment.

Experiment 2 - Estradiol implants and vitellogenin production

This experiment was conducted for 60 days from June to August in a water temperature of 15.6-17.6°C. Twenty-four cultured females which were 4,5,6 and 7 years of age (3 of each age group/treatment) and with body weights ranging from 3.8 to 29.6 kg were divided into two treatment groups : one was implanted as described in the previous experiment with 5 mg/kg of estradiol in cellulose pellets while the second group (control) were implanted with cellulose pellets. The fish were bled at two-week intervals (the hematocrit remained stable) throughout the experiment.

Experiment 3 - Estrogen, LHRHa and CP

This experiment was conducted for 73 days from October to December in a water temperature of 11-14°C. A total of 20 females which were 4.5 and 5.5 years old (body weight 4.7-14.6 kg) were used, including 16 fish previously implanted with estradiol in Experiment 1. These 16 fish were reimplanted with 15 mg estrogen in cellulose pellets 7 days prior to the initiation of the experiment. Four additional females were implanted with cellulose only. On day 0, all estradiol implanted fish were implanted with either LHRHa ([D-Ala^6]-LHRH Ethylamide ; 2.5 mg/fish obtained from Sigma Chemical Co., St. Louis), or carp pituitaries. The low dose of carp pituitary extract (CPL) consisted of a dose of 1.2 mg/fish and the high dose of carp pituitary extract (CPH) was 120 mg/fish. The controls received only cellulose. One group of nonestrogen treated females was implanted with 2.5 mg of LHRHa. There were four fish in each treatment group. Fish were bled on days 0, 10, 20, 34 and 73 of the experiment.

RESULTS

In Experiment 1, all concentrations of estrogen implanted resulted in a significant elevation in the plasma concentration of estrogens and the induction of vitellogenin production (Figure 1).

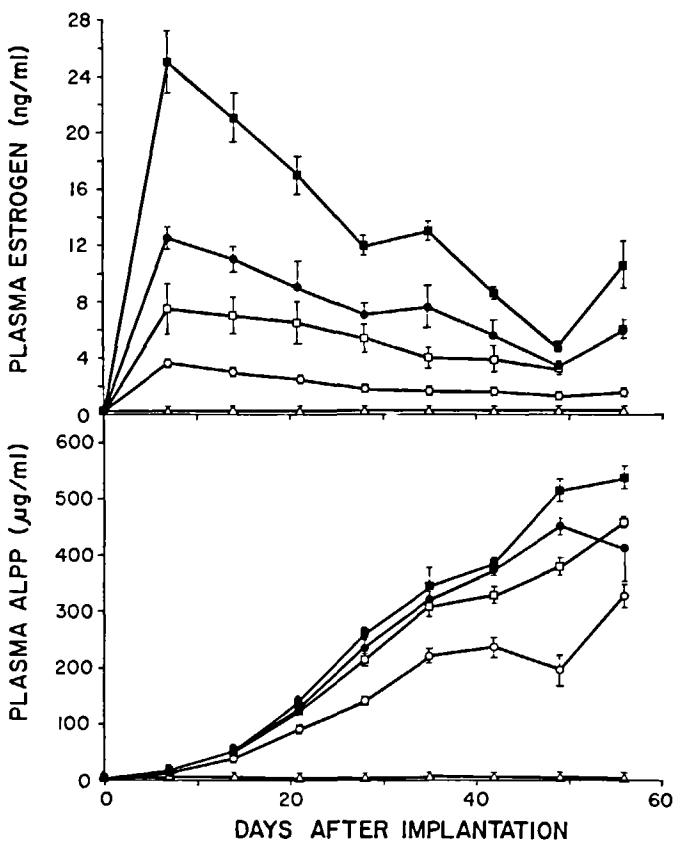


Figure 1 - The mean (+/- SEM) plasma concentrations of estrogens and ALPP in female sturgeon implanted with o 4, □ 8, ● 17.2, ■ 33.6 or ▵ 0 mg/kg of estradiol. (N=4 per group)

The plasma concentration of vitellogenin, as measured by ALPP, differed significantly ($P < 0.05$) between the lowest and highest E2 doses at all sampling points. By the end of the experimental period, ALPP concentrations of treated fish still remained at levels of 300-500 µg/ml, which is 6-10 times higher than mean ALPP values observed in vitellogenic wild females (Chapman et al., 1987). Throughout the experiment, the ALPP values of the untreated control fish remained at low basal concentrations (2-4 µg/ml).

The implantation of 5 mg/kg of estradiol in Experiment 2 resulted in a comparable increase in plasma estrogens in all age groups (Figure 2). Although the amount of estrogen in the plasma of the 1983 fish appear higher at some sampling times, these values are not significantly different from other age groups. The vitellogenin concentrations, as indicated by ALPP, showed a similar increase in all treated groups during the first 40 days of treatment reaching a high plateau of 300-500 µg/ml during the final 40-60 days of the experiment (Figure 2).

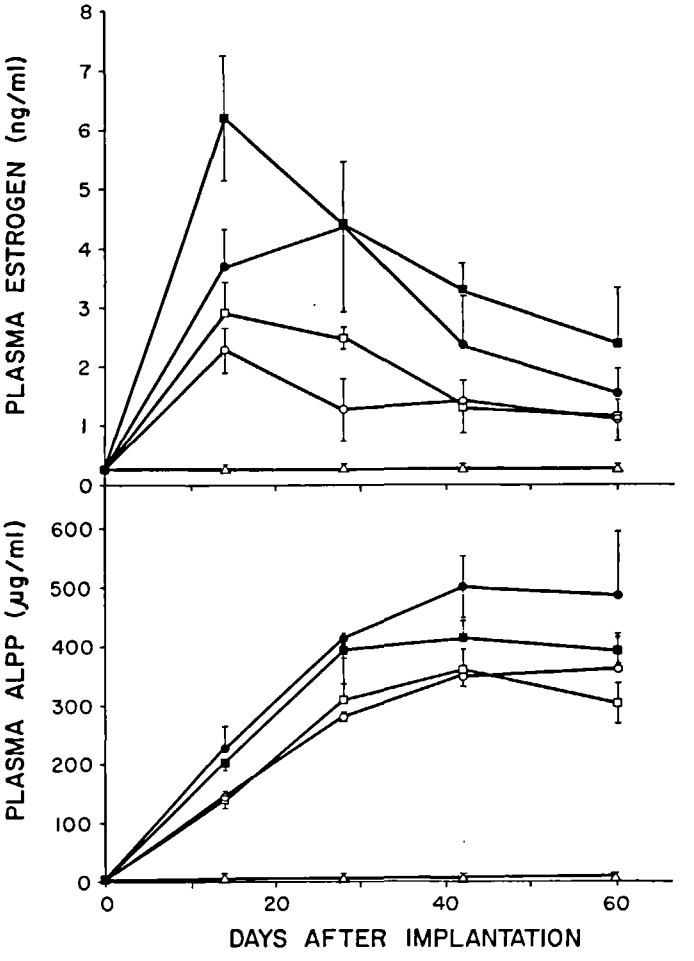


Figure 2 - The mean (+/-SEM) plasma concentrations of estrogens and ALPP of female sturgeon implanted with 5 mg/kg of estradiol. The fish were ■ 4, ● 5, □ 6, or ○ 7 years of age. The control groups (Δ) were implanted cellulose pellets. (N=3 for each age group/treatment)

Prior to the initiation of treatment in Experiment 3, the fish which had previously been implanted with estradiol still had slightly elevated concentrations of plasma estrogens as compared to the controls which had not been previously treated. Following implantation of estradiol, all of the treated animals experienced a further increase in the circulating concentrations of estrogen (Figure 3). Treatment with LHRHa alone did not result in an increase in circulating estrogen. The administration of estradiol along with LHRHa resulted in significantly ($P<.05$) higher plasma concentration of estrogen for the first 20 days following implantation than did treatments of either estradiol alone or in combination with CPL, but was not significantly different from the estradiol and CPH treatment. It is possible that LHRHa or the higher concentration of carp pituitary extract may have induced an increase of endogenous estrogen synthesis, however, LHRHa alone did not increase the amount of estrogen secreted and had no effect on the production of vitellogenin. The plasma concentration of ALPP was high from the previous estrogen treatment and remained elevated throughout the experiment in those fish which received estradiol treatment.

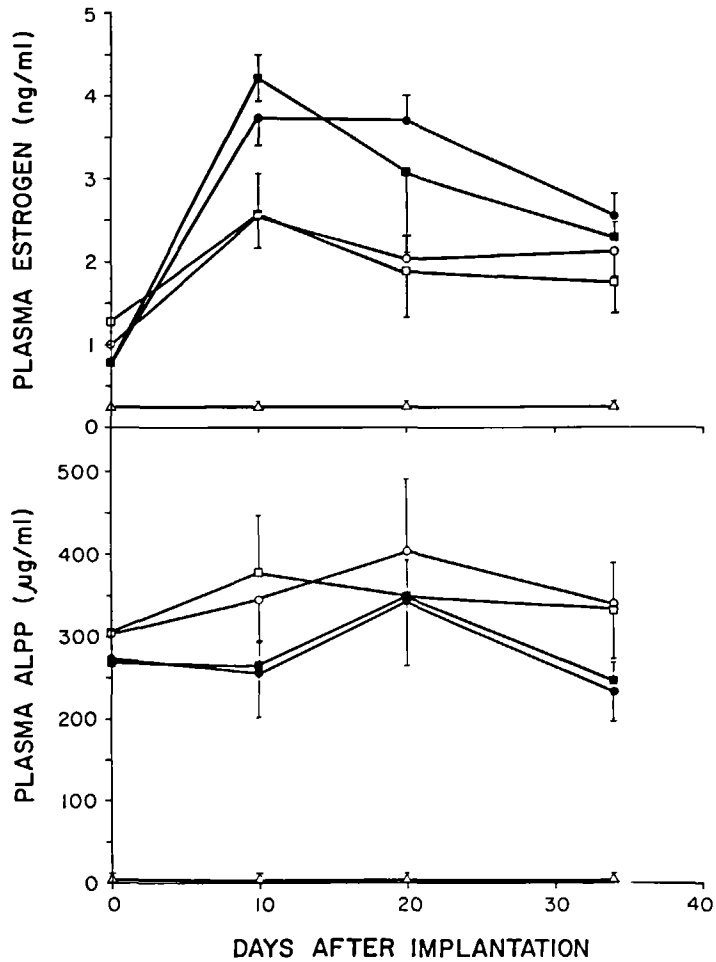


Figure 3 - The mean (+/- SEM) concentrations of estrogens and ALPP of female sturgeon implanted with □ estradiol, △ LHRHa, ■ estradiol + LHRHa, ○ estradiol + low dose of carp pituitary extract, or ● estradiol + high dose of carp pituitary extract. See text for details of treatments. (N=4 per group).

Seven to nine months after the completion of the experiments, plasma ALPP concentrations were measured in all of the experimental fish and were found to be still elevated in all estradiol-implanted fish (range of group means being - experiment 1 : 216-365 µg/ml ; experiment 2 : 119-227 µg/ml ; and experiment 3 : 72-139 µg/ml). The fish implanted with placebos or the non-estrogenized females implanted with LHRHa had low plasma concentrations of ALPP (2-5 µg/ml).

The ovarian stages were, in general, similar in all experimental animals on days 0. The oocytes were in the early growth phase, with basophilic cytoplasm and no structural yolk. Younger females (4 or 5 years old) exhibited substantial gonial cell proliferation and clusters of small, unfolliculated oocytes at the synaptic stage of meiosis. The follicular envelope in all fish was poorly differentiated : blood capillaries in the thecal layer and zona radiata were absent, and only a small number of undifferentiated granulosa cells was present (Figure 4,1).

The development of the ovarian follicle was not influenced by any treatment used, and no differences, detectable by light microscopy, were observed between day 0 and all treatment and control groups at the termination of each experiment. Estrogen-induced vitellogenin synthesis did not seem to influence ovarian differentiation and did not result in yolk deposition. There were no significant changes in the ovarian scores during experimental periods.

However, noticeable ovarian development did occur in some fish in 7-9 months after the cessation of experimentation (sampling in October 1988). Both estrogenized and non-estrogenized females of age groups 4 and 5 exhibited vascularization of thecal layer and proliferation of granulosa cells, although there was no histological evidence for secretion of zona radiata or presence of platelet yolk in cytoplasm (Figure 4,2).

Most significant changes were observed in three older females, age 6 and 7, that had been implanted with placebos in the Experiment 2. Two fish (age 7, body weights 24.1 and 27.8 kg) exhibited differentiated granulosa layer and thin secreted layer of zona radiata, which is clearly detected by PAS positive staining (Figure 4,3). One female (age 6, body weight 16.2 kg), in addition to differentiated follicular envelope, exhibited yolk microplatelets in the peripheral cytoplasm (Figure 4,4). Plasma ALPP concentrations in these three animals were 6, 7.5, and 8 µg/ml respectively, compared to range 2-5 µg/ml in all nonestrogenized females and range 4-275 µg/ml in estrogenized females. None of the animals implanted with estradiol in the Experiment 2 exhibited secretion of zona radiata and yolk deposition.

Mean ovarian scores observed in four age groups (Experiment 2) after 7-9 month additional rearing is shown in Figure 5. Estradiol treated and control fish were pooled since no statistically significant difference was observed, e.g. estrogen implants did not influence ovarian differentiation detectable at the light microscopy level. Pooled mean ovarian scores were low in the females from age groups 4 and 5, but significantly increased in the age groups 6 and 7, with body weight range 16-30 kg.

In all three experiments, animals ceased growth or lost body weight during respective experimental periods (Table 1). This was observed in all treatment groups, including control. This lack of growth may be attributed to the frequent sampling and possibly handling stress, rather than to hormonal treatment. After the 7-9 month rearing period following experimentation, all fish exhibited normal growth rates, ranging from 1.5 to 4.5% of body weight per month, which was similar to growth rates reported for the intact colonies of similar ages (Chapman, 1989). Difference in gains between the cohorts used in the experiments 1 and 3 (3.27-4.33%) and experiment 2 (1.5- 1.66%) is due to differences in age and body size. Hormonal treatments used in all three experiments did not affect growth (Table 1).

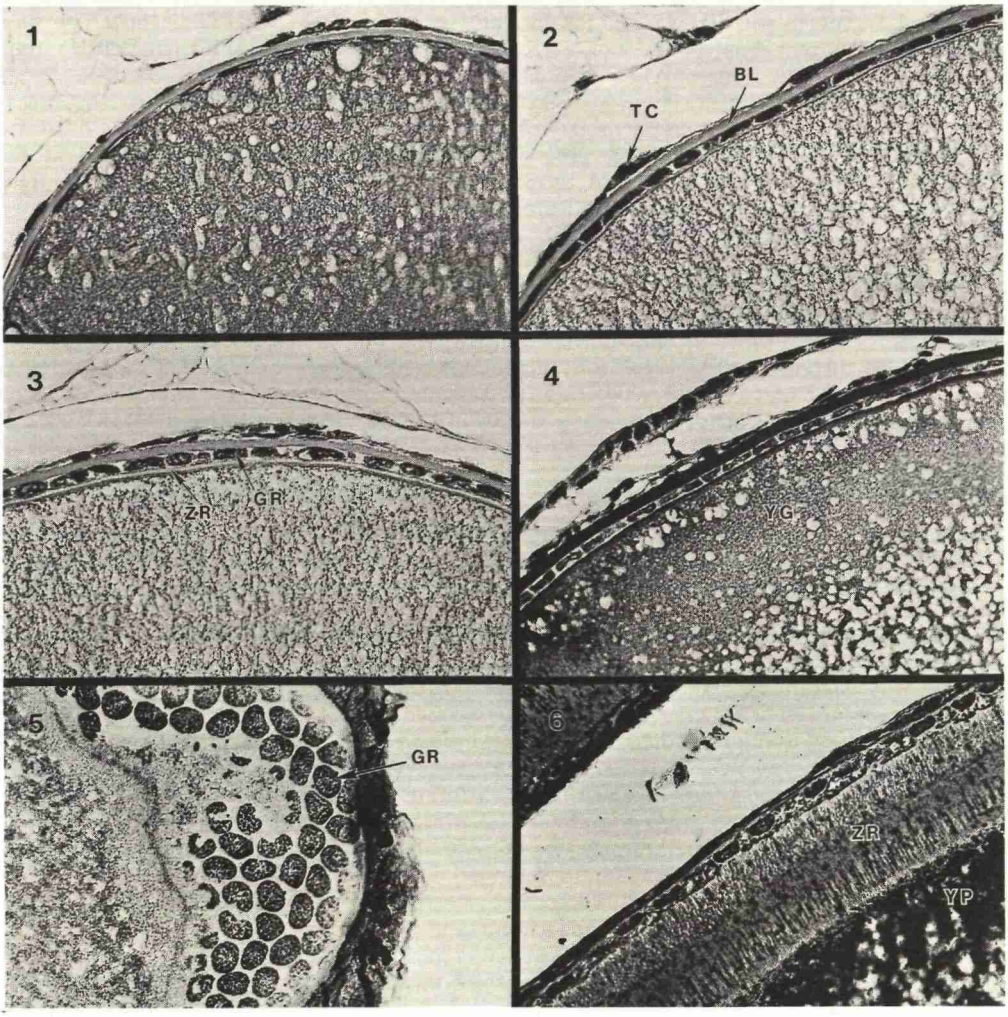


Figure 4 - Cortex of the ovarian follicles, magnification 390X. Ovarian biopsies were collected from experimental animals at the end of the experiment and in October 1988, 7- 9 months after experiments.
(1) estrogenized female age group 5, (Experiment 1), hematoxylin and eosin staining, egg diameter 300 µm
(2) estrogenized female, age group 5, (October 1988), retic stain, egg diameter 450 µm
(3) non- estrogenized female, age group 7, (October 1988), retic stain, egg diameter 550 µm
(4) non-estrogenized female, age group 6, (October 1988), PAS stain, egg diameter 600 µm
(5) granulosa cells, saggital section from the same fish
(6) follicular/egg envelope structure in a vitellogenic wild female, retic stain, egg diameter 1,300 µm (given for comparison).
Legends are : TC = thecal cells, BL = basal lamina, ZR = zona radiata, GR = granulosa cells, YG = yolk granules, YP = yolk platelets.

Table 1 - Body weight (kg, mean ± SEM) of sturgeon during the experimental and postexperimental periods. Gains or losses (percent per month) were all insignificant during experimental periods and significant for postexperimental periods (paired t-test, $p < 0.05$). See text for details of treatments.

Experiment 1		*Experimental Period*			*Postexperiment (228 days)*	
Treatment	N	Before	After	Gain	Weight	Gain
E2	16	7.8 ± 0.4	7.6 ± 0.4	-0.98	10.1 ± 0.6	4.33
Control	4	10.2 ± 0.6	9.4 ± 0.5	-4.16	12.3 ± 0.7	4.03

Experiment 2		*Experimental Period*			*Postexperiment (281 days)*	
Treatment	N	Before	After	Gain	Weight	Gain
E	12	13.5 ± 2.1	13.7 ± 2.2	0.83	15.6 ± 2.3	1.50
Control	12	13.7 ± 1.9	13.8 ± 1.8	0.59	16.0 ± 2.0	1.66

Experiment 3		*Experimental Period*			*Postexperiment (291 days)*	
Treatment	N	Before	After	Gain	Weight	Gain
E2	4	9.5 ± 0.7	9.3 ± 0.8	-1.01	12.2 ± 1.1	3.27
LHRHa	4	12.3 ± 0.7	11.4 ± 0.7	-2.89	16.0 ± 1.3	4.17
E2 + LHRH	4	10.4 ± 0.8	10.1 ± 0.8	-1.40	13.7 ± 0.9	3.76
E2 + CP	8	10.2 ± 1.0	9.8 ± 0.9	-1.75	13.0 ± 1.0	3.43

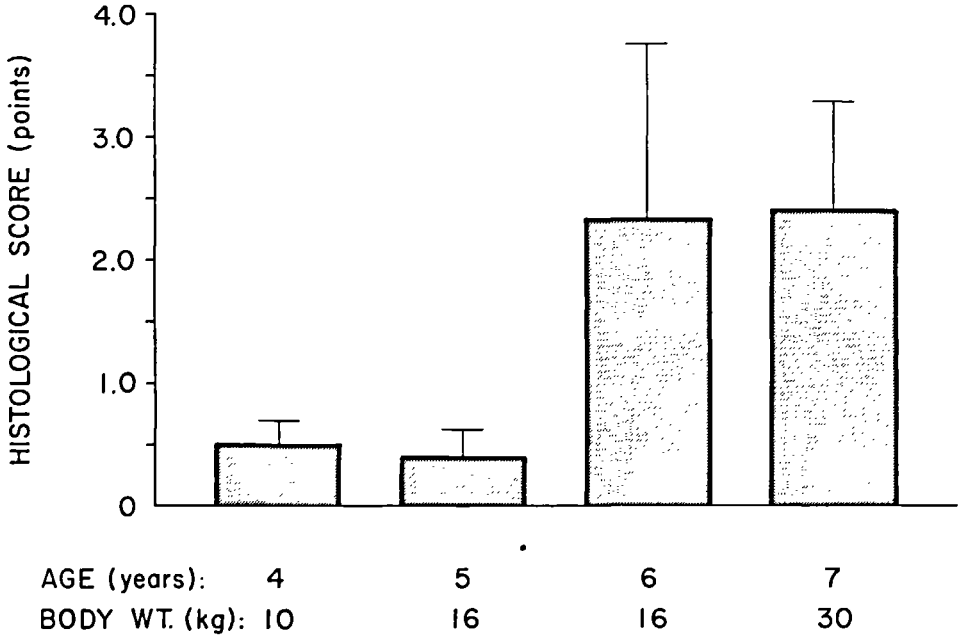


Figure 5 - Histological ovarian scores of females from Experiment 2, which were sampled in October 1988. Means (+/-SEM) for pooled estradiol treated and control age groups (N=8 per group). Mean body weight for each group is to be nearest kg.

DISCUSSION

These results indicate that the inability of the cultured female sturgeon used in this study to reach sexual maturity may have resulted from the failure of the hypothalamic-pituitary-gonadal axis (HPG) to secrete sufficient gonadotropins to stimulate the development of the reproductive system beyond the previtellogenic state. As a result, the follicular cells did not synthesize sufficient estrogen to stimulate vitellogenin production and the follicle did not develop sufficiently to permit the uptake of vitellogenin even when it was present in the plasma in high concentrations.

Certainly the cultured females were capable of synthesizing vitellogenin since all of the fish treated with exogenous estradiol responded with a marked production of vitellogenin. This is the first direct experimental evidence that vitellogenin synthesis in the female sturgeon is controlled by estrogen. However, even when vitellogenin was present in the blood in large quantities, the oocytes were incapable of incorporating the vitellogenin, suggesting that the primary problem of reproductive failure in these sturgeon is not their failure to synthesize estrogen.

If the cessation of reproductive development is the result of insufficient gonadotropin secretion, then one possible approach to solving the problem would be the administration of exogenous LHRH. Such treatment might stimulate the pituitary to synthesize and secrete sufficient gonadotropins to initiate the next stage of gonadal development. However, in this study, treatment with LHRHa alone did not have any effect on estrogen synthesis even though a comparable LHRHa treatment was effective in inducing spawning in sturgeon (Lutes et al, 1987). Since a gonadotropin assay for sturgeon was unavailable at the time of this study, it was not possible to confirm that the treatment used had any effect on the secretion of gonadotropins.

Treatment with estradiol and LHRHa combined did result in significantly higher plasma concentrations of estrogens than did either estradiol or LHRHa alone, suggesting that this combined treatment may have stimulated an increased secretion of endogenous gonadotropins resulting in estrogen synthesis. In immature salmonids, Crim et al (1981) found that treatment with exogenous estrogen resulted in a significant increase in the pituitary content of gonadotropins. If the estradiol treatment had a similar effect in our study, then the additon of LHRHa may have stimulated the secretion of the accumulated gonadotropins, which in turn stimulated estrogen synthesis. Nevertheless, the combined estrogen and LHRHa treatment still did not initiate sufficient follicular differentiation (granulosa cells) to permit the uptake of vitellogenin. It is possible if the combined treatment of estradiol and LHRHa had been extended for a longer time period, further follicular development might have been initiated. Such follicular development was observed during the postexperimental period, but occurred in both control and treated groups.

Based on histological observations, it may appear that captive white sturgeon female is incompetent to initiate normal vitellogenesis before reaching certain age and body size. However, there is no information at present time to suggest a certain specific age in cultured white sturgeon female, as a major determinant of vitellogenesis. Interactive effect of age, seasonality and endogenous factors (endocrine and metabolic) on the onset of vitellogenesis in sturgeon must be elucidated, and knowledge of this effect will be helpful in estalbishing appropriate hormonal treatment to accelerate sexual maturity.

In conclusion, the cultured females appear to delay sexual maturity because the hypothalamic-pituitary-gonadal axis does not secrete sufficient gonadotropins to initiate the next stage of reproductive development. It is unclear why this axis in the female fails to mature in culture while cultured males reach full sexual maturity at an age much younger than in the wild. It would appear that endocrine manipulation of the reproductive axis should induce sexual maturity, but at this time we have not been able to identify the appropriate hormone therapy.

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GROWTH OF HATCHERY PRODUCED JUVENILES OF ITALIAN STURGEON,*Acipenser naccarii* BONAPARTE,REARED INTENSIVELY IN FRESH WATER.

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RESUME

Un stock de 5800 larves a été élevé de facon intensive durant 8 mois en eau douce à 15°C avec une nourriture artificielle. A la fin du période la survie a été du 78% et le poids moyen est passé de 12.5 mg à 108 g, avec une longueur correspondante de 10 à 317 mm. La biomasse finale a été de 489 kg, avec un coefficient de croissance istantanée de 1.8 % dans les derniers 190 jours d'elevage. Le facteur de conversion de l'aliment pendant la meme période est de 1.2.
Ces résultats préliminaires montrent la possibilité de l'élevage de l'esturgeon italien non seulement pour des programmes de repeuplement, mais aussi pour une production commerciale.

Mots clés : *Acipenser naccarii*, larves, juvéniles, alimentation, croissance, survie.

ABSTRACT

A lot of 5800 hatchery produced larvae of the Italian sturgeon *Acipenser naccarii*, were raised intensively in fresh water on an inert diet at 15°C for 8 months. At the end of this period, fish survival was 78% with an increase in body weight from 12.5 mg to 108 g, and in total length from 10 to 317 mm. The istantaneous growth rate was 1.8% in the last 190 days of culture and the final biomass gain 489 kg, with a food conversion factor of 1.2.
These preliminary results suggest that the Italian sturgeon could be bred not only for restocking and mitigation programs, but also for commercial production.

Key-words : *Acipenser naccarii*, larvae, juveniles, alimentation, growth, survival.

INTRODUCTION

The Italian sturgeon, *Acipenser naccarii* Bonaparte (1836), is an amphibiotic, anadromous species which is endemic to the Adriatic sea and the middle and downstream course of the Po river and its main tributaries, where it lives sympatrically with the common sturgeon, *Acipenser sturio* L..

In recent years, the stability of the wild population of *A.naccarii* has been threatened by the construction of fluvial dams and barriers which prevent the migrating adults from reaching the spawning grounds upstream. As a consequence, the number of catches per year by sport and professional fishing has consistently declined.

To help preserving this species in its native environment, on June 1988, we have bred artificially a broodstock of sturgeon grown in captivity for 11 years in a fish farm at Orzinuovi, near Brescia, in order to produce juveniles for restocking operations (Arlati et al., 1988).

This note reports the results of a weaning and growing out experiment on a selected group of that offspring from the larval stage to the 8th month of life (July 1988 - March 1989).

MATERIALS AND METHODS

We have utilized 5800 larvae, part of a batch of eggs stripped from a single female (20 kg bw) injected, 8 h apart, with 10 and 25 µg/kg of the synthetic LH-RH analogue, des Gly 10[D-Ala6]-LH-RH ethylamide, which were fertilized with the milt of a male (13 kg bw) treated once with 10 µg/kg LH-RHa. Hatching occurred 8-10 days after fertilization and yolk resorption after further 10-12 days (Arlati et al.,1988).

On July 1988, all the larvae were moved into a 450-l fiberglass tank (300 x 75 x 25 cm) supplied with 2 l/sec of fresh water at a constant temperature of 15°C with 90-92 % oxygen saturation, flowing from a 112 m-deep artesian well.

As fish grew, they were equally subdivided into more tanks, with identical characteristics to avoid overcrowding (Tab.1). After 8 months, they were transferred into concrete tanks where they are still growing.

Table 1- Stocking density during the rearing period.

Months of rearing	Mean weight (g)	Number of fish	Number of tanks	Stocking density	
				Initial (kg/m^3)	Final (kg/m^3)
0	0.0125	5800	1	0.16	1.16
1	0.101	5148	2	0.58	3.61
2	0.7	4646	4	1.81	8.82
3	3.5	4537	6	5.88	13.26
4	7.9	4532	12	6.63	29.70
6	35.5	4518	20	17.82	30.12

Larvae were immediately weaned on an inert diet based on Larvit (Hendrix) supplemented with egg yolk, curdled milk and vitamins, which was administered for 15 days as moist thin strings. Thereafter, fish were fed 5-7 times during daytime and once at midnight on semi-moist pellets composed of 75% commercial sturgeon feed (Storioni 1, Hendrix), 32% brown trout starter (Microgranulina, Arc-en-ciel, Chezzi), 2% fresh fish and 1% vitamin mixture.

To monitor growth, the individual total length of 20 fish and the average weight of two samples of 100 sturgeons each were measured monthly during the first 5 months of culture. Afterwards monthly measurements of total length and weight were performed individually on groups of 30 fish.

The following expression was used to calculate the istantaneous growth rate (G) for the last 190 days of culture during which body weight increased exponentially (regression coefficient = 0.993 on semi-logarithmic plot):

$$G = (\ln Wf - \ln Wi) / T \times 100$$

where: Wi = mean initial body weight (g)
Wf = mean final body weight (g)
T = period of growth (days)

Overall conversion efficency (C) was calculated using the formula:

$$C = 1/F \times 100$$

where: F = food conversion factor = food supplied/biomass gain

The same prophylactic criteria were adopted for sturgeon rearing as in trout farming.

Sturgeons adapted well to life in tanks since mortality was low and limited to the first two months of culture (Fig.1), and 78% of them survived till the end of the observation period.

Growth of fish was continuous and very fast (Fig.2) as their average body weight increased from 12.5 mg to 108 g (W /W = 8670) in about 8 months (246 days), thus doubling 13-fold, with an istantaneous growth rate of 1.8% in the last 190 days of culture.

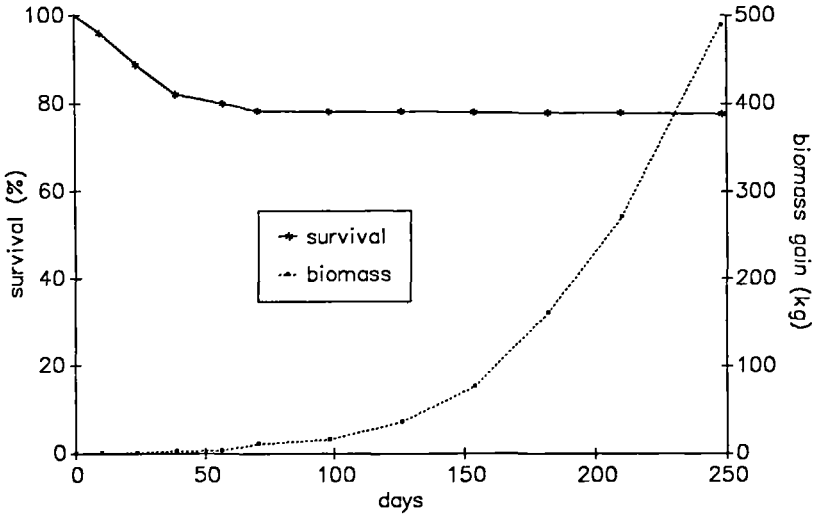


Figure 1- Survival (%) and biomass gain (kg) of reared juveniles of *A.naccarii*.

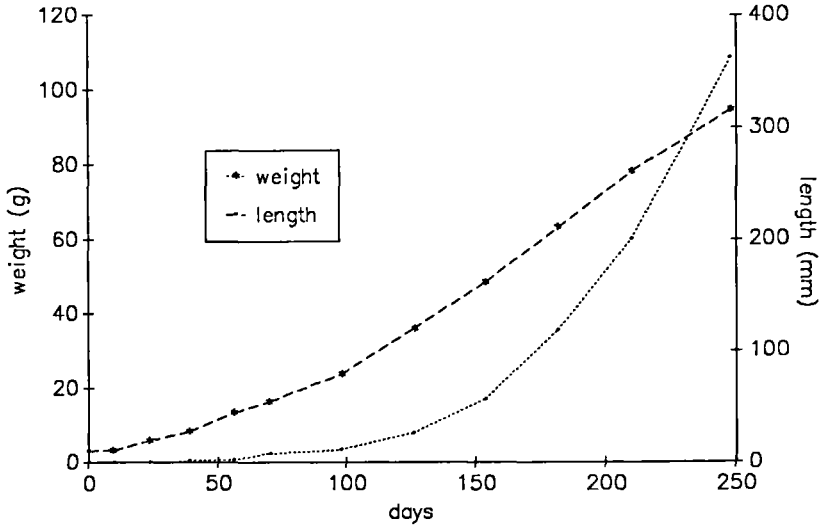


Figure 2- Growth of juveniles of *A.naccarii* expressed as mean weight (g) and length (mm).

The average total length increased from 10 mm to 317 mm (Fig.2), and in the last 4 months of culture it was linearly correlated (r =0.993) with the logarithm of body weight.

The final biomass gain amounted to 489 kg (Fig.1) and since it was obtained with 580 kg of feed (dry weight), the food conversion factor was 1.2 and the overall conversion efficiency 83.3.

These figures compare favorably with those reported for *A. baeri*, which can grow to 300 g at 16-18°C with F = 1.7 (Williot, 1986), and *A. transmontanus* which was found to reach 280 g in 10 months (Monaco et al., 1981). The performance in culture of *A. naccarii* is particularly promising because it was obtained at a constant temperature of 15°C, which is below the thermal preferendum of the other two species.

These preliminary results suggest the opportunity to consider the breeding of the Italian sturgeon not only for restocking and mitigation programs, but also for the commercial production.

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ALIMENTATION ARTIFICIELLE DES LARVES DE L'ESTURGEON SIBERIEN (*Acipenser baeri Brandt*)

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RESUME

Des larves d'esturgeon sibérien (*Acipenser baeri* Brandt) ont été élevées à 17 °C pendant 47 jours en utilisant exclusivement des aliments artificiels (2 aliments expérimentaux R1 et R2 à base de foie et de levure pendant les 29 premiers jours, puis un aliment du commerce pour alevin de truite). Les densités initiales d'élevage étaient comprises entre 33 et 66 larves.l^{-1}. A la fin de la 1ère période, la survie moyenne a été selon les régimes de 73% et 68% et le poids vif moyen de 392 et 379 mg. L'indice de "distribution" moyen (rapport quantité d'aliment distribuée/gain de poids) a été de 2 et le taux de croissance spécifique de 10%.j^{-1}. A partir du huitième jour, un comportement de cannibalisme a été noté, se traduisant par des morts mutilés et non par une disparition d'animaux. Entre le seizième et le vingt-troisième jour, les animaux mutilés représentaient environ 50% des morts. Pendant la 2ème période, la survie moyenne a été de 99% et le poids moyen final de 1,5 g avec un indice de "distribution" de 1,2 et un taux de croissance spécifique de 8%j^{-1}. Des modifications importantes de la morphologie des larves ont été mises en évidence par le calcul du coefficient de condition K = 100 P.L^{-3} et du coefficient de régression b de la relation P = a.L^{b}. Entre J_{22} et J_{47}, K et b diminuent respectivement de 0,95 à 0,52 et de 3,27 à 2,66. Ces résultats obtenus avec des densités initiales élevées (33 à 66 larves.l^{-1}) montrent que les larves d'*Acipenser baeri* se prêtent à des conditions d'élevage intensif. Des expériences complémentaires en plus grand volume seraient cependant nécessaires.

Mots clés : *Acipenser baeri*, larve, alimentation artificielle, survie, croissance, coefficient de condition, indice de consommation

ABSTRACT

Siberian sturgeon (*Acipenser baeri* Brandt) larvae were reared during 47 days using artificial food only : 2 experimental diets R1 and R2 based upon beef liver and yeast for the 29 first days, and after those a commercial diet for trout fry. Initial stocking rates ranged between 33 and 66 larvae.l^{-1}. At the end of the first period, mean survival, according to the diets, was respectively 73% and 68%, and mean live weight 392 and 379 mg. The ratio delivered food/weight gain was 2 and the specific growth rate 10%.day^{-1}. From day 8, attempts of cannibalism were observed, resulting in damaged dead larvae without fish disappearance. Between day 16 and day 23, damaged fish constituted about 50% of the dead ones. During the second period, survival was 99% and final mean weight was 1.5 g. The ratio delivered food/weight gain was 1.2 and the specific growth rate 8%.day^{-1}. The condition factor K = 100 W.L^{-3} and the regression coefficient b of the relationship W = a.L^{b} were used for the determination of morphological changes. K and b both decreased between D_{22} and D_{47} from 0.95 to 0.52 and from 3.27 to 2.66, respectively. These results obtained with high initial stocking rates (33-66 larvae.l^{-1}) show that *Acipenser baeri* larvae can be reared under intensive conditions, but they should be verified in larger tanks.

Key-words : *Acipenser baeri*, larvae, artificial feeding, survival, growth, condition coefficient, feed gain rate

INTRODUCTION

Les méthodes traditionnelles utilisées pour la production d'alevins d'esturgeons sont l'élevage en étangs fertilisés et l'élevage en bassins avec distribution de nourriture vivante (*Enchytreus albidus, Daphnia* sp., *Moina* sp. ou *Artemia salina*). Ces techniques sont pratiquées à grande échelle en Union Soviétique : depuis 1975, de 100 à 140 millions d'alevins (*Acipenser gueldenstaedti, Acipenser stellatus* et *Huso huso*) sont relâchés chaque année à des fins de repeuplement (Barannikova, 1987).

Des méthodes plus récentes consistent à remplacer les proies vivantes par des aliments artificiels (Semenkova, 1983; Buddington and Doroshov, 1984; Doroshov, 1985; Dabrowski et al., 1985).

Dans le présent travail, on a étudié les possibilités d'adaptation des larves d'*Acipenser baeri* à des conditions d'élevage intensif, avec une alimentation exclusivement artificielle, à des densités initiales d'élevage plus élevées que celles utilisées par Dabrowski et al. (1985) chez la même espèce : on a cherché à déterminer l'importance éventuelle du cannibalisme comme facteur de mortalité et l'influence de la quantité d'aliment distribuée sur la survie et la croissance des larves ; on a enfin précisé l'influence de l'alimentation et de l'âge sur le coefficient de condition et la relation longueur-poids.

La grande plasticité et le potentiel de croissance important d'*Acipenser baeri* ont déja été signalés par Mil'shtejn (1972). L'intérêt de cette espèce pour l'aquaculture a été confirmé plus récemment par Barrucand et al.(1979) et Williot et al.(1988).

MATERIEL ET METHODE

L'expérience a commencé le 11/04/84 (J_0) sur des larves écloses 4 jours plus tôt et venant de l'écloserie du CEMAGREF (Pisciculture INRA, Donzacq, Landes) ; elle s'est déroulée en 2 parties :

1ère partie (J_0 - J_{29})

1750 larves ont été réparties en 10 lots et placées dans un système d'élevage dont le principe général a été décrit par Charlon et Bergot (1984).

Deux séries de 5 bacs ont été utilisées ; chaque série comportait 2 bacs carrés de 2,6 l, 2 bacs rectangulaires de 5,3 l et 2 bacs ronds de 5,2 l présentant une surface du fond de respectivement 325, 475 et 491 cm^2. Le débit d'eau dans chaque bac était de 0,5 $l.min^{-1}$ correspondant à des temps moyens de renouvellement compris entre 11 et 5 minutes.

Avec une même charge initiale de 175 larves par bac, les 3 types de bacs testés correspondaient à des densités de 33 à 66 $larves.l^{-1}$.

Ces bacs étaient intégrés dans un circuit semi-fermé thermo-régulé à 17 °C représentant un volume total de 1500 l, avec un apport d'eau neuve pour l'ensemble du système de 1 $l.min^{-1}$ environ.

Deux aliments expérimentaux R1 et R2 ont été testés, la distribution étant assurée dès le premier jour par une série de distributeurs rotatifs programmables (Charlon et Bergot, 1986) placés sur chaque bac et fonctionnant pendant la période d'éclairage.

Le motoréducteur (1 $tour.min^{-1}$) qui assurait la rotation de l'axe supportant les distributeurs était couplé à un programmateur à cames utilisé en doseur cyclique (durée du cycle : 10 min) dont le réglage assurait 7 distributions d'aliment par cycle.

De J_0 à J_8 l'aliment a été distribué de 7h à 19h, et de J_8 à J_{29} de 5h à 22h. Pour réaliser des conditions d'alimentation en excès, le nombre de repas a été progressivement augmenté (240 repas par jour de J_0 à J_6, 320 de J_6 à J_8 et 453 de J_8 à J_{29}) ainsi que la quantité distribuée par jour et par bac (de l'ordre de 0,7 g la première semaine et 4,2 g à la fin de la quatrième semaine).

Les régimes R1 et R2 avaient pour base un mélange de levure (IFP pour R1 et Protibel pour R2) et de foie de boeuf. Leur formulation correspondait à celle des régimes A et D donnée par Bergot et al. (1986) ; leur composition chimique a été détaillée dans le travail de Kaushik et al. (1986).

Ces 2 types d'aliment ont été tamisés à :

100 - 200 µm pour la période J_0 - J_7
200 - 400 µm pour la période J_7 - J_{16}
400 - 630 µm pour la période J_{16} - J_{29}

Pour chaque granulométrie et chaque bac, les quantités d'aliment distribuées ont été pesées.

Tous les jours les morts ont été comptabilisés avec observation des phénomènes de cannibalisme (identification des morts mutilés). Pour chaque régime un échantillon de 20 poissons a été prélevé à J_0, J_8, J_{15}, J_{22} et J_{29} et conservé plusieurs mois dans du formol à 10% pour les mesures de poids. Les survies ont été calculées en tenant compte de ces prélèvements (Bergot et al., 1986).

A J_{22} et J_{29}, les poissons de chaque bac ont été anesthésiés au phénoxy-2-éthanol, pesés globalement, comptés et remis en élevage ; en outre, des mesures individuelles de longueur et de poids frais ont été faites à J_{22} et J_{29} sur des échantillons de 20 poissons.

2ème partie (J_{30} - J_{47})

Les 5 lots élevés sur R1 ont été regroupés de même que ceux élevés sur R2. Ces 2 nouveaux lots R'1 et R'2 ont été placés chacun dans un double bac circulaire de 300 l. Un troisième bac permettait d'effectuer un transvasement quotidien des poissons et un nettoyage complet des bacs par rotation des 2 lots.

L'aliment utilisé était un aliment du commerce pour alevin de truite (Aqualim Fario, France), tamisé à 400 - 630 µm pour la période J_{30}-J_{36} et >800 µm pour la période J_{36}-J_{47}. Il a été distribué pendant la période d'éclairage (5h - 23h) au rythme de 480 repas j^{-1}.

Tous les jours les morts ont été comptabilisés. En fin d'expérience au jour J_{47}, tous les animaux des lots R'1 et R'2 ont été anesthésiés et pesés individuellement ; des mesures de longueur et de poids frais ont été réalisées sur des échantillons prélevés dans ces 2 lots.

Le test statistique utilisé pour comparer les survies (S) est une analyse de variance à 2 voies après transformation angulaire $y = \arcsin(S^{1/2})$. Le même test a été appliqué aux poids (P) après transformation $z = \log P$.

RESULTATS

L'aliment a été distribué dès le premier jour d'expérimentation alors que les larves présentaient encore un bouchon de mélanine au niveau de la valvule spirale. Lors du nettoyage quotidien, la présence de ces bouchons après expulsion a été notée entre J_4 et J_{10}.

La répartition des effectifs contrôlés au cours de l'expérience est présentée dans le tableau 1.

Les survies entre J_0 et J_{29} ont été calculées pour chaque bac (fig. 1) ; la survie moyenne pour l'ensemble des lots a été de 70% à J_{29}. La survie au cours de la 2ème période a été de 99%.

L'analyse à 2 voies des survies à J_{29} ne montre pas d'effet significatif du régime ($p = 0,19$), du type de bac ($p = 0,12$) ni d'interaction ($p = 0,28$). Pour les poids moyens à J_{29}, les différences observées entre traitements ne sont pas non plus significatives (régime : $p = 0,80$; type de bac : $p = 0,19$; interaction $p = 0,58$).

A partir du huitième jour d'expérience, des manifestations de cannibalisme ont été notées : des larves capturaient d'autres larves par la queue ou les barbillons ; les individus mutilés (avec en particulier la nageoire caudale tronquée) ne tardaient pas à mourir. Le nombre total de morts ainsi mutilés est donné dans le tableau 1.

Tableau 1 - Dénombrement des individus et survie au cours des 2 périodes J_0-J_{29} et J_{30}-J_{47}

Période J_0 - J_{29}

REGIME	R1	R2	PME_i*
Effectif initial	875	875	100%
Morts enregistrés	237	281	29,6%
dont mutilés	61	73	7,7%
Prélevés vivants	85	87	9,8%
Disparus	14	5	1,1%
Survie à J_{29}	73%	68%	70%

Période J_{30} - J_{47}

REGIME	Aliment du commerce (R'1)	(R'2)	PME_i*
Effectif initial	539	502	100%
Morts enregistrés	5	7	1,2%
dont mutilés	2	0	0,2%
Survie à J_{47}	99%	99%	99%

* PME_i : pourcentage moyen de l'effectif initial

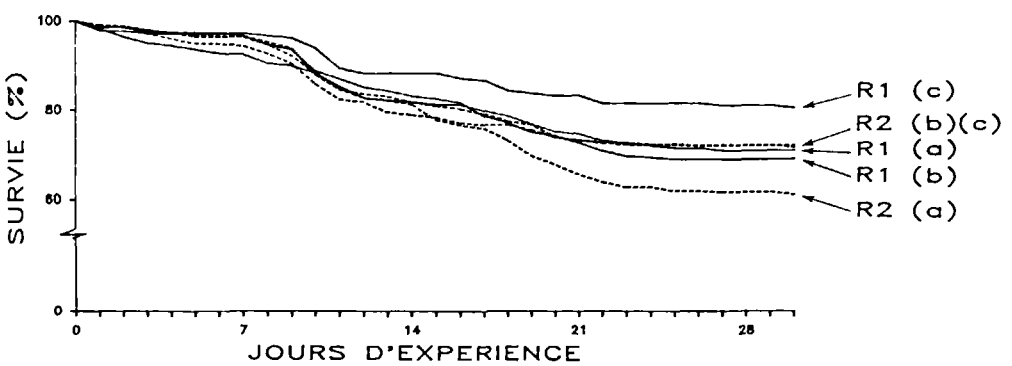


Figure 1 - Survie des 10 lots de larves élevées sur R1 (ááá) et R2 (-----) en bacs rectangulaires (a), carrés (b) ou ronds (c).

La figure 2 donne le nombre de morts mutilés et le nombre total de morts enregistrés pendant la période J_0 - J_{29}. A J_{12} et entre J_{16} et J_{23}, le nombre d'animaux mutilés représente chaque jour environ la moitié du nombre total de morts.

Les observations de cannibalisme sont nombreuses surtout dans les lots ayant de petits individus ; en effet, les nombres de morts mutilés, enregistrés pour les périodes J_0-J_{22} et J_0-J_{29}, sont corrélés négativement ($p < 0,01$) avec les poids moyens frais mesurés respectivement aux jours J_{22} et J_{29} (fig. 3).

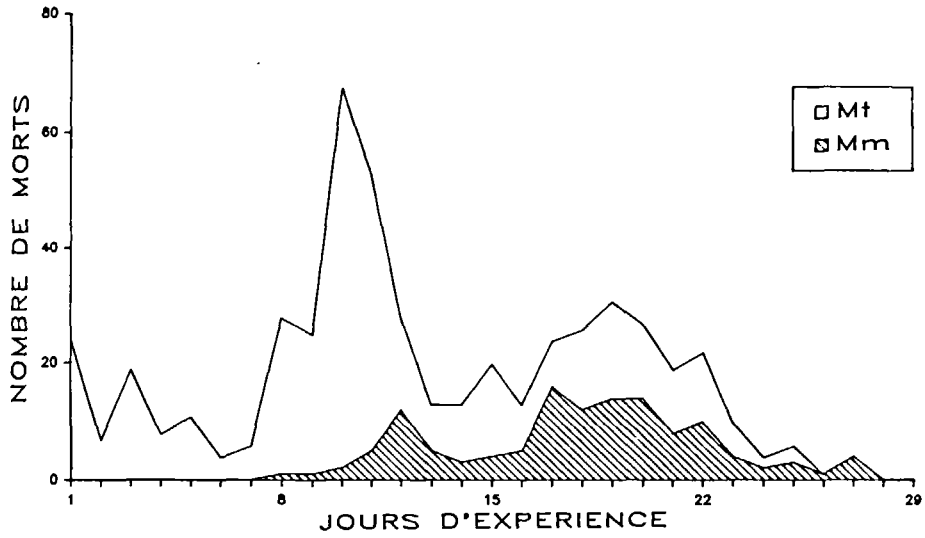


Figure 2 - Nombre total de morts (Mt) et nombre de morts mutilés (Mm) enregistrés quotidiennement pendant la période J_0-J_{29}

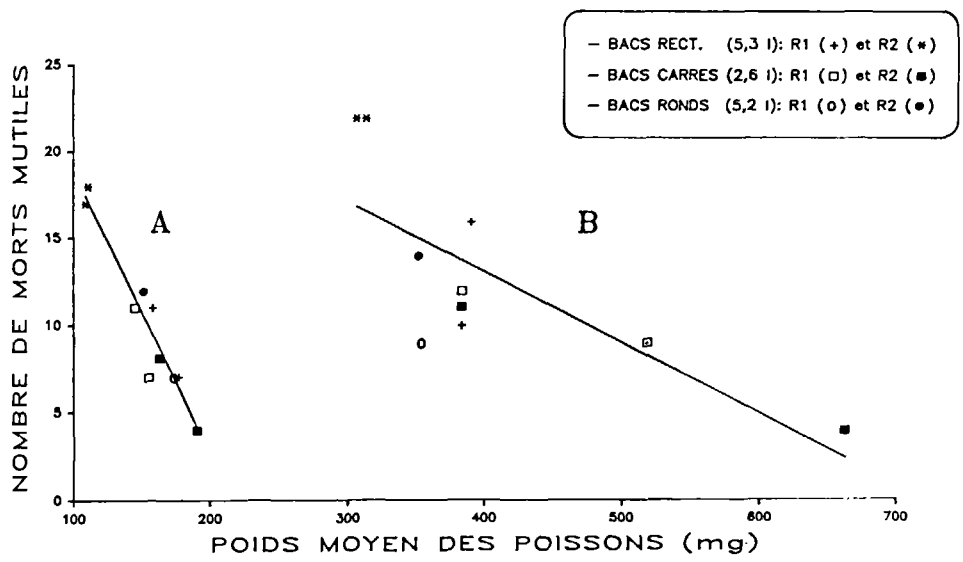


Figure 3 - Relation entre le nombre total de morts mutilés (manifestation de cannibalisme) et le poids frais moyen des poissons dans les différents bacs
A : nombre de morts entre J_0 et J_{22} par rapport au poids moyen des lots à J_{22}
B : nombre de morts entre J_0 et J_{29} par rapport au poids moyen des lots à J_{29}

La relation entre la survie et la quantité d'aliment distribuée pour une même période (J_0-J_7, J_7-J_{16} et J_{16}-J_{29}) est présentée dans la figure 4. La corrélation entre survie et quantité d'aliment distribuée est positive ($p<0,05$) pour la période J_7-J_{16}.

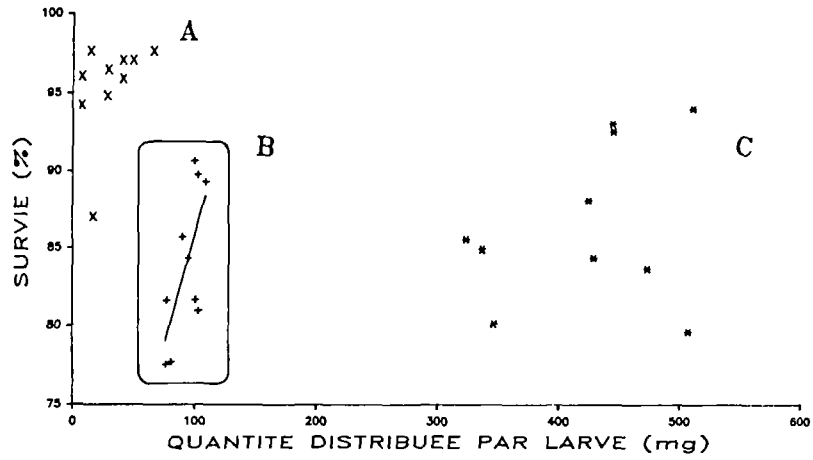


Figure 4 - Relation entre la survie et la quantité d'aliment distribuée par larve (mg)
A : 100 - 200 µm entre J_0 et J_7 B : 200 - 400 µm entre J_7 et J_{16}
C : 400 - 630 µm entre J_{16} et J_{29}

La figure 5 donne les poids moyens relevés en cours d'expérience. Les courbes A et B correspondant aux régimes de démarrage R1 et R2 ont été établies aux jours J_0, J_7, J_{16}, J_{22} et J_{29} sur un échantillon de 20 individus conservés dans du formol. Le taux de croissance spécifique calculé entre J_0 et J_{29} est d'environ $10\%.j^{-1}$. Les courbes C et D très similaires entre elles quel que soit le régime de démarrage donnent les poids frais établis aux jours J_{22}, J_{29} et J_{47} sur la totalité des lots en élevage.

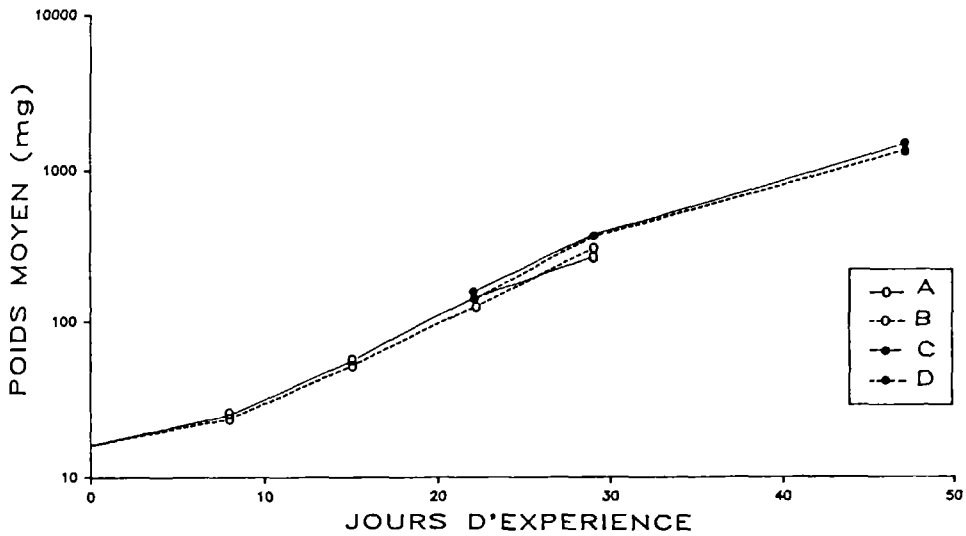


Figure 5 - Croissance pondérale en fonction des régimes de démarrage R1 (——) et R2 (-----), mesures effectuées sur des lots formolés (courbes A et b) ou frais (courbes C et D)

Ces résultats permettent de mettre en évidence une augmentation du taux de croissance spécifique pendant la période J_0-J_{29} puis un ralentissement (8%.j^{-1} entre J_{30} et J_{47}).

L'histogramme de fréquence de répartition des classes de poids en fin d'expérience est donné dans la figure 6. La structure des populations est unimodale. Pour R'1 (effectif de 533 poissons), le poids moyen frais est de 1,58 g. Pour R'2 (effectif de 492 poissons), il est de 1,44 g. Les coefficients de variation pour R'1 et R'2 sont respectivement de 40 et 44%. Les valeurs extrêmes des poids individuels sont 0,22 et 3,95 g.

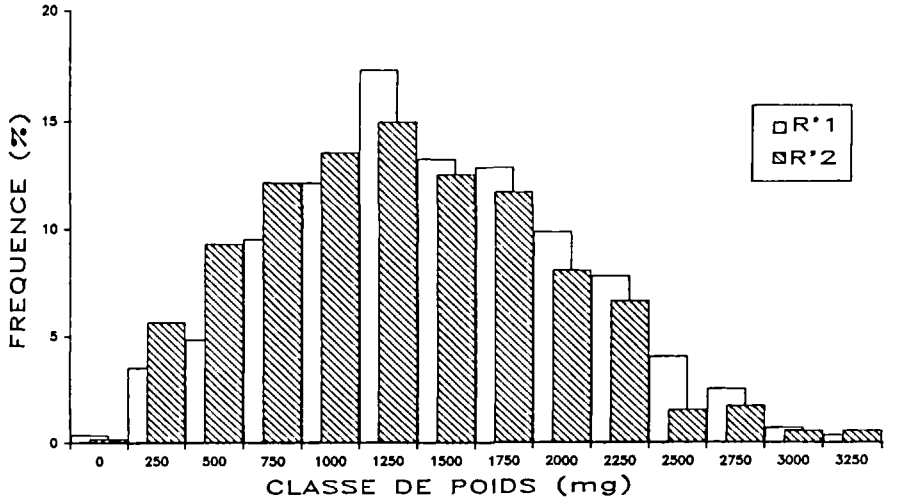


Figure 6 - Histogramme de fréquence de poids au jour J_{47} (intervalle : 250 mg) pour les lots R'1 (N = 533) et R'2 (N = 492)

Les coefficients de condition K = 100 $P.L^{-3}$ (P en mg et L en mm) et le coefficient de régression b de la relation $P = a.L^b$ ont été calculés sur des échantillons légèrement anesthésiés prélevés aux jours J_{22}, J_{29} et J_{47} (Tabl. 2). On note que K diminue entre J_{22} (moyenne = 0,95) et J_{47} (moyenne = 0,52) ; b diminue également pendant cette même période (de 3,27 à 2,66).

DISCUSSION

Les résultats de survie (70% en fin d'expérience) et de croissance (taux de croissance d'environ 10%.j^{-1}) obtenus dans les présentes conditions expérimentales de densité élevée, sans être optimaux, sont comparables à ceux de la littérature (Tabl. 3).

Les plus fortes densités initiales testées (66 larves.l^{-1}), correspondant aux plus petits bacs, ne semblent pas entraîner de baisse notable des performances par rapport aux densités plus faibles (33 larves.l^{-1}), obtenues dans les plus grands bacs ayant un renouvellement d'eau 2 fois plus faible.

Les 2 aliments artificiels R1 et R2 qui ne diffèrent que par la nature de la levure incorporée donnent des résultats voisins pour la survie et la croissance. Ces 2 régimes donnaient également de bons résultats chez le corégone (Bergot et al., 1986).

Tableau 2 - Poids frais individuel moyen Pm (mg) et longueur moyenne Lm (mm) mesurés aux jours J_{22}, J_{29} et J_{47}.
Calculs du coefficient de condition $K = 100\ P.L^{-3}$ et du coefficient b de la relation $P = a.L^b$.
(Pm à J_0 = 16 mg).

DATE	J_{22}		J_{29}		J_{47}	
REGIME	R1	R2	R1	R2	*Aliment du commerce*	
Effectif	20	20	20	20	78 (R'1)	61 (R'2)
Pm	177	164	347	378	1596	1611
Lm	26,30	26,10	33,90	34,60	68,40	66,80
K	0,97	0,93	0,89	O,92	0,50	0,54
b	3,24	3,33	2,64	2,83	2,68	2,64
Ecart-type de b	0,11	0,13	0,13	0,21	0,04	0,05

Tableau 3 - Comparaison des conditions d'élevage des larves d'*A. baeri*

A_i : âge initial (nombre de jours après l'éclosion)
P_i : poids initial (mg)
t_m : température moyenne (°C)
T : durée de l'expérience (jours)
C : charge initiale (larve.l^{-1})
S : survie (%)
TCS : taux de croissance spécifique

Réf	A_i	P_i	t_m	T	C	S	TCS
(1)	8	33	15-16,6	37	3,8	58-85	-
(2)	8	32,6	22	22	3,3	69-86	12-14
	6	33,6	20	22	13,3	58-84	7,7-10
(3)	9	20	17,5	30	28	52	13
(4)	9	20	17,5	30	28	45	11
(5)	4	16	17	30	33-66	70	10

(1) Evgrafova et al. (1982): alimentation naturelle
(2) Semenkova (1983) : alimentation artificielle (Ehkvizo)
(3) Dabrowski et al. (1985) : alimentation naturelle (Tubifex)
(4) Dabrowski et al. (1985) : alimentation artificielle (foie + levure)
(5) Présent travail : alimentation artificielle (foie + levure)

$TCS = 100(\ln P_f - \ln P_o).T^{-1}$ avec P_f = poids final au temps T et P_o = poids initial

Chez *Acipenser baeri* nourri sur aliment artificiel, Dabrowski et al. (1985) signalent la disparition de 7,3% de l'effectif de départ et l'attribuent au cannibalisme. Dans la présente expérience, le nombre d'animaux mutilés qui a été régulièrement

comptabilisé témoigne d'un comportement agressif. Cependant le bon accord entre les effectifs observés et théoriques en fin d'expérience montre que pratiquement aucune disparition complète de poissons ne peut être imputée au cannibalisme puisque le taux d'animaux disparus n'est que de 1,1% pour la période J_0 - J_{29}.

Emel'yanov (1981) considère le cannibalisme chez l'esturgeon comme un réflexe de capture de proie lorsque la distribution d'aliment n'est pas assez importante. Des études complémentaires sur la prise de nourriture et le comportement alimentaire seraient à développer pour préciser ce phénomène.

La mortalité initiale (J_0-J_7) observée paraît trop précoce pour être attribuée à un phénomène de jeûne. En effet, en prenant comme critère de première prise de nourriture l'expulsion du bouchon de mélanine (Krasnodembskaya et al., 1983), on peut estimer que les larves ont commencé à se nourrir entre J_4 et J_{10}, c'est-à-dire entre le huitième et le quatorzième jour après l'éclosion. Dans ses expériences sur la même espèce, Semenkova (1983) situe même le passage à l'alimentation exogène encore plus tardivement (entre 13 et 16 jours après l'éclosion pour des températures de 20-22 °C). Dabrowski et al. (1985) observent aussi une mortalité importante au cours des 7 premiers jours d'expérience, en mettant en cause la qualité des larves plutôt que la nature de leur alimentation ; il pourrait en être de même dans le présent travail. Krasnodembskaya et al. (1983) signalent l'apparition précoce de larves présentant des anomalies de développement, ces larves mourant 5 à 7 jours après l'éclosion.

La corrélation positive observée entre la survie et la quantité d'aliment distribuée pendant la période J_7-J_{16} suggère que la distribution a été temporairement insuffisante dans certains bacs.

Avec un taux de croissance spécifique TCS voisin de 10% par jour et un indice de "distribution" (rapport quantité d'aliment distribuée/gain de poids) proche de 2, on peut estimer le taux d'alimentation à environ 20% du poids vif par jour. Semenkova (1983) avec l'aliment artificiel "Ehkvizo" utilise un taux d'alimentation de 30% du poids vif par jour pendant la période initiale, ce qui correspond, pour des taux de croissance spécifique de 8 à 14% par jour, à des indices de "distribution" de 3,8 à 2,1.

Pendant la deuxième partie de l'expérience c'est-à-dire entre J_{30} et J_{47}, le taux d'alimentation a été plus faible (environ 10% du poids vif par jour pour un indice de "distribution" de 1,2) ainsi que le taux de croissance spécifique ($8\%j^{-1}$). Chez les juvéniles d'*Acipenser baeri*, le taux de croissance spécifique est encore plus bas ; Kaushik et al. (1989) indiquent une valeur d'environ $1\%j^{-1}$ pour des individus de 90 g élevés à 17,5 °C.

La période de croissance initiale s'accompagne de modifications importantes de la morphologie. Dabrowski et al. (1985) signalent une différence importante entre animaux de même âge nourris avec des aliments différents. Pour le coefficient b de la relation $P = a.L^b$, ces auteurs indiquent une valeur de 2,15 pour un lot (de poids moyen 1,45 g) nourri avec des *Tubifex* et de 2,45 pour un lot (de poids moyen 0,96 g) nourri avec un aliment artificiel à base de levure et de foie de porc lyophilisé.

Le présent travail montre que la valeur de b diminue avec la taille (diminution de 3,2 à 2,7 entre J_{22} et J_{47}). Cela suggère que la différence attribuée au régime pourrait être également due à une différence de taille des animaux comparés.

Entre J_{22} et J_{47}, le coefficient de condition diminue de 0,95 à 0,52. Cette dernière valeur est du même ordre de grandeur pour des individus plus grands chez différentes espèces d'esturgeon : de 0,35 à 0,61 pour les esturgeons sibériens (*A. baeri*) de 2 à 3 ans (Sokolov et al., 1986), de 0,35 à 0,58 pour les esturgeons atlantiques (*A. sturio*) entre 20 cm et 2,50 m (Magnin, 1962 ; Ninua, 1976).

Aucune donnée n'est disponible à notre connaissance sur le coefficient de condition des larves. Les présents résultats suggèrent qu'une morphologie de type adulte est atteinte précocement, dès la septième semaine. Les modifications de la morphologie des larves liées à la qualité de l'aliment restent à préciser. Ce travail qui

montre la possibilité de produire des alevins d'*Acipenser baeri* de façon intensive (densité élevée et alimentation entièrement artificielle) ne constitue cependant qu'une étape préliminaire ; il conviendrait de vérifier à grande échelle les résultats obtenus en petits bacs.

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GROWTH COMPARATIVE TRIAL OF FINGERLINGS OF STERLET (Acipenser *ruthenus* L.) AND THAT OF ITS HYBRID WITH MALE SIBERIAN STURGEON (*Acipenser baeri* B.)

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RESUME

L'élevage expérimental de l'esturgeon sibérien a commencé en Hongrie au début des années 1980. Lorsque les esturgeons sibériens mâles ont été pubères, on a cherché à les croiser avec le sterlet, indigène en Hongrie.

L'hybride et le sterlet ont été produits en mai 1987 durant la période de reproduction naturelle du sterlet du Danube. Ensuite on a comparé la croissance du sterlet et de son hybride avec l'esturgeon sibérien mâle dans un circuit fermé à une température de 22-23 °C.

Quel que soit l'aliment utilisé (naturel et 3 formules différentes d'aliments artificiels), la croissance de l'hybride est meilleure que celle du sterlet.

Mots clés : *Acipenser ruthenus, Acipenser baeri* mâle, hybride, croissance, alimentation.

ABSTRACT

The experimental breeding of the Siberian sturgeon has been started at the beginning of the eighties in Hungary. When the Siberian sturgeon males became sexually matured, an attempt has been made to crossbreed this sturgeon with the native sterlet.

The hybrid and the sterlet were propagated in May 1987 during the natural spawning season of the sterlet. After successfull propagation, a comparative trial on the growth rate of sterlet and the hybrid was carried out in a water recycling system at a water temperature of 22-23 °C.

Natural feed and three different type of formulated feeds have been applied in the experiments.

The growth rate of the hybrid was proved to be higher in each experimental group.

Key-words : *Acipenser ruthenus, Acipenser baeri* male, hybrid, growth, feeding.

INTRODUCTION

The experimental breeding of the Siberian sturgeon has been started at the beginning of the eighties, when a small stock of Siberian sturgeon was introduced to Hungary. Because the growth of hybrids of *Acipenseridae* appears to be intermediate between that of the parental species (Burtsev and Serebryakova, 1973 in Chevassus, 1983) an attempt has been made to crossbreed the Siberian sturgeon with the native sterlet that is the only sturgeon species in Hungary can be found in large populations in natural waters.

The major goal of the experimental work was to determine whether the hybrid has a better viability and growth than at least one of the parents. The results of the comparative trial on the growth of sterlet and that of the hybrid of sterlet and Siberian sturgeon have been summarized in this paper.

MATERIAL AND METHODS

The hybrid and the sterlet were bred in May 1987 during the natural spawning season of the sterlet. The sterlet spawners captured from the Danube river while the Siberian sturgeon males were collected from fish ponds of the Fish Culture Research Institute where they has been raised until their sexual maturation.

The release of the sexual products was induced by a synthetic LH-RH hormone (D-Ala^6 Pro^9 - NH-ethylamide). Single treatment was applied with a dose of 100 µg/kg for the female sterlets and 50 µg/kg for the male Siberian sturgeons. The fertilization, treatment and incubation of the eggs were done according to the method commonly used for the propagation of sturgeons (Milstein, 1972). The larvae were nursed in 100 litres raceways for twenty days where they feeding on zooplankton, shopped tubifex (Oligochaeta) and dry feed.

The comparative trial on the growth rate of sterlet and the hybrid was carried out through two consecutive experiments in a water recycling system where the water temperature was kept between 22 and 23 °C.

The first experimental period took 53 days when 130 nursed sterlets and 130 nursed hybrids were stocked into one experimental tank with a water volume of 300 litres. The fish were fed on zooplankton, tubifex and formulated feed *ad. libitum.*

During the second, 56 days long experimental period 40 sterlets and 40 hybrids were stocked into a 300 litres tank. Four experimental groups were used in this experiment.

In three of these groups the initial body weight of the hybrid was higher than that of the sterlet in accordance with the average final body weight of the fish after the first experimental period. In these groups the fish were feeding on three different feeds as shown in Table 1.

Table 1 - The composition of the feeds applied in the experiment

Ingredients	Feed No. 1	Feed No. 2	Feed No. 3
Dry mixture	65[1]	65[1]	100[2]
Boiled blood (from slaughterhouse)	35	-	-
Minced fish (Without head)	-	35	-
Dry matter	63	69	89
Crude protein	37,4	37,6	42

[1] The composition of the dry mixture : 40 % wheat meal, 30 % yeast, 20 % fish meal, 9 % meat meal, 1 % vitamin premix.
[2] The dry mixture contains fish meal, soya meal, yeast, wheat meal, oil, vitamin premix, lysine and starch. (Majoros F., 1987 personnal communication).

There was however a fourth group, where, the fish were selected so that the sterlet was bigger than the hybrid. This experimental group was feeding on Feed No. 3.

The specific growth rate, food conversion ratio and specific food rate given in Table 2 and 3 were calculated according to the following formulas :

$$\text{SGR (Specific Growth Rate)} = \frac{\ln Wt - \ln Wo}{t} \quad 100 \ (\% \ day^{-1})$$

$$\text{FCR (Food Conversion Ratio)} = \frac{F}{Wt - Wo}$$

Where F = total amount of feed given to the fish

SFR (Specific Food Rate) = SGR x FCR (% day $^{-1}$)

Table 2 - Growth and survival rates of sterlet and its hybrid raised together during the first experimental period

	Age (day)	Number of fish	Average body weight (g) W± SD*	Specific growth rate (% day^{-1}) SGR	Survival rate %	Coefficient of variation CV
Sterlet	20	130	0,218±0,004			
	73	87	15,90±4,66	8,09	66,9	29,3
Hybrid	20	130	0,250±0,013			
	73	102	26,00±4,89	8,76	78,5	18,8

* Standard Deviation

RESULTS AND DISCUSSION

As it is shown in Table 2, during the first experimental period the growth and survival rates of hybrid was respectively 8 % and 17% better than those of sterlet.

The ratio of the stunted individuals was only 4 percent in the hybrid population, it was 8 percent in the sterlet population.

In the second experimental period (see Table 3) the growth rate of the hybrid was 10 percent higher in average, but in the fourth group the growth rate of the hybrid was 36 percent higher than that of the sterlet. The coefficient of variation (CV) of the average final body weights shows that the hybrid population was more uniform than the sterlet.

In the feeding experiments during the second experimental period the Feed No. 2 containing 35 percent minced fish gave the best result, however the large-scale application of this feed is limited because its storage, handling and distribution to the fish tank by automatic feeder is difficult.

In order to utilize the full growth potential of the fish and to eliminate the feeding competition between individuals, the feed was applied *ad libitum*. Therefore the feed conversion ratio could be improved when the feeding ratio is well controlled.

Table 3 - The specific growth rate (SGR) and the feed conversion ratio (FCR) of the sterlet and those of the hybrid of sterlet and Siberian sturgeon between the 74th and 130th day of growing applying three different feeds.

Parameters	Feed No. 1		Feed No. 2		Feed No. 3			
	Sterlet	Hybrid	Sterlet	Hybrid	Sterlet	Hybrid	Sterlet	Hybrid
Number of fishes*	40	40	40	40	40	40	40	40
Average initial weight per fish Wo (g) ± SD	17,0±3,0	28,5±3,8	17,2±2,1	29,1±2,0	17,5±5,4	28,6±5,3	17,8±1,6	15,4±2,1
Average final weight per fish Wt (g) ± SD	50,2±13,4	94,1±18,0	56,7±10,0	113,4±4,5	55,6±5,3	95,4±5,3	52,5±3,9	67,3±6,6
Total food consumed F (g)	14268		15036		9908		7422	
Specific growth rate SGR (% day $^{-1}$) individual	1,93	2,13	2,13	2,43	2,06	2,15	1,93	2,63
Feed conversion ratio FCR (%)	3,61		3,04		2,36		2,14	
Specific food rate SFR (% day $^{-1}$)	7,44		7,06		5,00		4,92	
Coefficient of Variation of initial weight (CV_i)	17,6	13,3	12,2	6,9	30,8	18,5	9,0	13,6
Coefficient of Variation of final weight (CV_f)	26,7	19,1	17,6	4,0	9,2	5,6	7,4	9,8

* There was no fish loss during the experiment.

Since the growth rate is depending on the initial body weight, the better growth of the hybrid has become even more evident. During the second part of the experiments the average initial body weight of the hybrid was 200 percent higher in comparison with that of the sterlet, but the growth rate of the hybrid was higher even in these circumstances.

Although both the hybrid and the sterlet populations were relatively uniform in size, it should be noted however that the *ad libitum* feeding reduced the feeding competition between the individuals that was further helped by the availability of zooplankton as "diffuse-feed" during the first part of the experiments.

The average final body weight of the hybrid fry was 250 mg after a 20 days long nursing period at 19-23 °C water temperature, that found to be favourable compared to data from literatures. In an experiment with *Acipenser baeri*, the average body weight of the fry was 271 mg after a 26 days long rearing period when the fish was feeding on tubifex at a water temperature of 17,5 °C (Dabrovsky et al., 1985).

Different formulated feeds were applied by Semenkova (1983) during one experiment with Siberian sturgeon when the best average body weight was 167 mg after a 23 days growing period at19-23 °C water temperature.

CONCLUSION

Based on the results of the experiments, it was found that both the sterlet and the hybrid of the sterlet and the Siberian sturgeon can be cultivated with good results in tanks in intensive systems.

The hybrid was found to be not only viable but its growth rate was higher than that of the sterlet under similar conditions. By maintaining a smaller stock of Siberian sturgeon spawners and using hybridization, large scale commercial production of sturgeon becomes reality even in those regions where sterlet is the only readily available sturgeon species.

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GROWTH OF SIBERIAN STURGEON (*Acipenser baeri* B.) AND THAT OF ITS BOTH HYBRIDS WITH THE STERLET (*Acipenser ruthenus* L.) IN RECYCLING SYSTEM

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RESUME

On a mené des travaux sur la croissance de l'esturgeon Sibérien et celles de ses 2 hybrides avec le sterlet. Les expérimentations ont été réalisées dans un circuit fermé avec des densités de poissons importantes nourris de façon intensive.

Dans nos conditions expérimentales et sur la base de nos résultats, on peut admettre que les croissances de l'esturgeon sibérien et de ces 2 hybrides est principalement influencées par des facteurs technico-écologiques ; l'influence génétique étant secondaire.

Mots clés : *Acipenser baeri, Acipenser ruthenus*, hybrides, croissance, taux de nourrissage.

ABSTRACT

The investigations on the growth of Siberian sturgeon and that of the two hybrids of Siberian sturgeon and sterlet was carried out in a recycling system where high stocking density and intensive feeding were applied.

In our experimental conditions based on the results it can be assumed that the differences in the growth of the Siberian sturgeon and the hybrids were primarily influenced by the technological - ecological factors and the effect of the different genetical background had only secondary importance.

Key-words : *Acipenser baeri, Acipenser ruthenus*, hybrids, growth, feeding rate.

INTRODUCTION

Based on the results of a previous comparative trial on the growth of sterlet and the hybrid of sterlet and Siberian sturgeon in 1987, it was found that the hybrid was not only viable but its growth rate was much higher than that of the sterlet (Ronyai et al., 1990).

Meanwhile the Siberian sturgeon females raised in fish ponds became sexually mature, and thus the Siberian sturgeon could be propagated and the reciprocal hybrid, i.e. the hybrid of Siberian sturgeon and sterlet could also be produced. (When the name of the hybrid is mentionned in this paper, the first fish is always the female).

In 1988, the main goal of our experimental work was to collect information on the growth potential of the Siberian sturgeon and its hybrids in intensive system in the conditions of large scale production.

MATERIAL AND METHODS

The propagation was carried out at a water temperature of 16-17 °C using synthetic LH-RH hormone (Ronyai et al., 1990). The larvae were fed on chopped Tubifex during the first two weeks then dry feed with 48 percent protein and 9 percent of lipid content was applied.

Experimental groups, consisting of one thousand individuals each, were separated from the large stock of Siberian sturgeon, sturgeon x sterlet hybrid and reciprocal hybrid.

Samples were taken in every four weeks when the average body weight of a randomly selected group (about 10 percent of the population of the experimental group) was measured. The body weights were individually measured only on the first and last days.

The fish were also graded and divided into groups by size in every four weeks and the daily feeding ratio was corrected according to the followings.

Average body weight (g)	5-10	10-50	50-100	100-200	200-400
Daily feeding ratio (% body weight)	8,0	6,0	5,0	4,0	3,0

After 87 days of rearing the largest twenty individuals from each of the three experimental groups have been selected and marked.

These fishes were stocked and reared together in a common tank. The method of sampling and feeding was the same as described above.

Since feeding is one the most important factors effecting growth one additional experiment was carried out in order to investigate the effect of the different daily feeding ratios. Six experimental groups were formed consisting of sterlet x Siberian sturgeon 50 individuals each. The average initial body weight of the fish was 5 grams. The duration of the experiment was 98 days. These groups were fed with a daily feeding ratio of 3,4,5,6,7,8 percent body weight respectively. The specific growth rate and food conversion ratio were calculated according to the following formulas :

$$\text{SGR (Specific Growth Rate)} = \frac{\ln Wt - \ln Wo}{t} \times 100$$

$$\text{FCR (Feed Conversion Ratio)} = \frac{F}{Wt - Wo}$$

where F represents the total amount of feed given to the fish during the experiment.

In case of all experiments the stocking density and the water flow were adjusted so that the relative oxygen saturation of the outflow water did not decrease below 70 percent (Williot et al., 1988). The experiments were carried out in 2 m^3 tanks, that were placed in a large scale recycling system having 80 m^3 total water volume. The water in the system was heated by geothermal energy and its temperature was kept at 22 ± 1 °C.

Student t-test was applied in the statistical analysis.

RESULTS AND DISCUSSION

The results of that experiment where the growth rate of 1000 individuals from the Siberian sturgeon and the two hybrid population have been investigated are shown in Table 1.

Table 1 - Growth rate of the Siberian sturgeon, Siberian sturgeon x sterlet hybrid and Sterlet x Siberian sturgeon hybrid in intensive conditions.

Age (days)	Average body weight (g)			Stocking density (kg/m^3)
	Siberian sturgeon	Siberian sturgeon x Sterlet hybrid	Sterlet x Siberian sturgeon hybrid	
26	0,46 (0,09)*	0,54 (0,07)	0,41 (0,08)	
54	4,10	6,80	4,60	5,0
82	22,69	23,36	21,05	10-15
117	56,41	57,95	56,18	20-30
138	80,83	89,68	83,44	25-35
165	134,10	134,83	113,61	20-50
193	211,05	235,82	253,00	25-60
222	258,25	277,30	290,60	30-80
264	334,35	348,75	335,27	35-85
297	417,00 (66,72)	469,30 (98,55)	403,30 (77,43)	35-85

* The numbers in bracket show the standard deviation of the average body weight of a sample consisting of 100 fish.

Although the average body weight of the Siberian sturgeon x sterlet hybrid is 13-14 percent higher than that of the other hybrid and the Siberian sturgeon, the difference is not significant. ($P>0,05$).

High diversity in the sizes of the individual fishes could be observed in each group. The coefficients of variation of individual body weight of Siberian sturgeon, Siberian sturgeon x sterlet hybrid and sterlet x Siberian sturgeon hybrid were 16,0 %, 21,0 % and 19,2 % respectively.

The competition between the individuals was further increased by the high stocking rate, however it is assumed that applying regular grading, more uniform stock could be produced. Different results have been obtained in that experiment where 20-20 individuals of the Siberian sturgeon and the two hybrids were grown in one common tank. The results of the experiment are shown in Table 2.

Table 2 - Growth of the Siberian sturgeon and its hybrids raised together in a common tank *

Age (days)	Average body weight (g)		
	Siberian sturgeon	Siberian sturgeon x Sterlet hybrid	Sterlet x Siberian sturgeon hybrid
87	33,56 (3,17)	36,28 (3,50)	33,74 (3,98)
95	49,70	50,00	49,37
115	96,90	94,24	98,94
138	143,70	138,43	150,78
165	221,00	20,8,25	243,16
270	456,50	457,30	547,30
333	596,53 (114,18)	553,80 (127,71)	662,68 (112,20)

* During the experiment the stocking density did not exceed 30 kg/m^3.

The average body weight of the 11 month-old sterlet x Siberian sturgeon was significantly higher ($P<0,05$) than that of the Siberian sturgeon x sterlet hybrid. One of the reasons why different results have been got comparing to that experiment where the three groups were separated may be the competition between the three different kind of fishes.

The effect of the relative daily feeding ratio on the growth of the sturgeon species as well has been described (Hung et al., 1987, 1989).

In our experiments it could also be observed that the daily feeding ratio had a primarily effect on the growth rate of the Siberian sturgeon and its two hybrids with sterlet as shown in Table 3.

Table 3 - The growth rate of the Sterlet x Siberian sturgeon hybrid at different daily feeding ratio.

Relative daily feeding ratio * (% day $^{-1}$)	Mean final body weight (g)	SGR (% day $^{-1}$)	FCR
3,0	40,0	2,12	1,19
4,0	68,0	2,66	1,27
5,0	88,0	2,93	1,42
6,0	105,3	3,11	1,65
7,0	134,5	3,36	1,94
8,0	153,0	3,76	2,20

* Based on the weekly samplings. There was no feeding on the day before sampling.

These experiments were carried out during semi-scale production, therefore the results could be considered as preliminary ones and further investigations are needed to compare the growth potential of these three kind of sturgeons.

It can be concluded however that based on our results and observations the growth potential of the Siberian sturgeon and its two hybrids with sterlet are very similar. It can be assumed that in our experimental conditions their growth rate is primarily influenced by the different technological factors (stocking density, feeding, etc...) and the effect of the different genetical background had only secondary importance. It should be noted that practically there was no mortality from that phase of the growing period when the size of the fish exceeded 2,0 gram body weight.

CONCLUSION

The results of the experiments clearly indicated that the production of the two hybrids of the Siberian sturgeon (*Acipenser baeri* B.) and the sterlet (*Acipenser ruthenus* L.) has a great potential in intensive conditions.

Based on the results of our experiments it can be concluded that the production of the hybrids in intensive conditions might give at least the same results than that of the Siberian sturgeon.

During the experiments the average body weight of the Siberian sturgeon and its hybrids have been increased slighthy above 400 grams in 10 months.

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BILAN SANITAIRE DE 2 ANS D'ELEVAGE D'*Acipenser baeri* EN PISCICULTURES INTENSIVES

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RESUME

Un suivi pendant deux années de l'élevage de l'esturgeon sibérien *A. baeri* dans des pisciculture de type intensif, ainsi que des analyses au Laboratoire, permettent de dresser un bilan sanitaire. Deux syndromes, déformation de la colonne vertébrale et nécrose des holobranchies, sont discutés. Les résultats des analyses parasitologiques, bactériologiques, mycologiques et virologiques sont présentés. Ils ne font apparaître aucune pathologie spécifique mais mettent en évidence la sensibilité de l'esturgeon vis à vis de *Y. ruckeri*, *Vibrio anguillarum* et de *Flexibacter columnaris*.

Mots clés : *Acipenser baeri*, pathologie, elevage intensif.

ABSTRACT

After two years of rearing experiment and laboratory analysis on *A. baeri* (siberian sturgeon) we can draw up a pathological assessment. Two syndromes : spine deformation and branchiae necrosis are described. Results of parasitologicals, mycologicals and virologicals analysis are presented. These results show there is no specific pathology but sturgeon is sensitive to *Y. ruckeri*, *Vibrio anguillarum* and *Flexibacter columnaris*.

Key-words : *Acipenser baeri*, pathology, intensive rearing.

INTRODUCTION

L'esturgeon sibérien *A. baeri* a été introduit en France en 1975 et élevé dans une pisciculture alimentée par une eau à température constante provenant d'une résurgence. Dans ces conditions d'élevage, aucune pathologie spécifique n'a été diagnostiquée et cette espèce montrait même de réelles potentialités de croissance.

Toutefois, avant de développer l'élevage d'*A. baeri* un suivi sanitaire a été mis en place durant deux années, dans les six pisciculture ayant reçu un lot de ces animaux dont l'état sanitaire a été contrôlé au préalable. Ainsi, il a été possible d'observer et d'analyser le comportement de ces lots dans des conditions d'élevage différentes.

Dans le cas où les pathologies étaient d'origine infectieuse, deux objectifs ont été recherchés, d'une part, mise en évidence des agents pathogènes spécifiques de l'esturgeon et, d'autre part, recherche des agents pathogènes des autres espèces, essentiellement des salmonidés, pathogènes ou non pour l'esturgeon et pouvant nuire à l'élevage des truites.

MATERIEL ET METHODES

Pisciculture

Leurs caractéristiques et leurs antécédents pathologiques sont indiqués dans les deux tableaux ci-après.

Tableau 1- Caractéristiques des pisciculture

n°	Origine de l'eau	Type de pisciculture	Température Moyenne	Maxi.	Mini.	Localisation des esturgeons dans la pisciculture
1	Rivière	Salmoniculture (Reproduction-grossissement)	13 à 14°C	18°C	4°C	En tête de pisciculture
2	Rivière	Salmoniculture (Grossissement)	13 à 14°C	17°C	6°C	Bassins d'alevinage (en tête au début) bassin de grossissement ensuite
3	Rivière	Salmoniculture (Grossissement)	5 - 16°C	22°C	8°C	Bassin en amont des des bassins truites
	Rivière + forage	Salmoniculture (Reproduction-grossissement)	7 - 8°C	16°C	3°C	Selon les cohortes en amont ou en aval des bassins d'alevinage de truites
5	Résurgence	Salmoniculture et élevage d'esturgeons (Reproduction - grossissement)	16 - 18°C	18°C	14°C	En amont, puis en aval des bassins d'alevinage de truites
6[1]	Pompage dans estuaire + rivière	Grossissement esturgeons	16 - 18°C	26°C	14°C	Ensemble des bassins

(1) la salinité peut atteindre exceptionnellement 18 °/°° dans cette pisciculture.

Tableau 2- Antécédents pathologiques (sur 2 années) des six pisciculture ayant reçu des esturgeons

n° Pisciculture	Septicémie Hémorragique Virale	Nécrose Pancréatique Infectieuse	Yersiniose	Hépato-Néphrite Parasitaire	Vibriose	Cytophagales (myxo-bactéries)
1	-	+	+	+	-	+
2	-	+	+	+	-	+
3	NC	NC	NC	NC	NC	NC
4	+	+	+	NC	NC	+
5	-	NC	+	NC	-	+
6	NC	NC	NC	NC	NC	NC

NC : non controlé avant le suivi des esturgeons
+ : présence de l'agent responsable de la maladie
- : absence de l'agent responsable de la maladie sur une durée d'au moins deux ans.

Animaux

Les esturgeons avaient, pendant cette expérimentation, de 2-3 mois à 2 ans d'âge, à part la pisciculture 5, dans laquelle se trouvaient des géniteurs (entre 10-14 ans), des oeufs, des alevins et des juvéniles.

Interventions sur la pisciculture :

Elles comportent différentes étapes:
Entretien avec le responsable de la pisciculture
Cet entretien a pour but de faire le point sur la gestion de l'élevage au cours du mois écoulé. A cet effet, les fiches d'enregistrements quotidiens remplies par le pisciculteur sont analysées. Ces documents comportent notamment: la température journalière de l'eau, le taux d'oxygène dissous, les quantités quotidiennes d'aliments distribués, les mortalités et les événements ayant pu avoir une incidence sur la santé des animaux. Si nécessaire, en fonction des problèmes rencontrés au cours du mois précédant cet entretien, la qualité du milieu d'élevage est vérifiée (température, oxygène, sursaturation...)

Observation des animaux
Avant de procéder aux opérations de tri ou de pesée, le comportement des animaux est observé, ainsi que l'appréciation de l'état général.

Recherche d'anomalies externes
La recherche de ces anomalies concerne d'une part les animaux dont le comportement ou l'aspect attire l'attention et d'autre part des animaux apparemment sains pris au hasard. Les poissons sont sortis du bassin d'élevage, posés sur une tablette humide et sont soumis aux observations suivantes: - peau, examen minutieux sur l'ensemble du poisson afin de déceler les lésions (plaies, hématomes) - nageoires, (recherche de nécroses éventuelles) - orifice ano-uro-génital, examen de fèces (aspect) après légère pression abdominale - colonne vertébrale, vérification de la rectitude de celle-ci - branchies, chaque holobranchie est contrôlée avec soin. Pour ce faire, il est nécessaire de maintenir l'opercule écarté et de bien séparer, au moyen d'une petite spatule, les holobranchies de façon à voir apparaître toutes les lamelles branchiales constituant chaque holobranchie.

Prélèvements
En fonction des observations réalisées, plusieurs prélèvements peuvent être effectués :- frottis (peau et lamelles branchiales) - prélèvements coprologiques à l'aide d'un embout souple adapté à une seringue ou à l'aide d'un écouvillon stérile - prise de sang - animaux malades et/ou animaux apparemment sains. Ces prélèvements seront exploités sur place ou, ultérieurement, au laboratoire.

Examens de laboratoire

Autopsies
Les autopsies sont effectuées en laboratoire. Les lésions externes et internes, après ouverture de la paroi abdominale, sont notées. Les organes sont prélevés pour effectuer les analyses complémentaires : parasitologie, bactériologie, mycologie et virologie.

Parasitologie
L'examen parasitologique s'effectue par recherche directe de parasites sur les animaux au moment de l'autopsie et l'observation de ceux-ci au microscope optique si nécessaire (x100 ou x400). La recherche de l'agent de l'hépatonéphrite parasitaire de la truite se fait sur les calques de reins après coloration au May-Grünwald-Giemsa.

Bactériologie - mycologie

L'isolement s'effectue selon les techniques habituelles de bactériologie et de mycologie utilisées en médecine vétérinaire. Des examens directs sont effectués sur les frottis d'organes lésés après coloration au bleu de méthylène. Les prélèvements de foie, rate, reins et autres organes lésés selon les cas, sont ensemencés sur gélose trypticase soja et incubés à 20°C.

Les recherches de champignons sont effectuées sur le milieu gélosé de Sabouraud. Souvent, un simple examen à l'état frais, au microscope, du produit pathologique permet de les mettre en évidence.

Les caractères culturaux (morphologie des colonies, morphologie des bactéries après coloration de Gram) et le profil biochimique obtenu en galeries Api (Api 20E, Api 20NE, principalement), permettent l'identification des souches bactériennes isolées.

Virologie

Recherche des virus des salmonidés

Virus recherchés : les birnavirus virus de la nécrose pancréatique infectieuse (NPI), Cohen et al. (1973), Dorson et al. (1978) et les rhabdovirus des salmonidés virus de la septicémie hémorragique virale (SHV) Zwillenberg et al. (1965), virus de la nécrose hematopoïetique (NHI) Mc Allister et al. (1974), Darlington et al. (1974), ont été systématiquement recherchés sur chaque lot d'esturgeons.

Les analyses sont effectuées selon les techniques décrites par de Kinkelin et al. (1985), à partir des prélèvements de reins, rate et cerveau de 2 à 5 esturgeons par lot et, dans certains cas, à partir du sperme et des ovules.

Recherche des virus des esturgeons

Dans certaines manifestations pathologiques (notamment l'érosion des holobranchies et les déformations vertébrales) observées, des recherches histologiques en microscopie optique sont entreprises pour essayer de mettre en évidence des lésions d'origine virale.

Histologie

Les organes lésés sont fixés dans du liquide de Bouin. Les analyses ont été effectuées au laboratoire d'anatomie pathologie de l'Ecole Nationale Vétérinaire d'Alfort, afin de déterminer la nature des lésions et d'obtenir des indications permettant d'en identifier la cause.

RESULTATS

Les interventions réalisées dans les pisciculture ont permis de mettre en évidence, dans la pisciculture 5,- des déformations vertébrales sur des animaux de 2 à 6 ans (animaux atteints 30 %, mortalité 7 % sur 3 ans),- des nécroses et une érosion des holobranchies sur différentes cohortes âgées de quelques mois à 6 ans (animaux atteints 50 %, mortalité négligeable),- une mortalité importante d'origine bactérienne (*Y. ruckeri*) sur des alevins (mortalité 31 %).

Dans la pisciculture 6, une vibriose a entraîné une mortalité de 7 %.

Déformation vertébrale

Sur quelques esturgeons, nous avons observé des déformations de la colonne vertébrale. Celles-ci ont été surtout remarquées la première fois sur des animaux ayant entre 2 et 3 ans. Dans la plupart des cas, le point d'inflexion de la colonne se situe soit légèrement dans la partie postérieure, soit au niveau des 11 et 12ème écussons dorsaux comptés à partir de la tête. Les animaux nagent difficilement, perdent l'équilibre, ont des difficultés à s'alimenter et meurent après une période d'asthénie qui peut durer plusieurs mois. L'observation de coupes histologiques, pratiquées sur le cerveau et sur le canal médullaire, montre des lésions de démyélinisation et sur les muscles on constate fréquemment une myosite nécrosante. Elle n'a toutefois pas permis de mettre en évidence une lésion dominante ou caractéristique. D'autre part, une lésion au niveau

du tissu cordal ne peut être totalement exclue, bien qu'elle n'ait pas été systématiquement recherchée.

Nécrose et érosion des holobranchies

En 1986, un examen de routine sur une cohorte de l'année a montré une érosion importante des holobranchies. Ce syndrome peut se manifester de différentes manières : pour les cas banals, l'érosion est superficielle et se rencontre très souvent sur des tissus qui ne sont pas suffisamment recouverts par l'opercule ; dans les cas plus graves, on peut remarquer la disparition irrégulière, sur une ou plusieurs holobranchies, des lamelles branchiales ; très souvent, celles-ci présentent l'aspect d'un tissu qui aurait été découpé à l'aide d'un emporte pièces. Des disparitions totales de tissu branchial ont également été observées ; celles-ci, toutefois, se limitaient à une holobranchie par animal.

Les lésions observées apparaissent irréversibles. Il y a cicatrisation, mais le tissu branchial reste détruit. Les examens histologiques mettent en évidence des lésions d'hyperplasie, d'infiltration cellulaire, de nécrose.

Parasitisme

Des parasites externes ont pu être observés très ponctuellement sans apparition de lésions particulières, ce sont :

- *Costia necatrix.*
- *Ichthyophtyrius multifiliis.*
- *Trichodina reticulata.*
- *Gyrodactylus sp.*

Ce sont des parasites identiques à ceux retrouvés dans les élevages de truite d'Aquitaine. Toutefois, il faut noter que l'infestation était à un taux très faible. Aucun parasite interne n'a été observé.

Yersiniose à Yersinia ruckeri

Au mois de juin 1986, des mortalités anormales ont été constatées sur un lot d'*A. baeri* de 15 à 30 g, âgés de 2 mois. Les poissons étaient répartis en six bacs circulaires alimentés par une eau de résurgence à température constante de 17°C. Un seul bac a été atteint, sa charge était de 6,8 kg/m^3. Les lésions observées étaient des suffusions péribuccales, une congestion du pore ano-uro-génital, un ballonnement de l'abdomen, une diarrhée hémorragique, une congestion généralisée des viscères. *Y. ruckeri* a été isolée des organes des poissons autopsiés (Vuillaume et al., 1987).

Vibriose à Vibrio anguillarum

Dans la pisciculture 6, un lot de *A. baeri* âgés d'un an a subi des mortalités (10 %) en juin 1986 (Miossec, com. pers.). La température de l'eau était de 22-23°C. Les symptômes notés étaient les suivants :- suffusions hémorragiques sous-cutanées, surtout en zone périanale,-surcharge branchiale,- congestion hémorragique de la partie terminale du tube digestif,- suffusions hépatiques, suffusions sur la sereuse de la vessie natatoire.

La recherche de la bactériologie a mis en évidence *V. anguillarum* (Mureau, com. pers.).

Myxobactériose externe

Des bactéries appartenant à l'ordre des cytophagales ont été observées sur les nageoires, où elles provoquent une érosion, sur la peau et sur les branchies d'alevins *A. baeri* âgés de 1 à 2 mois. Elles se sont multipliées sur le milieu d'Anaker et Ordal. Les températures de croissance, la morphologie des colonies font penser à *Flexibacter columnaris*, (Bernardet, 1989).

Autres bactéries

Diverses espèces fréquemment retrouvées chez la truite (Trust, 1974 ; Lesel, 1979), ont été isolées :

- *Aeromonas hydrophila*, qui est la plus fréquente,
- *Acinetobacter sp*,
- *Plesiomonas shigelloïdes*,
- *Enterobacter agglomerans*,
- *Enterobacter cloacae*,
- *Pseudomonas cepacia, Pseudomonas maltophila, Pseudomonas fluorescens*,
- *Pseudomonas spp*,
- *Streptococcus faecium, Streptococcus lactis, Streptococcus spp.*

Mycoses

Dans toutes les pisciculture, des filaments mycéliens sont observés sur la peau de quelques individus rachitiques, qu'il s'agisse d'adultes ou de juvéniles. Ce phénomène est plus fréquent dans la pisciculture d'eau froide (4) (3 à 16°C).

Pathologie virale

Aucune mortalité d'origine virale n'a été observée pendant ces deux années. Les recherches histologiques effectuées à partir des branchies érodées, observées dans certains élevages, n'ont pas permis de mettre en évidence des lésions microscopiques évoquant l'action d'un virus. La recherche des birnavirus et des rhabdovirus des salmonidés a donné les résultats suivants : sur 25 séries d'analyses effectuées en deux ans, aucun rhabdovirus n'a été mis en évidence. Un birnavirus a été isolé une fois sur un lot d'esturgeons de 130 g, sur la pisciculture 4.

Les analyses effectuées à partir de la semence de 9 mâles et les ovules de trois femelles n'ont pas permis de mettre en évidence de virus.

DISCUSSION

Déformations vertébrales

Il n'est pas exclu que ce syndrome puisse avoir plusieurs origines. Pour le moment, nous émettrons deux hypothèses :

Causes alimentaires

Hung, (1987)décrit un syndrome identique chez *A. transmontanus* pour lequel il emet une hypothèse nutritionnelle.

En ce qui concerne *A. baeri*, il est difficile encore de connaître l'étiologie exacte de ce phénomène. Chez les poissons, des carences en minéraux ou en vitamines peuvent être responsables de telles lésions. La carence en vitamine C en particulier, entraînant un défaut de synthèse d'hydroxyproline, provoque des lésions vertébrales. Cette carence pourrait s'expliquer, en partie, par le comportement de l'esturgeon vis-à-vis de l'aliment dont on dispose actuellement. Ce poisson, qui ne présente aucune voracité, consomme lentement les granulés ; ceux-ci s'hydratent, se délitent et libèrent les substances hydrosolubles, dont la vitamine C, avant qu'elles ne soient ingérées. Dans le cas où cette hypothèse serait confirmée, il conviendrait d'élaborer un aliment de structure, voire de consistance différente, qui permettrait de conserver intactes jusqu'à l'ingestion ses qualités essentielles.

Causes traumatiques

Dépourvu d'arêtes et faiblement ossifié, le squelette des esturgeons apparaît comme fragile aux chocs et aux manipulations. A plusieurs reprises sur des animaux

qui, lors de pesées ou de tris, nous ont échappé nous avons constaté une déformation vertébrale.

Nécrose et érosion des holobranchies

Sur une même cohorte élevée dans le même bassin, l'ampleur des lésions est extrêmement variable selon les individus. Les causes de ce syndrome n'ont pu être mises en évidence, aucune étiologie infectieuse n'a pu être avancée pour le moment, les analyses mycologiques, bactériologiques et virologiques ont toujours donné des résultats négatifs. La nature même de la lésion, qui se situe dans de nombreux cas sur des holobranchies mal protégées (opercule atrophié) et qui laisse apparaître un tissu découpé pourrait être la conséquence d'une morsure provoqué notamment par des anguilles que l'on trouve fréquemment dans les bassins d'esturgeons. Cette hypothèse d'un phénomène traumatique n'est, toutefois, pas confirmée.

Une observation réalisée par le Laboratoire National de Pathologie des Animaux Aquatiques de Brest, montre que les filaments présentent des malformations à différents niveaux ; sur une portion relativement importante et généralement médiane, les lamelles sont absentes. A ce niveau, les filaments sont souvent réunis les uns aux autres en une seule lame conjonctivo cartilagineuse recouverte d'un épithelium. A partir de ces lames et vers la partie distale, s'individualisent parfois de nouveaux filaments porteurs de lamelles. L'orientation des lamelles est irrégulière, mais aucune lésion constitutive n'apparaît à ce niveau.

Sur ces tissus observés, les lésions sont anciennes et peuvent être dues, selon les conclusions du laboratoire, à des sursaturations ou à des agressions bactériennes ou parasitaires importantes mais limitées à certains secteurs ou arcs branchiaux.

Sur *A. transmontanus*, Hedrick (comm. pers.) a également constaté un syndrome de nécrose branchiale auquel il attribue une origine virale. Il est difficile de dire si, dans les deux cas, l'étiologie est la même, car pour *A. baeri* aucune inclusion virale n'a pu être mise en évidence.

Parasitoses

Pour ce qui est du parasitisme externe, on ne peut pas prédire quelle intensité prendra le phénomène en intensifiant l'élevage, mais on peut penser que des problèmes apparaîtront comme pour les autres espèces. Le pouvoir pathogène de ces parasites est dû à l'irritation qu'ils provoquent à la peau et surtout aux branchies. Les protozoaires, en particulier *Trichodina* spp par ses denticules, causent des microlésions. D'où la nécessité de mettre au point des protocoles de traitement. Toutefois, Dergaleva (1978) pense que la peau des acipensérídés, de par sa structure et son épaisseur, est moins favorable au développement des parasites que celle des téléostéens.

Bactérioses

Pour la yersiniose, la maladie s'est probablement développée à partir de truites élevées dans la pisciculture et sur lesquelles cette pathologie avait été diagnostiquée un mois auparavant. Des essais ultérieurs de contamination expérimentale ont confirmé la sensibilité des esturgeons à cette bactérie (Chêne, 1989). Au moment de l'épidémie, deux cents poissons provenant du bac contaminé ont été isolés et n'ont subi aucun traitement afin d'observer l'évolution de la maladie. Les mortalités se sont échelonnées sur un mois et ont atteint 31 % . Les autres poissons ont été traités à deux reprises, une première fois au pic de l'épizootie (12 mg/kg de poids vif de fluméquine, pendant 5 jours), et une seconde fois un mois après (12 mg/kg de poids vif d'acide oxolinique, pendant 5 jours). Un lot de ces animaux a été transféré dans la pisciculture 6. La maladie s'est à nouveau déclarée deux semaines après le transfert, à la suite vraisemblablement du stress subi lors de cette manipulation. L'administration d'un traitement à l'acide oxolinique, selon les modalités précédentes évoquées, a permis d'enrayer l'évolution de la maladie.

Pour la vibriose, qui est apparue dans un lot élevé en eau saumâtre, deux séquences de traitement de 5 jours et à 10 jours d'intervalle ont été réalisées avec fluméquine puis acide oxolinique aux doses déjà indiquées (Miossec, comm. pers.).

Parmi les autres bactéries isolées, *A. hydrophila* est rencontrée très fréquemment sur des poissons moribonds ou stressés. Hedrick (com. pers.) a montré qu'*A. transmontanus*, contaminé par inoculation expérimentale, manifeste une certaine sensibilité vis-à-vis de cette bactérie. Par ailleurs, elle est considérée comme pathogène chez les poissons placés dans de mauvaises conditions (De Figuereido, 1977).

Myxobactérioses

Ce sont des infections très fréquentes en élevage piscicole, d'autant plus fréquentes que les poissons sont maintenus dans de mauvaises conditions d'élevage (charges importantes, mauvaises conditions de milieu...). Un traitement externe quotidien à base de chloramine T (18 g/m^3) par bain pendant 1 heure, associé à un traitement général d'oxytétracycline (80 mg/kg de poids vif) pendant 5 jours a permis d'enrayer l'épidémie.

Viroses

Tous les lots d'esturgeons analysés provenaient de pisciculture où les truites étaient porteuses de birnavirus (Nougayrede, 1988) ; une seule fois, un birnavirus a pu être mis en évidence sans signes cliniques particuliers. Des recherches sont en cours sur des alevins d'esturgeons. Plus de la moitié des analyses a été effectuée à partir d'esturgeons élevés dans une pisciculture de salmonidés chroniquement infectée par un rhabdovirus (pisciculture 4). Alors que le rhabdovirus de la septicémie hémorragique virale a été régulièrement isolé sur *Oncorhynchus mykiss* Walbaum analysées en même temps, il n'a jamais été trouvé sur les esturgeons qui, d'ailleurs, n'ont jamais présenté de signes cliniques pouvant évoquer cette maladie.

Cette observation pourrait signifier que cette espèce est insensible au rhabdovirus de la septicémie hémorragique virale.

CONCLUSION

Il apparaît, à la lecture du bilan du suivi sanitaire de deux ans, qu'aucun problème majeur, qui aurait pu compromettre l'élevage d'*Acipenser baeri*, n'a été observé et qu'aucun agent pathogène spécifique de l'esturgeon n'a pu être mis en évidence.

Toutefois, les anomalies constatées sur les branchies, ainsi que les déformations vertébrales, sont des syndromes pour lesquels les causes restent inconnues.

Il a été observé sur le terrain une yersiniose à *Yersinia ruckeri* transmise probablement par les truites, une vibriose à *Vibrio anguillarum* en eau chaude et légèrement saumâtre (3 à 6 °/$_{oo}$), et sur des alevins une atteinte des branchies avec présence de bactéries de l'ordre des cytophagales.

Les parasitoses sont toujours restées négligeables. Quant aux viroses, il semble bien que l'esturgeon *Acipenser baeri* soit résistant aux virus des salmonidés (rhabdovirus et birnavirus) et qu'il pourrait être, comme de nombreuses espèces, porteur de birnavirus apparenté au virus de la nécrose pancréatique infectieuse.

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VIRUS INFECTIONS OF CULTURED WHITE STURGEON (*Acipenser transmontanus*)

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RESUME

Deux virus ont été découvert chez le jeune esturgeon blanc, élevé en Californie, USA. Nous avons rapporté en 1985 la découverte d'un "adenovirus" associé à des infections de la muqueuse de l'appareil digestif.

Des infections avec l'adenovirus ont été trouvé chez les jeunes en 1984, 1985, 1986 mais n'ont pas été vu depuis. Nous pensons que le virus a engendré la mortalité dans les premières phases d'élevage et le virus qui venait des esturgeons malades a été transmis aux esturgeons en bonne santé du laboratoire. Un virus supplémentaire, "l'iridovirus", a été récemment découvert chez plusieurs populations d'élevage d'esturgeons blancs en Californie. Le virus contamine les cellules epithelial de la peau et des ouïes des esturgeons. Les cellules se sont aggrandies et souvent teintées profondément à l'hematoxyline. Des virus d'un diamètre de 262 nm ont été trouvé dans les cellules infecteés. Lors d'infections importantes, on a observé de nombreuses cellules infectées sur la peau et les ouïes de poissons morts et moribonds. Aucun autre pathogène n'a été découvert parmi certaines populations d'esturgeons qui ont succombé à la maladie, et nous présumons que le virus a été la cause de la mortalité. Une étude a montré que le virus a été transmis par l'eau. La propagation étendue du virus parmi les populations de poisson à plusieurs élevages d'esturgeons en Californie, USA a été l'obstacle majeur à la production en 1988.

Mots clés : Iridovirus, maladie des branchies, *Acipenser transmontanus*

ABSTRACT

Two viruses have been detected in juvenile white sturgeon (*Acipenser transmontanus*) reared in California, U.S.A. We reported in 1985 on the discovery of an adenovirus associated with infections of the mucosa of the alimentary tract.

Infections with the adenovirus were found among juveniles reared in 1984, 1985 and 1986 but have not been seen since. The virus was believed to contribute to mortality in the early rearing phases and was shown to be transmitted from diseased sturgeon to previously health sturgeon in the laboratory.

An additional virus, the white sturgeon iridovirus, has recently been detected in several farm-reared populations of white sturgeon in California. The virus infects epithelial cells in the skin and gills. These cells become enlarged and often deeply stained with hematoxylin. Virions with a mean diameter of 262 nm were found within infected cells. In heavy infections, numerous infected cells are observed in the skin and gills of dead and moribund fish. No other pathogens were detected in certain populations of sturgeon which succumbed to the disease and we presume that the virus was the cause of the mortality. A laboratory study showed that the virus was transmitted via the water to previously uninfected white sturgeon held below infected fish. The rapid spread of the virus among populations of fish at several sturgeon farms in California, U.S.A. was a major obstacle to production in 1988.

Key-words : Iridovirus, gill disease, *Acipenser transmontanus*

INTRODUCTION

The artificial culture of white sturgeon (*Acipenser transmontanus*) in California has been growing steadily since its beginning in the 1970s. Several farms are actively engaged in rearing juveniles resulting from gametes obtained from feral broodstock. Growth rates obtained in captivity have been exceptional and these fish have been marketed both for the aquarium trade and as food fish. Diseases encountered in the early-rearing phases and broodstock development are the major difficulties now

encountered by this growing industry. Control or effective management of these diseases will be important to the success of this growing industry.

Although diseases due to environmental, nutritional and genetic causes are important, several of the more serious diseases have been due to infectious agents. Those due to external protozoans, bacteria or fungi can be treated in most cases but diseases due to the viral pathogens present additional problems. For the viral diseases there are currently no approved anti-viral drugs and secondly these agents can potentially be moved with live fish or eggs and are therefore easily disseminated to new geographical regions (Wolf, 1988). In this paper we describe two viruses that have been found during our investigations into diseases affecting cultured white sturgeon.

MATERIALS AND METHODS

Fish

Virus-infected juvenile white sturgeon were obtained from commercial farms between 1983 - 1989. Additional groups of juvenile white sturgeon (control, uninfected) reared from eggs at the laboratory were used in transmission studies.

Light and Electron Microscopy

Sturgeon less than 5 g in size were fixed whole in Davidson's solution (Humason, 1979) for 24 h and then transferred to 70 % ethanol. Portions of the gill, skin (of the operculum), intestine, liver, spleen and kidney of larger fish were fixed in the same manner. Whole fish or individual tissues were embedded in paraffin, sectioned to 5 µm and stained with hematoxylin and eosin (H and E).

Tissues for electron microscopy were fixed in 2.5 % glutaraldehyde in 0.1 M cacodylate buffer (pH 7.4), post-fixed in 1 % aqueous OsO_4, embedded in plastic and sectioned. Sections were stained with uranyl acetate and lead citrate, observed and photographed with a Philips EM 400 electron microscope.

Transmission Trials

Iridovirus Trials : Two sturgeon from a population, shown to be infected with the iridovirus by histological and electron microscopical examinations of the gills, were placed in the upper section of a 70 L trough with running 17 C well water. In the lower section of the trough, separated from the upper section by a screen, 10 healthy sturgeon were added. In a second identical trough, the upper section was left empty, while the lower section contained 10 healthy sturgeon.

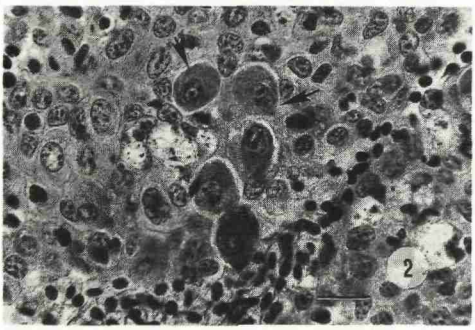
Adenovirus Trials : Eight juvenile sturgeon (10 g) from a control population were inoculated intraperitoneally with 0.1 ml of filtered (0.45 µm) homogenate made from the intestine of an infected sturgeon. An equal number of control fish were inoculated only with a saline solution. Both the experimental and control groups of fish were maintained in 20 L aquaria receiving 16 C well water.

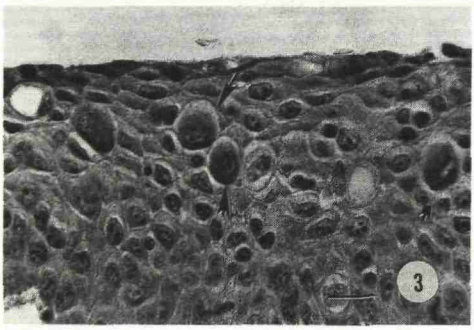
In transmission studies with both viruses, tissues from both mortalities and survivors were examined following histological preparation and staining with hematoxylin and eosin.

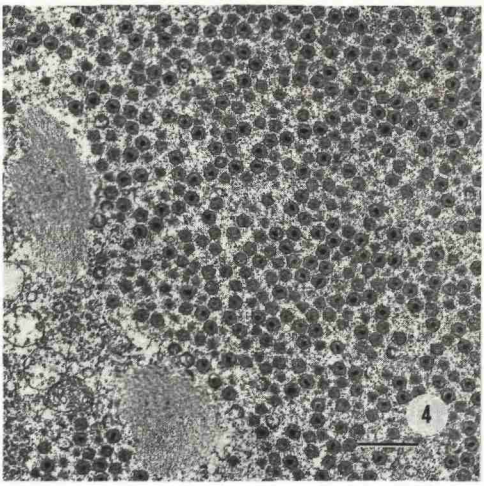
RESULTS

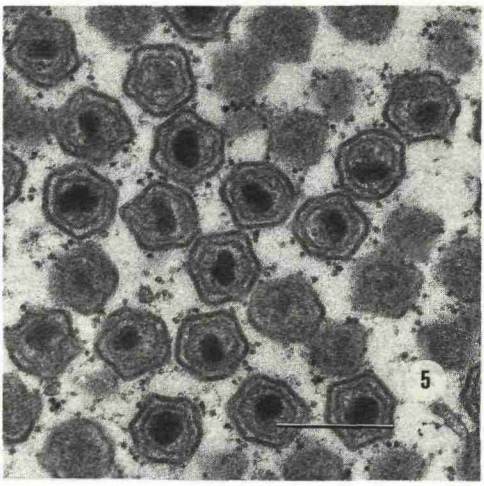
The appearance of fish with either virus as observed in outbreaks at commercial farms were similar. Affected juveniles are often the weakest fish in the population. These fish exhibit weight loss, drop to the bottom of the tank, cease to swim and eventually die. Gill pallor and hyperplasia may also be evident.



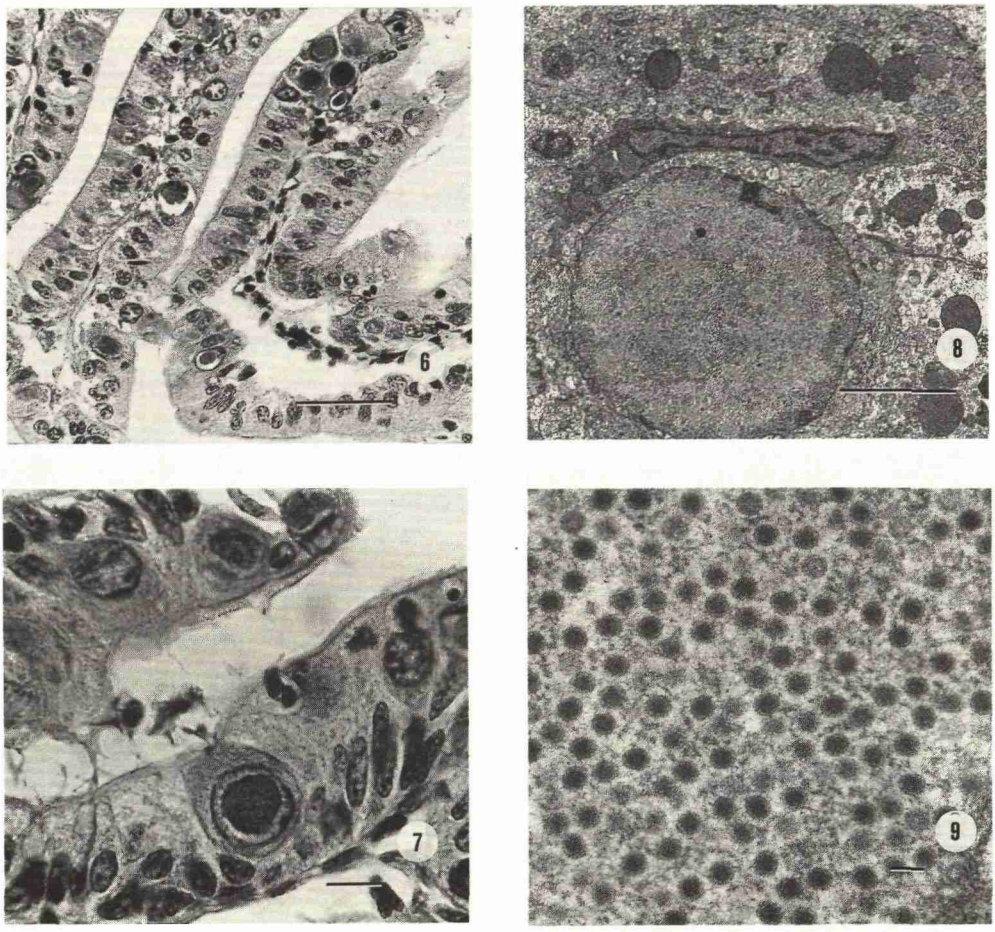








Figures 1 - 5 - Iridovirus infections in juvenile white sturgeon.
(1) Enlarged and deeply staining cells in the epithelium of the gills. Bar = 100 µm
(2) hyperplastic respiratory epithelium. Enlarged virus-infected cells. Bar = 20 µm.
(3) Affected cells in the epidermis. Bar = 20 µm. H and E stains.
(4) Numerous virions in the cytoplasm of an infected cell of the gill epithelium. Bar = 1 µm.
(5) Virions with dense cores and double capsid structure. Bar = 500 nm.



Figures 6 - 9 - Adenovirus infections in white sturgeon.
(6) Straight intestine showing mucosal epithelial cells with hypertrophied nuclei. Bar = 50 µm.
(7) nuclei in various stages of enlargement. Bar = 10 µm. H and E stains.
(8) Virus infected cell with enlarged virion filled nucleus. Bar = 100 µm.
(9) Virions with dense inner cores. Bar = 100 nm.

Complicating infections with fungi and gill bacteria are found with both types of viral infections but are more common with the adenovirus. Internally, fish infected with either virus have little to no body fat, pale livers and lack of ingesta.

Microscopic Signs of Disease

Iridovirus : Histological examination of the gills and skin showed numerous hypertrophied, and occasionally intensely basophilic cells in the gill epithelium and epidermis (Fig. 1 - 3). Crystalline rod-like bodies were observed in the cytoplasm of some infected cells and the nucleus was slightly enlarged (Fig. 2). Cells in proximity to the heavily stained cells were also affected as shown by cell swelling (Fig. 2).

In severely infected fish, many of the Malpighian cells of the epidermis were infected as judged by their appearance on light microscopy (Fig. 3). In light infections, the gills showed only a moderate hyperplasia with occasional basophilic staining cells (Fig. 1). In more advanced gill lesions however, hyperplasia of the respiratory epithelium was followed by necrosis of the of the pillar cells lining the lamellar vascular channels. Small hemorrhages were often associated with these vascular lesions. Hypertrophic, basophilic cells were observed but less frequently in these fish.

Enlarged cells containing numerous virions were seen on electron microscopy of gill tissues (Fig. 4). Virions were abundant in the cytoplasm of degenerating cells near the epithelial surface and accumulations of fibrillar material were present (Fig. 4). Mature virions were 262 nm in diameter as measured from one side to another and approximately 300 nm from vertex to vertex (Fig. 5). There was an internal nucleoid that surrounded a circular to bar-shaped electron-dense core. This internal capsid or nucleoid had a diameter of 184 nm (Fig. 5).

Adenovirus : Histological examination of midline sagittal sections of whole fish showed the principal sites of infection were the mucosal epithelial cells of the straight intestine and the spiral valve (Fig. 6). Affected cells had enlarged nuclei with a clear halo near the nuclear membrane (Fig. 7). Nuclear changes ranged from early, slightly enlarged stages to diameters 10 times normal (Fig. 7).

Electron microscopy showed that affected nuclei contained numerous virions (Fig. 8). Virions were detected only in the nucleus and were devoid of any envelope. A viral capsid with a mean diameter of 74 nm surrounded an electron dense inner core of 47 nm (Fig. 9).

Transmission Trials

Iridovirus : Infections were detected in white sturgeon that were held below infected sturgeon but not in control fish. Four of the 10 sturgeon exposed to infected fish had died by 40 d post exposure. Evidence for virus infection on histological examination of two of these fish that died during the study and among four of six fish which were sacrificed at the end of the study (40 d) was found. No control fish died during the study nor was there histological evidence of infection in this group upon examination of all survivors at the end of the study.

Adenovirus : There were no mortalities in the experimental or control groups over a 30 d period. Infections were however, detected in five of eight white sturgeon injected with filtered homogenates from infected sturgeon. The infections were detected in the mucosa of the spiral valve and hepatocytes. The number of infected cells detected by histology were few compared to those detected in natural infections.

No evidence of virus infection was found among control fish.

DISCUSSION

Two viruses have now been described from white sturgeon (Hedrick et al., 1985; Hedrick et al., 1989). Both viruses were observed in juvenile sturgeon experiencing above normal losses. Of the two agents however, the iridovirus seems most pathogenic and it is suspected as being a major cause of mortality in cultured white sturgeon. In

one commercial farm, 95 % of 200,000 juveniles were lost over a 4 mo period. Abundant numbers of infected cells were present in the gills and skin of dead and moribund fish from this site and there was no evidence of other causes of mortality. Infections of the skin and gills could compromise the osmoregulatory capabilities of affected sturgeon. These changes, if severe, could cause death without systemic spread of the virus. Transmission studies clearly demonstrated the ability of the virus to spread via the water from infected to uninfected fish. Another potential method of spreading the virus would be with wet equipment (nets, brushes, boots etc.) between tanks or fish rearing facilities.

Clinical signs of fish infected with either the iridovirus or adenovirus are similar. Infected fish are usually emaciated, lethargic and sink to the bottom of the tank. No other external signs may be noticed and detection of infection requires careful laboratory inspections including observations of stained tissue sections. The cellular changes induced by the two viruses are considered unique (pathognomonic) and suffice for a presumptive diagnosis. The iridovirus induces cellular enlargement (principally cytoplasmic) and strong basophilia (Fig. 1 - 3). The adenovirus induces nuclear hypertrophy (Fig 6. and 7). The tissue tropism displayed by the two viruses is also unique. The iridovirus to date has only been found in the epithelium of the skin and gills, while the adenovirus has been found in intestinal mucosa and occasionally liver.

Confirmation of infection with either virus is dependent on observation of characteristic virus particles by electron microscopy. Although we have made numerous attempts to isolate the viruses on primary and established cell lines from white sturgeon, we have had been unable to propagate the viruses in vitro. Until this can be accomplished our understanding of the two viral agents will be limited to field observations and experimentation with crude preparations derived directly from infected fish.

The source of both viruses is unknown but we suspect feral broodstocks since the virus has occurred in populations reared in isolated conditions. Examinations of a limited number of feral juvenile and adult white sturgeon however, has failed to show any evidence of infection and the potential impact of the viruses on these fish is currently unknown.

ACKNOWLEDGEMENTS

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AUTRES

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THE SUBSISTENCE LAKE STURGEON(*Acipenser fulvescens*) FISHERY OF THE INDIAN VILLAGE OF MUSKRAT DAM IN NORTHERN ONTARIO, CANADA

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RESUME

La communauté indienne du barrage de Muskrat a été fondée dans les années soixante sur le bord de la rivière Severn dans le nord-ouest de l'Ontario. C'était un site de campement traditionnel connu pour sa richesse en ressources sauvages dont l'esturgeon. La population de cette communauté a augmenté jusqu'à atteindre 220 personnes. Des tronçons de la rivière Severn avait été exploités dans le passé par une pêcherie d'esturgeon controllée depuis le sud.

Des informations historiques et ethnographiques ont été obtenues auprès des résidents lors d'interviews, de conversations ou en accompagnant des familles sur les sites de pêche d'esturgeon. Dans le cadre d'une étude destinée à quantifier l'utilisation par la communauté des ressources sauvages renouvelables, des informations sur les pêches vivrières ont été réunies pendant 2 ans.

Des données biologiques ont été obtenues sur une partie des captures de 1988; combinées à des estimations de l'effort de pêche et des sites de pêche, elles ont permis de décrire l'état de la pêcherie d'esturgeon et son importance dans l'économie de subsistence. On décrit l'utilisation de l'esturgeon à la fois comme ressource alimentaire et comme fonction sociale de la communauté. On a exploré le savoir traditionnel sur le comportement de l'esturgeon et sa biologie ainsi que son utilité pour la conservation des populations de poissons. Des suggestions sont faîtes pour que les communautés du nord contrôlent et gérent leurs stocks d'esturgeon. Ceci devrait être réalisé en reconnaissant la nécessité de conduire de telles études au niveau de la communauté et sous son contrôle, avec, si nécessaire, l'assistance de professionnels extérieurs compétents, plutôt que comme une réponse conventionnelle par les agences et institutions officielles.

Mots clés: *Acipenser fulvescens*, pêche de subsistence, biométrie

ABSTRACT

Muskrat Dam, an Indian community on the Severn River in northwestern Ontario, was founded in the 1960's at a traditional camping site noted for its abundant wildlife resources, including sturgeon, and has now grown to a population of 220. Parts of the Severn River, which connects a chain of lakes, have been exploited in the past by a commercial sturgeon fishery controlled from the south.

Historical and ethnographic information was obtained from residents in interviews, conversation, or when accompanying families to sturgeon fishing camps. Data on the native subsistence fishery were gathered for two years as part of a harvest study which quantified the use of all wild renewable resources by the community.

Biological data were obtained for a portion of the sturgeon harvest in 1988 and are combined with information on fishing effort and location to describe the state of the sturgeon fishery and its importance within the larger subsistence economy. Use of the sturgeon both for domestic food and for community social functions in described. Traditional indigenous knowledge on sturgeon behaviour and biology, and its usefulness for community-based conservation of sustainable fish populations, is explored. Suggestions are made for northern communities to monitor and manage their sturgeon stocks. This will likely have to be done in a fashion that recognizes the need for conducting studies at the community level under community control, with the assistance of sensitive outside professionals where required, rather than as conventional regulatory responses by established agencies an institutions.

Key-words: *Acipenser fulvescens*, subsistence fishery, biometry

INTRODUCTION

Purpose of the Study

This paper describes the history and present status of an indigenous lake sturgeon (*Acipenser fulvescens*) fishery in a small Indian community in the boreal forest region of the Hudson Bay watershed of Northern Canada. It presents some preliminary results from part of an extensive study of wildlife and renewable resource use by the community,and makes some comments towards exploring viable management. To our knowledge, no such detailed description of a subsistence *A.fulvescens* fishery has been published before, even though dozens of Native communities across the species' range fish for sturgeon, and probably consume more fish than sportsmen and commercial fishing combined.

Context

Northern Ontario is the home of the 20,000 Cree and Ojibway spesking Native people of the Nishnawbe-Aski Nation. Most live in villages of a few hundred to one or two thousand people on the small reserves lets them when the Canadian government had them forfeit legal title to their traditional lands in treaties signed in 1904 and 1929 (fig. 1).

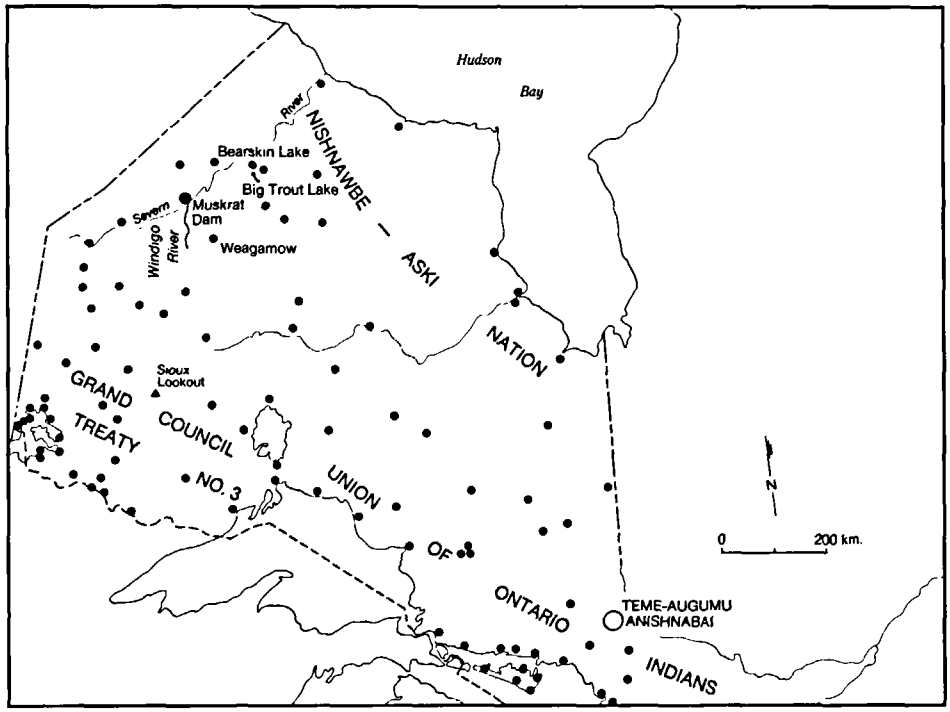


Figure 1- Indian Communities of Northern Ontario, Canada.
The range of Acipenser fulvescens includes the entire area shown on the map, and many of the communities in the Nishnawbe-Aski Nation would engage in sturgeon fishing.

Rights to hunting, fishing, and trapping are, however, maintained on the land covered by the treaty, and products obtained bu these traditional activities, often at considrable distance from the communities, constitute an important component of the domestic economy. *Acipenser fulvescens* occurs in most of the large rivers across the Nishnawbe-Aski territory and is a preferred species for village domestic fishers whenerver accessible. The species is also used or has been used by Native fishers in Quebec, and across its range into western and northern Canada.

Access to these resources increasingly faces problems with competing white sportsmen, commercial fishing, mining and forestry developments, hydroelectrical generation, tourism, and the generally encroaching infrastructure of the southern industrial society. Wildlife regulatory agencies, which are typically centralized, bureaucratic, and embedded in European culture and western science, attempt to manage resources through various measures of legalistic enforcement.

However, they remain largely ignorant of the ways, knowledge, abilities and needs of these native communities. The last few years have witnessed enormous political activity on the part of the native First Nations, with considerable emphasis on aboriginal rights, and the devolution of control over the management of wildlife resources to the nations or their constitutional communities.

The concept of shared or community controlled management of wildlife, often termed cooperative management or "comanagement", has recently become a subject of considerable discussion (Alberta society of Professional Biologists, 1986), and the first innovative attempts at comanagement are now being evaluated (Pinkerton, 1989).

The information offered in this paper, which describes the domestic sturgeon fishery of a small native community, is provided in a context that seeks to include indigenous wisdom and knowledge about wildlife and ecology community management or comanagement of renewable resources. The information may initially seem of more ethnographic than biological interest, but represents the kind of information that must be assembled if community-centered management schemes are developed.

The Community of Muskrat Dam

Muskrat Dam is located on the Severn River in northwestern Ontario (fig.1), and is accessible only by air or special transport vehicles over winter trails. The nearest town accessible by road is Sioux Lookout, 350 km to the south. The community was founded in 1966 by a group from Bearskin Lake 80 km to the northeast. The site on Muskrat Dam Lake (60 km^2), one of a chain of lakes formed by the Severn River, was a traditional summering spot for earlier generations who moved in a seasonal pattern over a wide area in pursuit of their hunting, fishing, and trapping actvities.

The community had 100 people in 1975 when it was the subject of a very detailed study of wildlife use from an anthropological perspective (Winterhalder, 1977), and has now grown to about 180 people in some 45 households.

MATERIALS AND METIIODS

General Methodology

Data were gathered in a comprehensive recall harvest study of all wild products obtained by all resident members of the Muskrat Dam community based on six interview sessions at four month intervals from late August 1987 to September 1, 1989. Methodological accuracy was ensured by various means, most of which have been reviewed by Usher and Wenzel (1987). Weights were taken to 50 g accuracy on samples of harvested animals for as many species as possible to establish mean weights. Edible portions were derived by weighing the products from butchering, processing, and consumption of harvested species.

Sturgeon

Sturgeon data (weight, length, sex, age) are mostly from July and August 1988 (62 specimens) with six from august 1989. Percent edible weight is based on specimens butchered by community members themselves. Pectoral fin rays for aging were cut from fresh or frozen fins, air dried, and stored dry until sectioned. Fin ray discs were cut with an isomet saw, and polished with a metal-bonded diamont disc to remove saw marks, before being attached to microscope slides with LePage's 5 minute epoxy glue (which was found to have superior adhesive properties while remaining satisfactorily transparent). Sections were then thinned to 250 to 350 µm with a water-cooled Bueler Petro-thin diamond sectioning system.

Cultural and historical information was collected by taped interviews, in July 1989, with elders who were familiar with the researchers and the purpose of the harvest study. Skilled interpreters were used as necessary. Other information was assembled from observations, field notes, conversations, survey questionnaires, management interviews and land use mapping sessions from February 1987 until September 1989. A summary of comments from the most comprehensive statement is provided in the following chapter; none of its assertions has been contradicted by other fishers' statements and it can thus be considered as representative of community knowledge on sturgeon.

All data gathering was done with consent and approval of the Muskrat Dam Band Council and the Elders of the community.

RESULTS

History of the Muskrat Dam Fishery

Early History:

Residents of the present village of Muskrat Dam know that at least the remembered previous three generations of their people have lived along the Severn River. Muskrat Dam Lake had a plentiful population of sturgeon whose main spawning grounds were 8 km from the village site, below the falls of the large tributary Windigo River about one km before its confluence with Muskrat Dam Lake.

Up to 100 people from some six extended families from the same clan centered on Big trout Lake, 70 km to the northeast, would gather for about three weeks in the spring at a camping spot near the mouth of the Windigo River in late May or early June when sturgeon were about to spawn. Spears were fashioned from barbed, flattement nails fixed to poles with twine. Each man would spear about 20 to 30 fish over the spawning period, and this would result in a harvest of an estimated 250 fish, sufficient to feed the camp for the period. Traps had been used in the past, but seem not to have been used by the early part of this century.

This spring spawning fishery probably ceased in the 1920's when the use of nets became current, and were used by the small number of families who would camp along Muskrat Dam Lake in the summer. These were hand made and took considrable effort. A man and woman working together could produce a 7-10 m. long net about 2.5 m. deep from cotton twine in four to five days. Each family had only one short net which lasted through three seasons. Only sticks were available for floats. Sturgeon were then plentiful enough that a longer net was not necessary, and fishers could sit on the bank to await movement indicating that a sturgeon had been caught. Most sturgeon netting was done in the spring and early summer, with lesser activity in the fall. Few sturgeon were ever netted in the winter.

The Period of Commercial Fishing:

By the 1940's, manufactured nets, boats, and outboard motors had begun to replace the earlier fishing technology. In about 1951, a white commercial fisherman established a base at Muskrat Dam Lake, having obtained a sturgeon fishing licence from the provincial Department of Lands and Forests. He fished the lake for some five

or six summers, with the assistance of another white man and occasional hired Native assistants, although never any of the local people.

He was well outfitted, in a way that local people could not afford, and actively prevented local Natives from continuing subsistence sturgeon fishing for their families. The fish caught for sale were kept alive nin pens made of poles and boards across small bays or pockets of the shorline. This way fish could be kept in relatively good condition until a float plane arrived to take them out.

Local people eventually reacted against the commercial fisherman's monomoly and prevailed on provincial government authorities to reduce the area coverd by the commercial licence. However, fish stocks were so reduced by this time that the fisherman departed : the Muskrat Dam Lade sturgeon fishery had effectively collapsed.

A second commercial sturgeon fishery downstream on the Severn was managed through the Hudson Bay Company in Bearskin Lake. This employed a number of Bearskin Lake men seasonally from 1953 or 1954 until the early 1970's, although heaviest fishing was in the first four years. Small numbers of sturgeon are also taken in commercial fishing for whitefish and pickerel in the Severn waterway. Because of depressed prices, there has been little commercial fishing of any kind in the area in the past few years.

The Fishery at Present

Since the founding of the village in 1965, sturgeon fishing at Muskrat Dam is almost completely for domestic use. A few fish are sent as gifts to people elsewhere, while many fish are shared with those who do not or cannot set nets themselves.

Spearing or gaffing during the spawning run no longer occurs, although it was occasionally practised by some of the present fishermen until just a few years ago. Net fishing for sturgeon is now done in three areas :

1. The Windigo River (8 to 32 km from the village).

2. A widening of the Severn River about 33 km downstream from Muskrat Dam.

3. A small lake ("A sipquobag") 42 km downstream from the village on the Severn.

These three areas are bounded at each end by unnavigable rapids or waterfalls. The journey to each of them is lengthy, and undertaken in aluminum boats with outboard motors. The round trip to the Windigo site takes a minimum of 2.5 hours and 17 liters of gasoline. The journey down the Severn requires skilled navigation through several hazardous rapids and one or two portages; seven hours and 35 liters of fuel are required.

Nets are checked either by the owner, or by a designated friend and assistant. Most of the time, the fishermen work.d out of the village, or, when going down the Severn, stay only overnight. Because of the distance, the nets are only checked every 3 to 7 days. Families do, however, camp out for a few days throughout the open water season, and at this time will often stay at camps convenient to sturgeon fishing locations, allowing nets to be checked once or twice a day. Fishing gear is almost exclusively 8 or 9 inch (20.5/23.0 cm) mesh nets of 100 yards (92 m) length. Fishermen tend to use the same fishing grounds each year.

Sturgeon caught at camps are either eaten immediately, or kept alive on tethers fastened to poles on shore until camp is broken. In 1988, eleven fishermen fishing at the two locations on the severn caught 330 fish, while seven men on the Windigo River caught only 38. An additional two fish were caught in general fish nets on Muskrat Dam Lake in late winter. Notes on effort have not yet been analyzed to determine the relative productivity of the two sites, although the downstream site on the Severn appears to yield the most fish. Results for the summer season up to Sept. 1 in 1989 show that 10 men on the Severn caught 196 fish while four on the Windigo caught 53. This latter figure included a substantial catch by a single individual who had not fished

the year before. In 1988 fishing commenced in late May and continued until July 10, when most nets were pulled for cleaning and drying. Fishing then resumed in mid August. Only one fisherman kept a net out as late as October.

The community of Muskrat Dam is renowend for its ability to stage large events and arrange banquets for considerable numbers of guests. For eight consecutive years it has hosted the Northern Elders Feast for some 100 to 200 guests for three or four days. In 1987 and 1988, 40 to 50 sturgeon were caught specifically for the feast. The feast was not held in 1989, and this should have resulted in decreased effort and a smaller catch.

When asked about the health of sturgeon populations in the area, most fishers stated that they felt there had been a decline in numbers, particularly on the Windigo River. There was less agreement about the situation downstream on the Severn, although opinion tended to the view that stocks have decline. All agreed that few fish have survived the commercial sturgeon fishing of the 1950's in Muskrat Dam Lake.

Biological Data

Table 1 shows a few preliminary data on fish caught in 1988. Sample sizes are admittedly small, but it is evident that the fish now being caught are fairly small and perhaps near the minimum size for reproductive maturity.

Table 1- Some bioligical characteristics of sturgeon caught by Muskrat Dam fishermen in 1988.

Parameter	Mean	Range (min/max)	Sample Size
Kept fish			
Weight (g)	5,090	1,900-32,350	50
fork length (cm)	85	66-138	49
minimum fork length of gravid females (cm)	88		3
male:female	22:17	39	
age (years)	21.4	14-31	24
Released fish			
weight (g)	1,075	700-1,650	6
fork length (cm)	58	40-66	6

The Sturgeon Fishery in the context of Community Harvesting Activity.

Sturgeon fishing and moose hunting require strength and are only undertaken by adult men, while general net fishing is one primarily by older women as well as a number of younger men and women. Only one woman, a particularly active harvester, reported fishing for sturgeon, and then only for three days in the two year survey period. Table 2 shows the number of able-bodied adult males who engaged in various

harvesting activities, irrespective of success, over a one year period. Some 38 % of these men, or 18 people, put out sturgeon nets of their own at least once. Of these, fifteen also put out nets for other fish species and eleven angled.

Table 2- Harvesting Activity of Adult Males.
Number of able bodied adult males (excluding high school students) engaged in harvesting for one year period beginning Dec. 15, 1987.

Harvest Category	Harvesters	
	Number	Percent
Waterfowl	46	96
Grouse and hare	46	96
Furbearers	37	77
Moose and caribou	34	71
Angling	33	69
General net fishing	27	56
Sturgeon fishing	18	38
Inactive	2	4
Total	48	100

There is a high degree of specialization in harvesting within the community : some individuals are skilled and highly knowledgeable trappers of marten, others concentrate on moose, and at least two individuals particularly favour sturgeon. It would be a mistake, however, to single out individuals, as so much of their harvest is shared with other members of the community.

By community decision, wild meat cannot be sold, as it is felt this would erode the system of reciprocal sharing that prevails, and the care and concern for other people's welfare that this constitutes. On one occasion, two sturgeon with a combined weight of 12,8 kg were observed to be shared amongst 5 households with 15 people in total. It is thus more meaningful to regard harvesting for food at the level of the total community harvest.

Table 3 summarizes the contribution of different harvested species to the total supply of edible wild meat to the community for a one year period. Sturgeon are almost invariably boiled, and only the entrails, fins and tail are discarded; there is much to be eaten from the heads, and the skin is also savoured. Only 15 % of the weight was discarded, the lowest loss for any of the species on the list. This is also a conservative figure, since some portions of the intestinal tract are occasionally eaten, but were not considered here.

Sturgeon accounted for 7.0 % of the total edible weight of meat, and 19.6 % of the edible weight of all fish combined. Only moose, whitefish, and sucker accounted for larger contributions to the community meat stock, while all species of ducks combined weighed only slightly less.

Muskrat Dam residents, as members of a hunting society, have very discriminating palates for meat. Sturgeon consistently ranked with moose and beaver as the most popular meats when individuals were asked to give their taste preferences. Sturgeon are thus so avidly pursued because they are deemed particularly delicious.

Table 3- Complete Harvest of Edible Wildlife Species for one year, ranked by percentage contribution of species group to total edible weight (Ed.Wei.), for period beginning Dec.15,1987. Mean weights and edible portions (Ed.Por.) derived from weighed samples except as indicated.

Species			Number		Species weight (kg)		Total edible weight	
			Taken	Used	Whole	Ed.Por.	kg	%
Moose	*Alces alces*		40	40	-	227.28[1],[3]	9,091	39,9
Lake whitefish	*Coregonus clupeaformis*		3,605	3,605	0.89	0.68	2,774	12.2
White sucker	*Catostomus commersoni*		5,342	3,098	1.16	.65	2,014	8.8
Lake sturgeon	*Acipenser fulvescens*		410[2]	370	5.09	4.33	1,602	7.0
DUcks			3,282	3,282	0.87[1]	0.48	1,575	6.9
Beaver	*Castor canadensis*		194	147	-	8.09	1,189	5.2
Canada goose	*Branta canadensis*		399	399	4.77	2.50	997	4.4
Walleye	*Stizostedion vitreum*	net	1,342	1,342	0.83	0.46	617	3.7
		angling	-[4]	648	0.68	0.35	227	
Northern pike	*Esox lucius*	net	2,178	566	2.91	1.08	611	3.0
		angling	-[4]	71	2.91[5]	1.08	77	
Muskrat	*Ondatra zibethica*		779	670	1.19	0.74	449	2.0
Grouse			1,230	1,230	0.53[1]	0.36	442	1.9
Caribou	*Rangifer caribou*		6	6	81.80	49.08	294	1.3
Snowshoe hare	*Lepus americana*		236	236	-	0.88[1]	207	0.9
Goldeye	*Hiodon alosoides*		336	336	0.64	0.48[6]	161	0.7
Snow goose	*Chen hyperborea*		99	99	-	1.63[1]	161	0.7
Common loon	*Gavia immer*		111	111	-	1.16[1]	128	0.6
Yellow perch	*Perca flavescens*		327	202	0.54	0.28[7]	56	0.2
Mariah	*Lota lota*		1,292	77	1.31	0.66[4]	51	0.2
Miscellaneous waterfowl			132	128	-	-	75	0.3
Total all fish species							(8,190)	(35.8)
Total all species							22,798	99,9

(1) Mean weight or percent edible weight from JBNQNHRC,1982.
(2) Calculated from "Number Used" assuming 10% release.
(3) Weights adjusted for sex and age composition of species.
(4) Fish released or abandoned not known.
(5) No direct weights. Mean edible weight of retained fish assumed similar to netted fish.
(6) Edible portion assumed similar to whitefish.
(7) Edible portion assumed similar to walleye.

Indigenous Knowledge and Conservation

Context:

Indigenous management systems, based on a variety of different kinds of acquired and shared information used in traditional decision-making processes to provide prescriptions for conservation practices, will likely provide the basis for effective, innovative management schemes in northern Canada (Usher, 1987). Here we provide some observations on the knowledge and values expressed by Muskrat Dam harvesters that indicate the possibility of viable community-controlled management schemes.

Values:

People in Muskrat Dam have a deep faith in the Creator who gave them their land and the animals on it for their survival. This wealth must be used properly, and only as much as needed is taken, in accordance with the Creator's designs. In a sense, they have obligations as custodians, and must protect the land. Part of the opposition of the community to recent mining and dam proposals comes from this sense of obligation to flufill the Creator's trust in them as the people chosen to live on this land.

Conservation Practices:

Muskrat Dam residents who specialize in the various harvesting activities acquire a wealth of ecological and behavioural knowledge about wildlife species. They are familiar with such things as age structure and growth rates of populations, relationships to food and habitat, the dynamics of disease, condition, the principles of sustained yield, and the choice of mechanisms to adjust harvest levels to population dynamics. When general commercial fishing was practised in Muskrat Dam, decisions were made how long to fish certain lakes, and then how long to leave them fallow. This is a different approach than government agency prescriptions aiming for an annual uniform sustained catch, but perhaps more manageable when effort, expenses, and cultural preferences are considered.

A code of conduct exists regarding the setting and checking of nets, disposal of fish carcasses, location of nets, respect for traditional fishing territories, the kinds of activities that are permissible in key fishing areas, and the introduction of contaminants to water bodies.

With sturgeon, smaller fish, below about 60-65 cm fork length, are invariably released alive (Table 1), because they have proportionately less meat than larger fish and it is felt they should be left to grow and fatten up. The community itself has chosen not to continue with commercial sturgeon fishing, even though the commercial licences they hold allow for a catch of 500 kg on the Severn. Fishermen, in common with all harvesters, continually discuss their success and offer observations they have made during activities. There is concern about the health of the sturgeon population and it is now a matter of discussion in the community. There is also recently much worry about contaminants in wildlife.

Knowledge About Sturgeon:

The information below is selected from interviews with an experienced trapper and fisherman whose wisdom and dnowledge are recognized by the community.

He estimated that the mean fork length of sturgeon caught is 82 cm; in fact it is 85 cm. He considered the population of spawning females to be largely within the 80-90 cm length, with a minimum of 77 cm. Our own information so far consists of only 3 fish, with the smallest 88 cm (table 1). He felt that fish of different sizes sorted themsleves to different water depths and current velocities, and that this accounted for some of the characteristics seen for different catches. He considered age a difficult matter to reckon, but considered that fish might have to live as long as 30 years to spawn.

Growth is recognized as being very slow. He based this mainly on observations made on a different river where he would visit the same spot after many intervening years, and find that the fish had increased little in size with time. Replenishment of an overfished area is known to be problematic and slow. The commercial fisherman on Muskrat Dam lake in the 1950's used about 2750 m of nets, and this intensity of fishing was obviously unsustainable. In his own experience as a commercial fisherman in another community, he would spread his activity over short intervals amongst a number of widely separated locations.

He considers sturgeon to be rather sedentary. This conclusion is based on the fact that the sturgeon from different areas have distinct differences in appearance, and fish are rarely noted in a noncorresponding area. Such differences include bodily dimensions, fat deposition, head shape, colour intensity, and colour patterns, as well as taste. In addition, depopulated spots remain empty of fish; they do not appear to move in from elsewhere.

Sturgeon from different areas have different diets, as revealed by gut contents, and this may account for differences in taste, as well as their preference for different locations. General availability of food, as well as food quality, may influence the growth rate of fish. Sturgeon observed just before freeze up are very inactive and a few fish netted at break up were covered with mud. Would this indicate some sort of torpor?

He maintains that sturgeon are known to be very susceptible to stress and disturbance. On one occasion he observed a large hole in a net that indicated a large sturgeon had escaped from a net. Some time later, a large fish bearing old scars from a previous netting was taken in a net some distance away below an intervening waterfall and rapids. The physical characteristics of the fish corresponded to those of the population from where the earlier fish had escaped. Excess motorboat traffic is widely held to disturb sturgeon.

In addition, they appear to have highly developed chemosensivity. Campfire ashes, forest fire smoke, mineral ore drilling and associated activities, fish wastes, dead fish, wash water, animal fats, and petroleum products are all considered detectable by or potentially harmful to sturgeon. Several informants independently concurred on a correspondence between the disappearance of sturgeon and mine exploration at a particular location. The white commercial fisherman operating at Muskrat Dam from 1951-1956 sometimes used gasoline to drive sturgeon into nets; this action was condemned by informants and such practices are unknown in the community.

Threats to the Community Fishery and Steps to a Resources Policy.

The community of Muskrat Dam has reacted strongly to incursions into their land area, and have consistently undertaken measures to develop concrete policies towards resource use and conservation.

A 1973 proposal to place a penstock hydroelectric generator at the Windigo Falls (the spawning grounds of the commercially depleted Muskrat Dam Lake sturgeon population) was declined by the elders of the village because of their concern for the integrity of the spawning grounds. In 1986, plans for an extensive harvest and land use study were put forward, resulting in the work presented here. In October, 1988, an active confrontation was found necessary to force a gold drilling camp from the Windigo Tiver, and persuade the company to undertake negotiations with the community.

In early 1989 a survey was completed of resident opinions and expectations towards formulationg a policy on resources; concern for sturgeon was mentioned in many of these interviews. In May, 1989, leaked government documents confirmed plans by a private consortium to build a series of 12 flow-of-river hydroelectric stations at the major waterfalls and rapids on the Windigo and Severn. The community faced this crisis by organizing other communities on the river to form the "Severn River Coalition", which will be conducting a referendum on the question of declaring the river system a "dam-free zone". Publicity statements issued by the Band Council specifically mentioned concern for sturgeon spawning areas as one reason for opposing the dams.

DISCUSSION

Sturgeon have an unusually important place in the subsistence food economy of Muskrat Dam. At 7 % of total edibe weight of wild meat, this dependence is far greater than other communities in Ontario such as Webequie where it is 0.2 % (Hopper, 1989), or two communities near Sudbury (Manitowabe, pers. comm.), as well as Quebec (JBNQNHRC, 1982) where it is 0.7 %.

The demise of the commercial fishery at Muskrat Dam Lake follows an all too familiar pattern in Ontario (Harkness and Dymond, 1961) and parallels the collapse of the commercial sturgeon fishery at Weagamow Lake, 50 km to the southeast, between 1950 and 1961 (Ontario Dept. of Lands and Forests, 1971). Recent evidence seems to indicate that at least some native sturgeon fisheries were viable in the past (see Holzkamm and McCarthy, 1988, for a study based on historical isinglas returns), but were too precarious to withstand white incursions (Tough, 1984).

The Ontario government has in the last few years sponsored considerable biological research into the state of major sturgeon populations of northern Ontario rivers, particularly those flowing into James Bay (Oliver, 1987; Threader and Brousseau, 1986), and these results provide us with the biological understanding and technical possibilities for effecting positive management options elsewhere in the province. However, there is a corresponding gap in our understanding of the subsistence economies of native communities associated with the streams bearing sturgeon populations in the north, and a lack of consistency and good faith in the actions of government agencies that destroys their credibility in the eyes of native communities. For example, elders at Muskrat Dam remember only too painfully the harassment they used to suffer at the hands of conservation officers when their nets were confiscated and they were forbidden to fish. Now the same agency is seen to be too close to developers, such as the group wanting to build dams on the Severn River.

Native communities must be given the option of producing and administering their own management schemes as part of an overall process of moving toward self government. An acceptance of aboriginal rights and the potential of indigenous knowledge within a framework of native moral and spiritual values would be necessary before the accumulated biological studies could be used towards the formation of management plans that are acceptable to communities.

The importance of spiritual values cannot be discounted. Sensitive studies have now shown that moral values effectively determine indigenous management systems that can yield results very similar to government quotas based on scientific population studies, at least for beaver (Feit, 1986). In addition, analyses of other native subsistence fisheries have shown that practices and technology are finely tuned to ecological considerations and can result in successful fisheries over the long term (Berkes, 1977). Other studies have revealed the possibilities for some internal restructuring of problematic portions of subsistence fisheries while still respecting the needs of the communities (Berkes, 1979). Of course, such problems are not necessarily unique to native fisheries and pose larger questions for developing more sophisticated and sensitive methods for studying the social relationships of fisheries (Vanderpool, 1987).

The community of Muskrat Dam is prepared to assess their sturgeon fishery. The incomplete information here suggests a decline in local strugeon populations. However, the community can be credited for three constructive actions. It has itself initiated comprehensive studies of its wildlife use and emphasized the state of its sturgeon fishery. At the same time, it has surveyed community members as the firest step in formulationg its own resource policies so that the community can govern its own land. And, it has actively defended and protected its waterways to maintain the ecological integrity of the areas that are entrusted to through the Creator.

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QUELQUES REFLEXIONS SUR LA PRISE EN COMPTE DE L'INCERTITUDE DANS LA POLITIQUE DE GESTION D'UNE ESPECE SAUVAGE: L'EXEMPLE DE L'ESTURGEON OUEST EUROPEEN (*Acipenser sturio*)

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RESUME

La gestion des espèces sauvages est affectée par de grandes incertitudes liées à la durée des périodes en cause, aux insuffisances des connaissances dans le domaine, et au caractère fondamentalement aléatoire de certains phénomènes. Nous montrons qu'un modèle semi-markovien peut fournir une trés utile structure d'analyse des décisions de gestion lorsque l'incertitude joue un r le critique. Ce cadre est appliqué à la gestion de l'esturgeon, une espèce menacée en Aquitaine. Nous montrons pourquoi la gestion des espèces menacées est fondamentallement une politique de minimisation des pertes, et nous procédons à quelques investigations sur les stratégies de recherche et de gestion.

Mots clés : Espèce menacée, *Acipenser sturio*, Incertitude, Modèle semi-markovien, Stratégie de recherche, Valeur économique

ABSTRACT

Wild species management typically involve substantial uncertainties due to the long time frame for analysis, the lack of knowledge in the field, and the intrinsically stochastic nature of some phenomenons. We show that a semi-markovian model can provide an insightful structure for analysing management policy decisions where uncertainty plays a critical role. This framework is applied to the management of the sturgeon, an endangered specie in Aquitaine. We show why management of endangered species is intrinsically a policy of loss minimization and make some investigations on research and management strategies.

Key-words : Economic Value, Endangered Species, Research Strategy, Semi-markovian Model, *Acipenser sturio*, Uncertainty

INTRODUCTION

La gestion des espèces sauvages est typiquement affectée par de grandes incertitudes. La durée des périodes en cause, l'état des connaissances dans le domaine, le caractère aléatoire d'événements ayant un impact determinant sur ces espèces, constituent autant de facteurs d'incertitude dont la combinaison peut décourager toute ambition prospectiviste. Et pourtant, au jour le jour, des décisions sont prises, retardées ou annulées avec des effets certains mais difficiles à prévoir. Dans un contexte de ressources rares, ou les projets soumis à des financements publics notamment doivent affronter une dure compétition, il devient indispensable de cerner mieux les zones d'incertitude et d'aléa qui caractérisent les politiques envisagées.

Les modèles déterministes de gestion sont bien s r ici totalement inopérants et l'on se sent très démuni pour aborder de tels problèmes. Par essence, les situations incertaines sont difficiles à structurer. Nous voudrions ici montrer comment une approche probabiliste de nature markovienne peut aider à cette structuration et soutenir ainsi la réflexion de la communauté impliquée dans cette gestion (scientifiques, services gestionnaires, professionnels, décideurs politiques).

L'ambition de ce papier n'est pas la recherche d'une politique optimale, mais la construction d'un cadre formel d'analyse. L'outil à mettre en oeuvre doit garantir une richesse et une souplesse d'approche de façon à conserver les aspects spécifiques et critiques du problème étudié.

Pour l'esturgeon Ouest Européen, *Acipenser Sturio* qui est ici au centre des investigations, on retrouve bien les grandes sources d'incertitude évoquées précédemment :

- Politique de long terme. Une femelle de cette espèce atteint sa maturité sexuelle entre 20 et 22 ans (MAGNIN 1962).
- Connaissances incomplètes. On ignore encore de trés nombreux aspects de développement biologique du sturio (par ex de 0 à 3 ans croissance, survie...). Les biotechniques sont en plein essor mais les essais de reproduction artificielle et d'alevinage s'ils ont progressé significativement ne sont pas encore un réel succès. Par ailleurs, on ignore le niveau des stocks, et la limite à partir de laquelle on tend irrémédiablement vers la disparition de l'espèce locale (Trouvery et al., 1984).
- Interventions de facteurs aléatoires. Nous pensons par exemple à l'intervention de pollutions accidentelles dans le milieu, ou encore à des incidents graves au stade de la reproduction artificielle ou de l'alevinage (Beddington et al., 1977).
- Présence de facteurs exogènes trés prégnants tels que la pratique de la pêche commerciale en mer qui peut limiter considérablement les taux de retour (Castelnaud et al., 1985).

Il faudrait encore tenir compte outre des incertitudes biotechniques, des incertitudes économiques. En effet, l'espèce fut autrefois source de richesse, les politiques actuelles visent à dépasser les valeurs symboliques et d'existence pour atteindre un stade dégageant un surplus économique marchand. S'introduisent donc ici des inconnues de nature économique : co t de production des alevins, substituabilité ou complémentarité entre aquaculture d'espèces exotiques (*Acipenser baeri*) et restauration de l'espèce locale dans le milieu naturel, évolution de la valeur de la ressource[1].

Face à de telles incertitudes, quelles sont les questions qui se posent ?

On peut d'abord chercher à évaluer les politiques passées. A un certain moment la population d'esturgeon bien qu'abondante a donné des signes de fléchissement. Quels auraient été, appréciés sur une longue période, les co ts et les bénéfices d'une politique d'intervention a ce stade compte tenu des aléas qui caractérisent le domaine. Comment se situent ces co ts et bénéfices par rapport à ceux auxquels on est confronté aujourd'hui.

On peut également s'intéresser à la réduction de l'incertitude. Face à des structures de probabilité variables qui conduisent à des résultats économiques différents, il peut être avantageux de réduire l'incertitude. Les recherches scientifiques contribuent entre autres à l'amélioration de l'information. Les co ts de ces recherches peuvent être confrontés à la valeur de l'information crée.

On peut aussi chercher à élaborer des stratégies de gestion qui éloignent d'états conduisant avec des probabilités significatives vers une disparition de l'espèce.

Ces questions n'épuisent pas bien entendu le problème, mais elles paraissent importantes. Le modèle est susceptible d'apporter des éléments de réponse pour chacune d'entre elles. Nous n'évoquerons ici, et encore superficiellement, que ses possibilités au regard du rendement des programmes de recherche et du choix d'une stratégie d'action.

LES PRINCIPES D'UNE APPROCHE SEMI-MARKOVIENNE

Essentiellement un modèle markovien est constitué d'une matrice ou processus comportant des probabilités de transition. Ces probabilités p_{ij} indiquent la vraisemblance qu'étant dans un état i le système se trouve dans un état j à la période suivante. La probabilité de passage de l'état i à l'état j dépend uniquement de l'occupation de l'état i et non de l'histoire des évolutions qui ont conduit à l'occupation de cet état i. C'est là d'ailleurs l'hypothèse centrale de Markov.

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[1] Sur la relation entre prix de marché et exploitation d'un même espèce avec écloserie, et compétition entre aquaculture et pêche commerciale en mer, on pourra consulter (Anderson, 1985)

Le modèle de markov classique a la propriété qu'une transition est réalisée à chaque période. Certes la transition peut ramener à l'état déjà occupé, mais il y a tout de même transition. Pour les problèmes qui nous intéressent et qui se caractérisent par des phénomènes d'inertie, cette propriété est trés gênante. Aussi allons nous nous tourner vers des modèles plus complexes mais plus adaptés appartenant à la classe des modèles semi-markoviens. Dans ces modèles, il est possible de définir des processus tels que la durée entre deux transitions soit plus importante que le pas de temps et même tels que la durée d'occupation d'un état donné soit lié non à l'état, mais à la probabilité de transition.

Explicitons ceci. Soit p_{ij} la probabilité qu'un processus semi markovien qui vient d'occuper l'état i passe à l'état j à la prochaine transition. On a classiquement :

$$p_{ij} \geq 0 \quad (i = 1,2,...,N \text{ et } j = 1,2,...,N) \text{ avec } \sum_{j=1}^{N} p_{ij} = 1 \quad (i = 1,2,...,N)$$

où N est le nombre d'états du système. Ici, après avoir sélectionné i, mais avant de transiter vers j, le système occupe l'état i pendant une durée τ_{ij} . Ces temps d'occupation sont des valeurs entières positives gouvernées par une fonction de probabilité h_{ij} telle que $Prob(\tau_{ij} = m) = h_{ij}(m)$. On admet que les moyennes des temps d'occupation sont finies et que τ_{ij} est au moins égal à une unité de pas de temps.

Ainsi, il faut spécifier N^2 fonctions de durée d'occupation pour décrire complètement le processus semi-markovien.

Après avoir occupé l'état i durant τ_{ij} instants, le processus transite vers j et immédiatement sélectionne une nouvelle transition vers k au moyen des probabilités de transition. Il choisit alors une durée d'occupation τ_{jk} de l'état k selon la fonction de probabilité $h_{jk}(.)$ au bout de laquelle il effectuera une nouvelle transition.

L'introduction du temps d'occupation d'un état donné nous assure la possibilité de tenir compte de l'inertie des phénomènes, mais aussi de situations complexes ou une forte probabilité de transition d'un état i vers un état j peut être combinée à une occupation trés longue de l'état i par exemple alors que simultanément, une faible probabilité de passage de i vers k peut être associée à un faible temps de présence dans l'état i.

La dimension économique sera introduite en adjoignant au système des co ts et bénéfices pour chaque probabilité de transition. Compte tenu de la dimension temporelle des problèmes, nous incorporerons également un facteur d'actualisation.

L'algorithme de calcul dont nous nous inspirons est du à Howard (1971). Le principe est le suivant :

Soit $v_i(t,\alpha)$ l'espérance actualisée de la valeur dégagée par le système pour un intervalle de temps t si l'on se trouve dans l'état i au début de cet intervalle. Notons $b_{ij}(\tau,\alpha)$ la valeur actualisée du gain généré par le rendement b_{ij} (que nous considérons comme constant par période) pour un temps d'occupation τ de l'état i avec comme successeur l'état j :

$$b_{ij}(\tau,\alpha) = \int_0^{\tau} b_{ij}\, e^{-\alpha\sigma}\, d\sigma$$

L'équation livrant l'espérance de la valeur actualisée dégagée par le système est la suivante :

$$v_i(t,\alpha) = \sum_{j=1}^{N} p_{ij} \int_t^{\infty} h_{ij}(\tau)\,[b_{ij}(t,\alpha)]\,d\tau + \sum_{j=1}^{N} p_{ij} \int_0^{t} h_{ij}(\tau)\,[b_{ij}(\tau,\alpha) + e^{-\alpha\tau} v_j(t-\tau,\alpha)]\,d\tau$$

On peut l'interpréter en remarquant que la première transition hors de l'état i peut intervenir avant ou aprés l'instant t.

Si la transition intervient aprés t alors le processus occupera l'état i durant l'intervalle de durée t et le gain s'établira en actualisant sur la période [0,t]. Le premier terme somme ces valeurs actualisées en les multipliant par leur probabilité d'occurrence pour tous les états successeurs possibles et pour tous les temps d'occupation supérieurs à t.

Si la transition est réalisée à un instant $\tau < t$, alors on établira un gain actualisé sur la période $[0,\tau]$. Le processus entrera dans l'état j alors qu'il reste encore $t-\tau$ instants par rapport à l'intervalle initial. L'espérance de la valeur actualisée des gains futurs au moment de la transition est alors : $v_j(t-\tau,\alpha)$. Mais cette valeur est observée au début de l'intervalle τ. Elle doit donc être multipliée par le facteur d'actualisation $e^{-\alpha\tau}$ pour tenir compte du délai additionnel τ. Le deuxième terme de l'équation somme ces valeurs multipliées par leur probabilité d'apparition pour toutes les valeurs de j, et les temps de transition $\tau < t$.

Nous nous intéresserons ici au comportement limite du processus. Donc à la valeur :

$$V(\alpha) = \lim_{t \to \infty} v(t,\alpha)$$

On peut démontrer (Howard 1971) que le vecteur $v(t,\alpha)$ approche un vecteur limite $v(\alpha)$ de valeur:

$$V(\alpha) = [\,I - C\,]^{-1}\, r(\alpha)$$

où I est la matrice identité et C la matrice produit telle que $c_{ij} = p_{ij} h^e_{ij}$ quelques soient i et j, avec P matrice des probabilités de transition et H^e_{ij} matrice des transformées de Laplace des densités de probabilité h_{ij} .

Dans le cas particulier qui nous intéresse, on peut démontrer que $r_i(\alpha)$ élément du vecteur $r(\alpha)$ a la forme suivante :

$$r_i(\alpha) = \sum_{j=1}^{N} p_{ij} \int_0^{\infty} h_{ij}(\tau)\,[\,b_{ij}(t,\alpha)\,]\,d\tau$$

soit avec b_{ij} indépendant du temps :

$$b_{ij}(\tau,\alpha) = (\,b_{ij} / \alpha\,)[\,1 - e^{-\alpha\tau}\,] \quad \text{avec } \alpha > 0$$

La valeur de $r_i(\alpha)$ s'établit alors de la façon suivante :

$$r_i(\alpha) = \sum_{j=1}^{N} p_{ij} \int_0^{\infty} h_{ij}(\tau) \left[(b_{ij} / \alpha) (1-e^{-\alpha\tau}) \right] d\tau$$

soit encore

$$r_i(\alpha) = \sum_{j=1}^{N} p_{ij} (b_{ij} / \alpha) [1 - h_{ij}^{e}(\alpha)]$$

où $h_{ij}^{e}(\alpha)$ est la transformée de Laplace de $h_{ij}(\alpha)$.

L'élément $r_i(\alpha)$ s'interprète comme l'espérance de la valeur actualisée liée à la première transition hors de l'état i quelque soit le moment ou elle intervient.

MISE EN OEUVRE DANS L'EVALUATION DE STRATEGIES DE RESTAURATION DE L'ACIPENSER STURIO EN AQUITAINE

L'approche que nous proposons exige que l'on puisse se mettre d'accord sur une identification des strategies et des états pertinents pour l'espèce étudiée. En outre, il faut adopter un jeu de base de paramètres qui comporte les probabilités de transition, les temps d'occupation et les rendements associés.

L'identification des stratégies et des états possibles de la population

C'est là a l'évidence une étape cruciale de la modélisation. A titre provisoire, nous envisagerons trois stratégies :

- ne rien faire
- lancer un programme léger
- mettre en oeuvre un programme lourd de restauration.

Disons pour fixer les idées que la stratégie 2 correspondrait aux options initiales du plan esturgeon en Aquitaine et la stratégie 3 viserait à des actions confortant le milieu naturel avec un co t direct (passes à poisson parfaitement adaptées) et un co t d'opportunité (limitation sévère des rejets polluants) élevés.

Chacune de ces stratégies va être intégrée au modèle sous forme d'un état d'entrée du système.

Pour rester simple à ce niveau trés exploratoire de la recherche, nous considérerons cinq états possible pour la population d'esturgeon :

- stabilisation à un haut niveau de stock
- stabilisation à un niveau de stock moyen
- stabilisation à un niveau de stock faible
- état critique niveau de stock trés faible
- disparition pure et simple de l'espèce

Le premier état est à apprécier par rapport aux potentialités qu'a offert autrefois le milieu (Trouvery et al., 1984). En l'absence d'information sur les niveaux de stock, on pourrait se référer à des prises de l'ordre de 100 t/an chiffre inférieur à celui évoqué pour les années 1920 (120 t).

Le deuxième état correspondrait à la situation des années 1940 avec un potentiel de prise de l'ordre de 40 t/an.

Le troisième état pourrait s'identifier à la situation des années 1960 avec un potentiel de pêche d'environ 6 t/an.

Le quatrième état correspondrait à la situation actuelle. On enregistre encore une présence sporadique de l'espèce.

Le dernier état correspond à ce qui s'est passé dans la plupart des fleuves en Europe. Il est traité comme un état absorbant dans le processus markovien (probabilité de rester dans l'état =1).

L'identification des probabilités de transition

La première réaction ici est de considérer comme impossible l'exercice de fixation des probabilités de transition. Mais que signifie un tel constat ? Certainement pas qu'une approche non probabilisée est plus pertinente. Plus rassurante sans doute. Mais entre de simples valeurs dites "moyennes" sans que l'on sache trés bien à quoi cela se rapporte d'ailleurs, et l'examen de distributions de probabilité il nous semble qu'il n'y a pas à exclure l'explicitation de l'aléa (Barnthouse et al., 1984). En l'absence d'éléments objectifs tels que calculs de fréquence d'occupation des divers états, on peut s'appuyer sur des évaluations d'experts[2] ou encore sur des probabilités subjectives reflétant les convictions des décideurs. Dans ce dernier cas le processus d'explicitation des probabilités subjectives peut être considéré comme participant à la rationalité de la décision.

Nous avons retenu à titre trés provisoire la matrice de probabilité correspondant à la figure 1.

L'identification des bénéfices et des co ts

A chaque état i étant donné son successeur j, nous allons associer une marge nette (différence entre chiffre d'affaire et co ts) par période d'occupation de l'état. Cette marge nette sera constante par unité de temps. Bien entendu, selon l'état et la transition considérés, cette marge peut être positive ou négative.

La détermination de telles valeurs est, nous l'avons dit, soumise à de grandes incertitudes. On sait que les bénéfices devraient être évalués par référence à deux grandes catégories de valeur (Point, 1986) :

- des valeurs d'usage : valeurs commerciales et valeurs de loisir,
- des valeurs non liées à l'usage direct : valeur d'option, quasi valeur d'option, valeur d'existence, valeur de legs[3].

Ces concepts ne font pas l'objet d'un consensus total. En particulier, les deux derniers, évoqués par Krutilla (1967), malgré des tentatives de mesure empirique (Walsh et al., 1984), n'ont pas encore un statut scientifique trés établi.

Pour l'esturgeon on ne dispose que de trés peu d'informations sur sa valeur d'usage et bien entendu on ignore tout de sa valeur d'option, d'existence ou de legs.

Il n'entre pas dans l'objet de cette recherche de procéder à des estimations en la matière. Nous les considérerons ici comme ayant le statut de paramètres.

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[2] Les expériences étrangères ne sont bien s r pas transportables, mais peuvent se révéler d'enseignements et aider à cadrer des probabilités (Williot, 1984)

[3] - La valeur d'option serait le consentement à payer une sorte de prime d'assurance pour conserver la possibilité d'un usage futur de la ressource

- La quasi valeur d'option serait la disponibilité à payer pour retarder la perte irréversible de la ressource compte tenu du fait qu'on pourrait vouloir l'utiliser ultérieurement

- La valeur d'existence serait la disponibilité à payer pour la satisfaction de savoir que la ressource est préservée sans considération d'une utilisation éventuelle

- La valeur de legs recouvrirait le consentement à payer pour la satisfaction de transmettre la ressource aux générations futures

Le point d'ancrage du profil de référence à partir duquel nous allons travailler s'appuie sur une estimation des co ts annuels du plan esturgeon engagés pour le contrat de plan Etat - Région Aquitaine 1984-1988[4]. Cela conduit grossièrement à un co t annuel de l'ordre de 2.0 MF (en francs constants 1989) pour l'ensemble de la filière esturgeon. Nous allons considérer qu'au début du nouveau plan, le co t à engager pour la seule filière *Acipenser sturio* afin d'assurer une continuité de programme, est de l'ordre de 1,0 MF. Le co t de référence associé à la stratégie 2 sera donc : C2 = 1,0 MF.

Le co t de la stratégie 1 est considéré comme nul : C1 = 0. Le co t de la stratégie 3 est un multiple de C2. Dans le profil de base il est fixé arbitrairement à 5C2.

Les bénéfices seront également envisagés comme des multiples de ce co t annuel.

Bénéfice net état E4 : B4 = 0.25 C2
Bénéfice net état E5 : B5 = 1.25 C2
Bénéfice net état E6 : B6 = 2.50 C2

Compte tenu d'une valeur moyenne de 0,68 MF/t départ pêcheur[5] et des hypothèses de prise posées précédemment, on obtient des bénéfices nets commerciaux de l'ordre de 3,7% du CA pour B6, 4,6% pour B5, 6,1% pour B4.

Les bénéfices et co ts des états E7 et E8 sont fixés provisoirement de la façon suivante :

Bénéfice net état E7 = 0
Co t net état E8 = -B6= -2.50 C2

La perte irréversible est réputée égale au gain maximum que l'on aurait pu obtenir : celui délivré par l'état E6.

Nous retiendrons donc comme jeu de base les valeurs suivantes :
C1=0, C2=1.0, C3=5.0, B4=.25, B5=1.25, B6=2.5, B7=0, C8=2.5

L'identification des durées d'occupation des états selon les transitions

Nous admettrons ici que les densités de probabilité d'occupation d'état h_{ij}, sont continues et correspondent à des exponentielles[6]. Le temps d'occupation τ_{ij} est donc décrit par la distribution :

$$h_{ij}(\tau) = \lambda e^{-\lambda\tau} \text{ avec } \tau \geq 0$$

Les éléments de la matrice des transformées de Laplace sont alors :

$$h_{ij}^{s} = \lambda/(s + \lambda)$$

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[4] Nous parlons d'estimation car, pour ce programme diversifié, à c té des crédits de l'Etat et de la Région, il convient de faire figurer une part d'autofinancement des organismes participant à la recherche ce qui pose bien s r quelques problèmes d'imputation (Bories com. pers.)

[5] Pour 100t de prises (50t de femelles, 50t de m les) on obtiendrait 55t de chair et 5t de caviar aux prix respectifs de 60 F/kg et 650 F/kg

[6] Nous aurions pu également nous situer dans un modèle à temps discret et utiliser par exemple des distributions géométriques

On peut démontrer facilement que lorsque λ=1, l'algorithme dégénère en un algorithme plus simple tel que :

$$V = [I-(1/(1+s))P]^{-1}R$$

où R est le vecteur des co ts et bénéfices par état

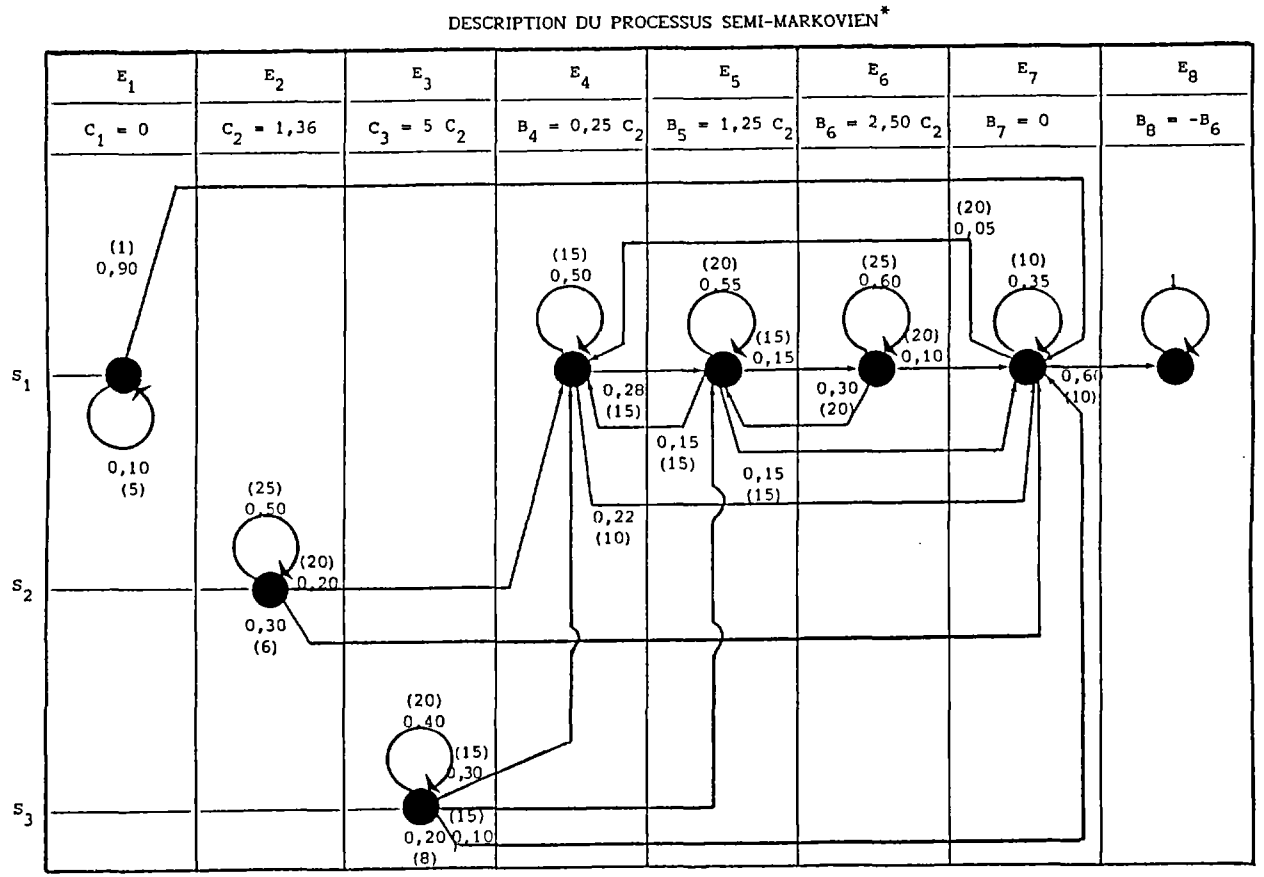


Figure 1- Description du processus semi-markovien. Les données entre parenthèses sont les durées moyennes d'occupation des états pour chaque probabilité de transition.

Nous avons retenu comme jeu de départ la matrice dont les valeurs lambda correspondent à la figure 1, s étant ici le taux d'actualisation. On observera que les durées moyennes associées aux probabilités de transition (entre parenthèses sur la figure 1) sont comprises entre 5 et 25 ans.

Les orientations de base du modèle

L'ensemble des hypothèses et paramètres sont rassemblés dans la figure 1.

Pour résumer la logique retenue à travers la fixation des probabilités, et les temps d'occupation, on dira que s'agissant des états de la population biologique, plus le stock stabilisé est important, plus la probabilité de rester dans l'état est élevée et plus la durée d'occupation est marquée. Plus le stock est faible plus on a des chances de transiter rapidement vers l'état absorbant. On tient compte d'une asymétrie fondamentale entre les durées de restauration de l'espèce et la vitesse de régression de la même espèce. Le risque est partout présent, à des niveaux variables. La politique la plus active en matière de restauration n'exclut pas complètement la possibilité d'une extinction de l'espèce locale. En observant les stratégies et les probabilités retenues, on constatera qu'elles tentent de refléter un plus ou moins grand potentiel de réussite.

Sauf précision contraire le modèle fonctionne avec un taux réel d'actualisation fixé à 5%.

LE FONCTIONNEMENT DU MODELE

Les résultats bruts du modèle avec le paramétrage retenu

Le modèle vise à comparer trois stratégies. On calcule l'espérance de gain actualisé associée à l'entrée dans chacun des trois états E1, E2, E3 représentant approximativement la situation actuelle combinée à trois stratégies.

Si l'on reprend le modèle avec les valeurs de paramètres telles qu'elles sont rappelées sur le schéma n 1, les résultats bruts sont les suivants :

Stratégie S1 (ne rien faire) = -24.26 M F

Stratégie S2 (agir modérément) = -19.03 M F

Stratégie S3 (agir plus intensivement) = -56.64 M F

La stratégie qui minimise la perte est clairement la stratégie intermédiaire S2.

Nous allons expliquer dans le point suivant pourquoi aucune de ces stratégies ne dégage une valeur actualisée positive en indiquant le r le joué par le co t d'opportunité d'une disparition de l'espèce, et par le taux d'actualisation.

L'influence du co t d'opportunité de disparation de l'espèce

La possibilité de disparition est traitée comme un état absorbant d'une chaîne de Markov. Dans l'analyse du fonctionnement asymptotique du modèle à laquelle nous procédons, ceci implique la nécessaire réalisation de cet état. Une telle propriété ne paraît pas inacceptable dans un modèle de trés long terme. Elle reflète une réalité indiscutable : celle d'un processus naturel de disparition d'espèces. Si cette disparition est appréciée négativement, ce co t pèsera significativement sur l'analyse, mais de façon différenciée selon les stratégies. Le tableau n 1 indique l'espérance actualisée du gain associé à chaque stratégie pour trois niveaux de co t d'opportunité.

Tableau 1- Espérance actualisée du gain associé aux trois stratégies et selon trois niveaux de côut d'opportunité

	$C_8 = 0*C_2$	$C_8 = 1*C_2$	$C_8 = -B_6 = 2,5*C_2$
S1	00.24	-09.56	-24.26
S2	-10.41	-13.86	-19.03
S3	-50.28	-52.82	-56.63

L'écart relatif le plus marqué est imputable à la stratégie d'inaction S1 (de l'ordre du facteur 100 pour les valeurs extrêmes), le plus faible concerne la stratégie S3 (facteur multiplicatif 1,12 pour les valeurs extrêmes).

L'introduction d'une valeur positive pour le co t d'opportunité de disparition de l'espèce amène dans la plupart des configurations à des stratégies dégageant une espérance de valeur actualisée négative. Ceci veut dire en clair que la politique de restauration d'une espèce est avant tout une politique de minimisation de perte.

L'influence de la valeur de cadrage C2

Le caractère trés général du modèle implique que les résultats doivent être interprétés en termes de tendance et non comme des prévisions financières. Nous nous intéressons à la confrontation de stratégies d'action et cherchons à détecter celle qui peut dominer, pour des ensembles de paramètres jugés pertinents par les spécialistes. La stratégie dominante sera celle qui dégage une différence de gain positive par rapport aux autres.

La valeur de C2 joue un r le de cadrage du modèle. Or, cette valeur retenue pour C2 à la fin du plan 1984-88 pourrait par exemple se révéler sous estimée compte tenu des autres paramètres, notamment des probabilités de transition du modèle. Jusqu'où peut-on aller sans qu'il y ait d'inversion de stratégie ? La figure n 2 représente une simulation de l'espérance de gain relatif de S2 pour des niveaux du co t annuel C2 variant entre -50% et +50%. On constate qu'il faut atteindre un co t annuel de 1,45 MF (donc 45% d'augmentation) pour que la stratégie d'inaction l'emporte.

L'influence du taux d'actualisation

On sait que le taux d'actualisation adopté par un pays peut s'interpréter comme un taux de substitution entre présent et futur. Une société parfaitement altruiste et qui accorderait autant de poids au bien être des générations futures qu'au sien propre utiliserait à la limite un taux nul. Cela ne correspond pas aux réalités actuelles et il est d'usage d'accorder un poids plus faible aux bénéfices à percevoir dans le futur, mais aussi aux co ts à acquitter à une date éloignée.

Il faut s'attendre à ce qu'un tel taux joue un r le important dans les résultats produits par le modèle. Nous avons procédé à une simulation numérique décrivant l'évolution des gains relatifs de S2 par rapport à S1 et S3 lorsque ce taux varie entre 0,5% et 26%. Le graphique n 3 qui synthétise ces résultats montre que jusqu'à 8,5% S2 domine S1. Au delà par contre, la stratégie d'inaction se révèle meilleure. Dans tous les cas, S2 domine S3.

QUELQUES ELEMENTS DE REFLEXION SUR UNE STRATEGIE DE RECHERCHE POUR LA RESTAURATION D'ESPECES SAUVAGES A EXPLOITATION COMMERCIALE

Action sur la durée des cycles ou sur les probabilités de réussite

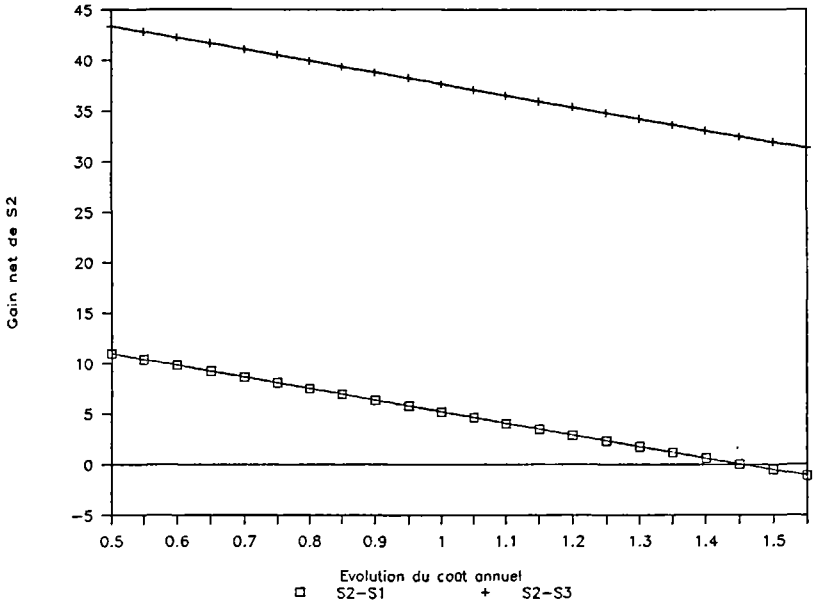


Figure 2- Espérance du gain relatif de S2 en fonction de l'évolution du côut de cette stratégie.

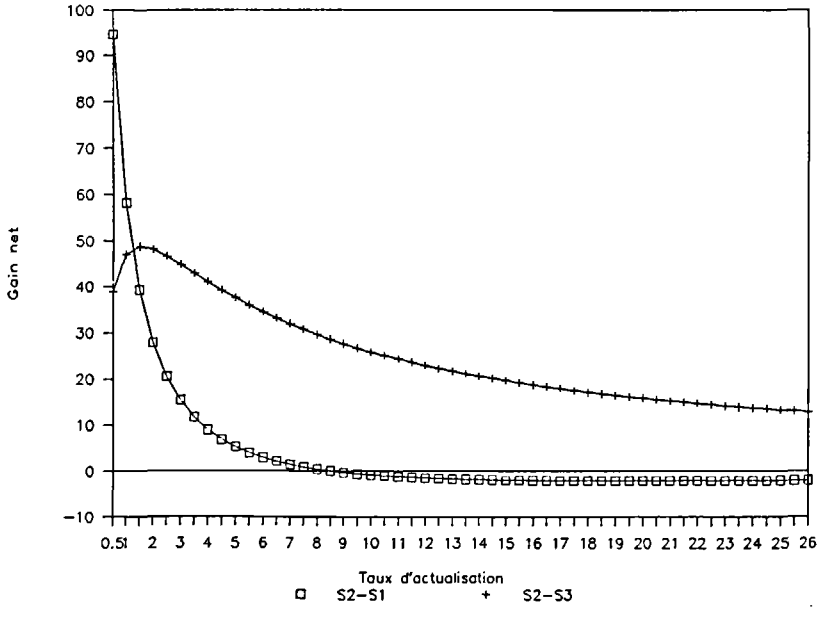


Figure 3 - Espérance du gain relatif de S2 en fonction du taux d'actualisation

Raccourcir les cycles ou améliorer les probabilités de réussite ? Certes, les deux possibilités ne s'excluent pas mutuellement, mais sur quoi doit on faire porter l'effort? Le taux de substitution entre réduction d'une année de cycle et amélioration de la probabilité de réussite n'est pas élevé. Il semble que dans une perspective de long terme ce soit la réduction de l'aléa, plus que celle des cycles qui améliore le plus les politiques.

. Effet d'une réduction de la durée associée à la transition de E2 vers lui même.

Le modèle initial considère que si par le jeu des probabilités on transite de E2 vers lui même, on reste bloqué 25 ans dans cette situation. Quelle amélioration des gains nets de la stratégie S2 peut on attendre si l'on parvient à réduire ce délai toutes choses restant égales par ailleurs ? La figure n 4 montre l'évolution des gains nets de S2 par rapport à S1 selon la réduction du temps d'occupation de l'état. L'effet est visible, il est non linéaire. En moyenne annuelle le gain net est de 0,0137 MF.

. Effet d'une réduction de la probabilité de transition de E2 vers lui même.

L'évaluation est ici plus délicate puisque, lorsque l'on réduit une probabilité de transition, il y a nécessairement compensation par amélioration des transitions vers un ou plusieurs autres états. Nous avons choisi ici de répartir la variation au profit du passage vers E4. Donc réduire p_{22} , c'est augmenter p_{24} et donc rendre le système plus performant. Si nous avions décidé de répartir équitablement la variation vers les deux transitions E4 et E7 on observerait une dégradation de la stratégie S2 car le passage vers E7 est trés pénalisant. Il paraît plus réaliste ici d'admettre que si l'on réduit p_{22} c'est en améliorant p_{24}. La figure n 5 retrace l'évolution de l'espérance de gain relatif de S2 par rapport à S1 lorsque p_{22} se réduit. Là encore la non linéarité est évidente. En moyenne une variation de .01 de probabilité entraîne un gain relatif de 0,403 MF.

Compte tenu de certaines réalités biologiques, il est sans doute plus facile d'agir sur la probabilité de réussite que sur la durée des cycles. On montre ici que l'effet économique en est beaucoup plus marqué.

Les co ts et les bénéfices d'une stratégie de recherche

. Effet d'une augmentation des bénéfices nets

L'augmentation homothétique (et dans une fourchette raisonnable) des bénéfices de chaque état n'introduit pas de modification dans l'ordre des stratégies. On a une relation linéaire croissante entre évolution des bénéfices et gain net de la stratégie dominante pour S2 contre S1 et décroissante pour S2 contre S3. Plus les bénéfices sont élevés, plus la rentabilité de S3 s'améliore (voir figure n 6). L'augmentation du rendement net associé à un des trois états bénéficiaires influe de la même façon, mais avec moins d'ampleur, sur le gain relatif des stratégies.

. Effet d'une modification du co t associé à une stratégie

Nous avons pu observer que la stratégie S3 lorsque les co ts associés sont 5 fois supérieurs à ceux de S2 ne peut être dominante pour des valeurs vraisemblables des autres paramètres.

A quel niveau de co t la stratégie S3 devient elle supérieure à la stratégie S2 ? Un calcul numérique illustré par la figure n 7 montre que par rapport à S1 la dominance est obtenue pour un co t C3=2*C2 et par rapport à S2 pour un co t C3=1,5*C2.

En clair si un programme de recherche représentant un co t supérieur de 50% au programme du plan esturgeon (stratégie S2) pouvait amener à une situation proche de celle de S3, alors il serait souhaitable de l'adopter.

. Effet d'une modification des probabilités de transition sur la hiérarchie des stratégies

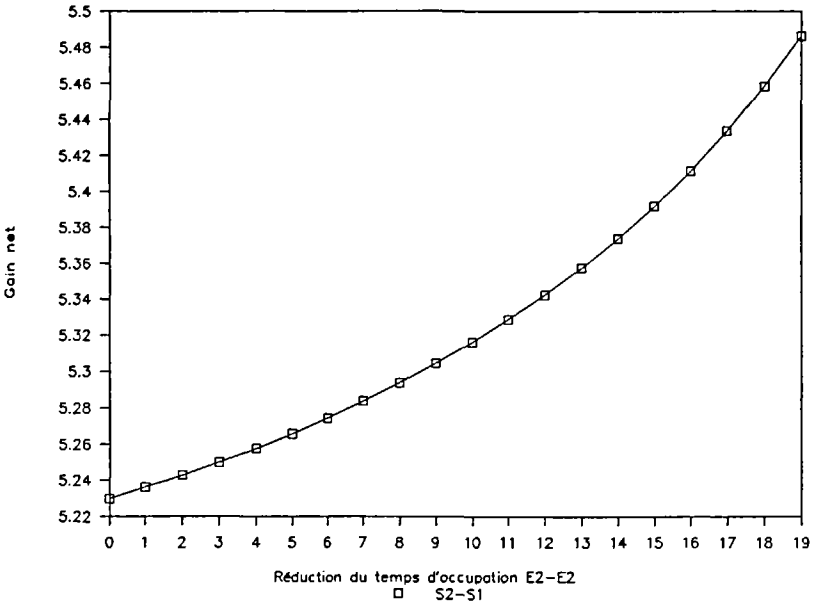


Figure 4- Espérance du gain relatif de S2 en fonction de la durée d'occupation pour E2-E2.

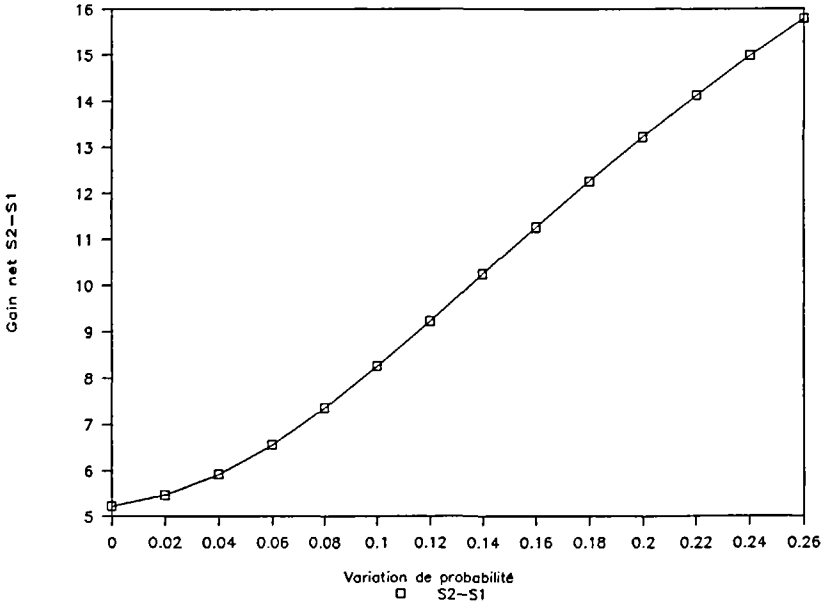


Figure 5- Espérance du gain relatif de S2 en fonction de la réduction de probabilité de passage E2-E2 et d'une compensation vers E2-E4.

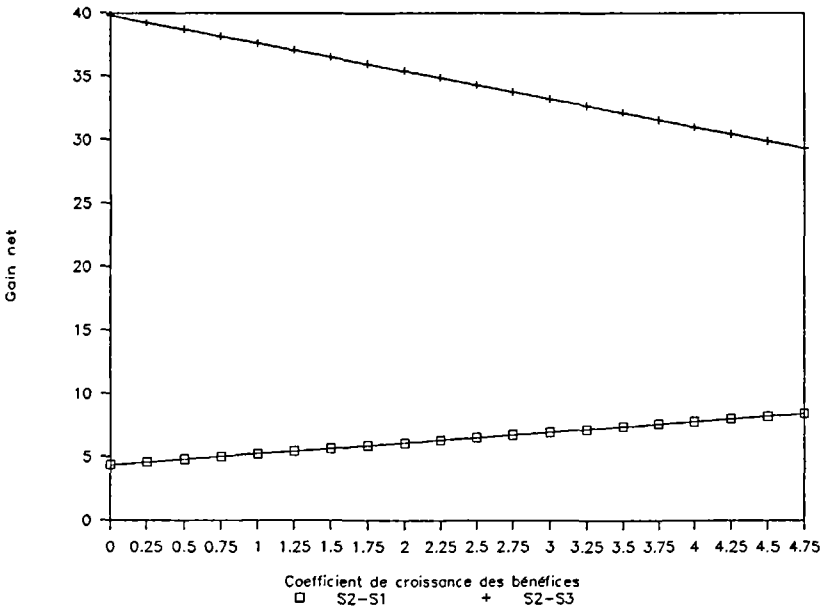


Figure 6- Espérance du gain relatif de S2 pour une évolution homothétique des bénéfices.

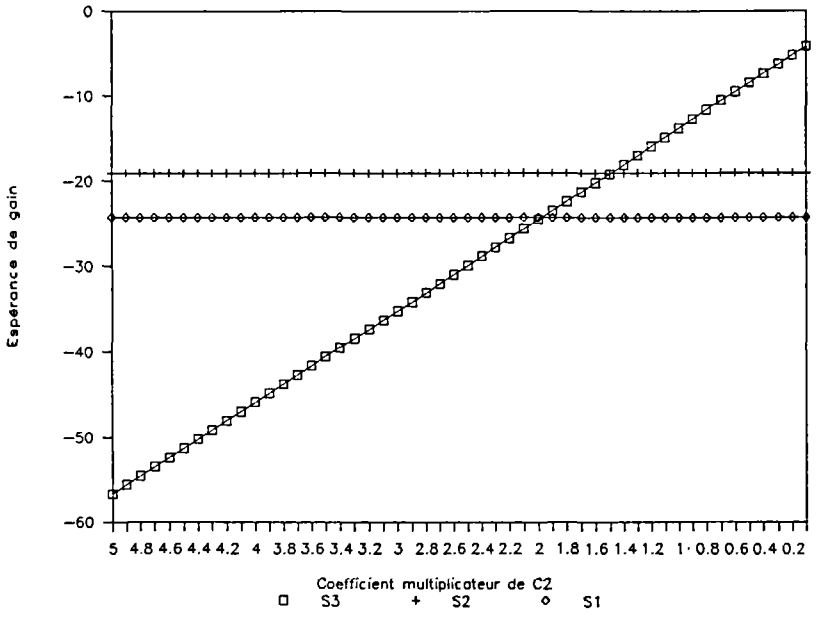


Figure 7- Espérance du gain de S3 en fonction de l'évolution de son côut.

En renversant le problème précédant, on peut se demander quelle structure de probabilité associée à la stratégie S3 pourrait, compte tenu de son co t assurer une dominance de S3. Nous avons programmé un calcul numérique qui s'appuie sur les hypothèses suivantes :

- réduction progressive et constante de p_{33} ainsi que de p_{37},
- réaffectation équivalente sur p_{34} et p_{35}.

La figure n 8 retrace l'évolution du gain relatif de S3 par rapport à S2 pour des variations au pas de 0,02. On constate que S3 devient dominante pour la structure de probabilité suivante :

p_{33} =0,235
p_{34} =0,465
p_{35} =0,265
p_{37} =0,035

Il est donc clair qu'un programme mettant en jeu des co ts 5 fois supérieurs à ceux de S2 ne sera efficace que s'il réduit considérablement la probabilité p_{37} de passage à l'état critique.

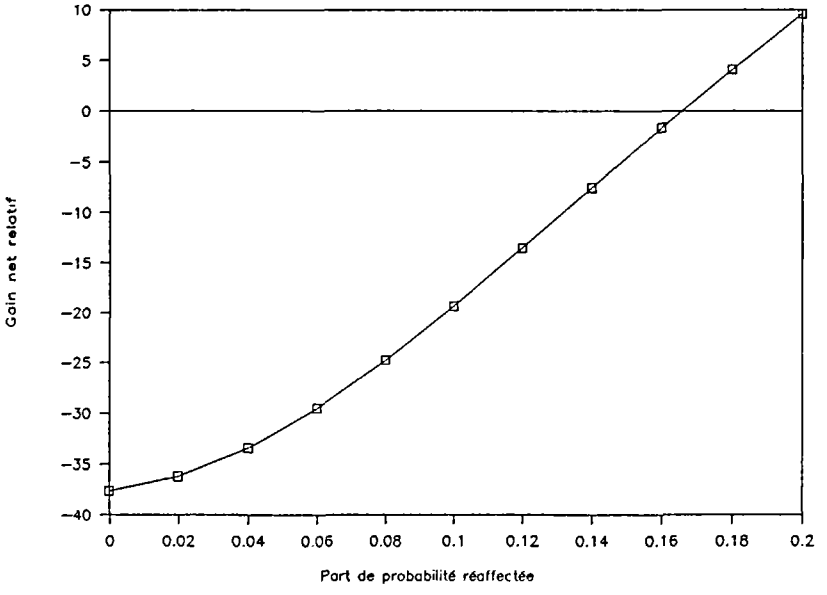


Figure 8- Espérance du gain relatif de S3/S2 en fonction d'une baisse identique de la probabilité de passage E3-E3 et E3-E7 avec un transfert vers E3-E4 et - E3-E5.

CONCLUSION

L'exercice auquel nous venons de nous livrer doit être apprécié avec toute la prudence qu'imposent les hypothèses inhérentes à l'approche markovienne et à la structure même du modèle retenu. Le modèle est intrinsèquement un modèle de long terme destiné au choix de stratégies fondamentales et non à des calculs financiers précis. Sous ces réserves, par sa souplesse, son caractère évolutif il peut aider les parties prenantes à la réhabilitation et la gestion de l'*Acipenser sturio*, à b tir un plan susceptible d'intégrer l'aléa qui se révèle être la composante dominante du problème. Les différents scénarios esquissés montrent un fonctionnement raisonnable (non contre-

intuitif) du modèle et une grande stabilité des solutions. On constate en particulier pour un domaine de variation assez large des paramètres une domination marquée de la stratégie S2.

La stratégie d'inaction S1 qui consiste à laisser les choses en l'état n'émerge que pour des valeurs élevées du taux d'actualisation ou pour des bénéfices attendus trés faibles peu réalistes.

Bien entendu, il faut y insister, il s'agit là de tendances qui doivent être confortées par des recherches complémentaires. Celles-ci, c'est tout à fait dans l'esprit du modèle, doivent contribuer à réduire l'incertitude, à limiter les scénarios possibles.

Ajoutons enfin qu'à coté des co ts et bénéfices commerciaux il convient de tenir compte des valeurs de loisir (pêche amateur), et des valeurs d'option de legs et d'existence auxquelles les instances régionales paraissent fort heureusement sensibilisées. L'accroissement des bénéfices qui en résulte, s'il est difficile à évaluer n'en conforte pas moins les stratégies d'action contre la simple défense passive.

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ELEMENTS DE SYSTEMATIQUE ET DE BIOLOGIE DES POPULATIONS SAUVAGES D'ESTURGEONS

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RESUME

Afin de permettre une vision globale de la famille des *Acipenseridae*, les auteurs rappellent certaines données systématiques, biologiques et écologiques de base. 24 espèces reconnues d'esturgeons constituent cette famille, dont 17 sont du genre *Acipenser*, 2 du genre *Huso*, 3 du genre *Pseudoscaphirhynchus* et 2 du genre *Scaphirhynchus*. Elles sont présentées successivement, avec leur principales caractéristiques systématiques, écologiques et biologiques. On constate une grande disparité dans la connaissance que nous avons des espèces, certaines ont été relativement bien étudiées, d'autres le sont encore très peu. Pour presque toutes il subsiste de grosses lacunes dans les connaissances de base. Au cours d'une brève synthèse, quelques caractères généraux de la famille sont mis en évidence. Les auteurs soulignent la nécessité de travaux systématiques sur cette famille et évoquent notamment le statut de certains taxons. On mentionne aussi l'importance de se doter de nouveaux outils plus performants reposant sur des données précises et récentes pour actualiser les critères de détermination des espèces.

Mots clés : *Acipenseridae*, Populations naturelles, Systématique, Ecologie.

ABSTRACT :

Some of the basic data about the systematic, the biology and the ecology of the *Acipenseridae* family are first given. From 24 recognized sturgeon species constituing this family, 17 are of the genus *Acipenser*, 2 of the genus *Huso*, 3 of the genus *Pseudoscaphirhynchus* and 2 of the genus *Scaphirhynchus*. All the species are successively presented with their main systematical, biological and ecological characteristics. There is still a great imbalance of knowledge about the different species, some have been relatively well studied but others are still not well known. For almost all the species, there is a lack of knowledge of their basic nature. In a brief synthesis, some general characteristics of this family are presented. The authors emphasize the need to work on the systematic of this family and mention in particular the case of certain taxa. The necessity to work on more efficient and accurate tools to update the determination criterion for the species is also discussed.

Key-words : *Acipenseridae*, Wild populations, Systematic, Ecology.

INTRODUCTION

Il y a actuellement pour l'ensemble des espèces d'esturgeons, un réel besoin de clarification des critères de détermination.

En milieu naturel, certaines espèces sont encore mal connues et difficiles à identifier à l'intérieur même de leur aire de répartition. D'autres s'hybrident assez fréquemment : *Scaphirhynchus albus* x *S. platorhynchus* (Carlson et al., 1985), *Acipenser ruthenus* x *A. gueldenstaedti* (Burtzev dans Charlon et Williot, 1978) et ceci complique encore les identifications. Quant aux animaux capturés en limite, voire en dehors de leur aire de répartition habituelle, il est très problématique de les identifier de façon sûre avec les clés de détermination actuelles.

En élevage, ce besoin va probablement s'accentuer avec le développement actuel de l'acipensériculture. Le polymorphisme intraspécifique et les conditions d'élevage (alimention, qualité de l'eau) rendent parfois difficile l'identification des animaux.

De plus qu'ils peuvent être originaires d'une région très éloignée de la pisciculture, c'est le cas par exemple des *A. transmontanus* originaires de la côte ouest

des Etats-unis et élevés en Europe. Les hybrides artificiels d'esturgeons sont parfois fertiles (*Huso huso* x *Acipenser ruthenus*) et pour la plupart extrêmement difficiles à identifier (*A. ruthenus* x *A. gueldenstaedti*), ceci complique encore les identifications.

Les appellations commerciales de la chair fraiche, fumée ou en conserve et des "caviars" varient d'un pays à l'autre et parfois même au sein du même pays sans qu'il soit possible d'établir des correspondances entre l'appellation et l'espèce d'origine. Ceci permet d'introduire sur le marché sous un nom flatteur la chair de poissons appartenant en fait à une espèce différente. Cette pratique est particulièrement aisée lorsque le produit est sous forme de conserve ou de semi-conserve car on ne peut plus identifier l'espèce (Morel, 1974).

METHODOLOGIE

Face à cette situation, indépendamment des résultats présentés au colloque Acipenser et publiés ici, il nous a paru nécessaire de dresser une liste alphabétique des espèces citées dans la littérature en donnant si possible les appellations synonymiques, les noms vernaculaires et les principales caractéristiques écologiques. Nous signalons également les divergences existant parfois entre les auteurs et les problèmes d'identification qu'elles peuvent entrainer. Certaines de ces données sont ensuite synthétisées sous forme de figures ou de tableaux.

Les noms vernaculaires des espèces sont issus en grande partie des ouvrages de : Banarescu et al. (1971); Bergeron et Brousseau (1983); Leim et Scott (1972); Lindberg et al. (1980); Quéro (1984); Scott et Crossman (1974). Ils sont donnés dans l'ordre alphabétique des pays ou des langues; les abréviations utilisées sont les suivantes : D, langue allemande; E, langue anglaise; F, langue française; R, langue russe. La richesse en noms vernaculaires de différentes langues, permet de situer et d'apprécier indirectement l'aire de répartition des espèces.

Les comptages d'écussons sont présentés sous la forme min-maxD (nombre d'écussons dorsaux minimum et maximum), min-maxL (nombre d'écussons latéraux minimum et maximum), min-maxV (nombre d'écussons ventraux à l'avant des nageoires pelviennes, minimum et maximum). La méthode de comptage des écussons qui est la plus utilisée est celle de Lebedev (1969), mais la façon d'effectuer ce dénombrement peut varier d'un auteur à l'autre. Les mesures maximales (exprimées dans les unités du système international) sont designées par Pmax (poids maximum) et LTmax (longueur totale maximum).

Nous donnons ci dessous quelques précisions concernant certains termes utilisés.

Caviar : ce mot d'origine turque (Gödecken, 1986) désigne historiquement et gastronomiquement les préparations d'ovocytes de *A. gueldenstaedti*, *A. stellatus* et *H. huso* de la mer Caspienne. Les spécialistes n'étant pas d'accord sur les espèces d'*Acipenseridae* pour lesquelles ce terme peut être utilisé, nous désignerons par caviar les préparations provenant des espèces précitées et par "caviar" (entre guillemets) les préparations d'ovocytes provenant de toutes autres espèces d'esturgeons.

Espèce : on appelle ainsi l'ensemble des organismes qui possèdent des génotypes identiques ou peu différents et qui se reproduisent exclusivement entre eux (Aron et Grassé, 1966). le terme espèce peut avoir actuellement deux significations: l'espèce typologique (basée sur "quelques individus de référence appelés types" et conservés dans des musées) et l'espèce biologique (basée sur "une population vivant dans un milieu déterminé"), la seconde tendant à supplanter la première (Daget, 1988). L'utilisation indifférente de l'un ou l'autre de ces deux concepts dans la systématique des *Acipenseridae* entraine des difficultés de classification.

Forme : du fait des incertitudes qui règnent pour certains taxons, ce terme pourra désigner la sous-espèce, la race où l'écotype.

Taxon : terme général pour désigner un groupe systématique : famille, genre etc. (Rickett, 1958 dans Lender et al., 1979)

PRESENTATION DES ESPECES

Les esturgeons appartiennent à l'embranchement des poissons, à la classe des ostéichthyens, au super ordre des Chondrostéens et à l'ordre des Acipensériformes. Cet ordre comprend deux familles : les *Acipenseridae*, au rostre conique, au corps revêtu de cinq rangées longitudinales d'écussons osseux et à la queue hétérocerque (esturgeons) et les *Polyodontidae* au rostre aplati en forme de spatule, au corps recouvert de très petites scutelles et à la queue homocerque (spatules) ou hétérocerque (*Psephurus*). Nous ne nous intéresserons ici qu'aux *Acipenseridae*.

La famille des *Acipenseridae* comprend deux sous-familles *les Acipenserinae* Bonaparte, 1845, avec les genres *Acipenser* Linné, 1758 et *Huso* Brandt, 1869 et les *Scaphyrhynchinae* Bonaparte, 1845 avec les genres, *Scaphirhynchus* Heckel, 1835 et *Pseudoscaphirhynchus* Bogdanov, 1874 (Rostami, 1961; Magnin, 1962). Les *Acipenserinae* généralement de grande taille possédent un spiracle, leur rostre est plus ou moins conique et ils n'ont pas de filament caudal (Magnin, 1962), par contre les *Scaphyrhynchinae* sont des animaux de petite taille, ne possèdent pas de spiracle, ont un rostre aplati en forme de pelle et un long filament caudal.

Nous allons présenter ci dessous la liste des 24 espèces qui nous semblent le plus unanimement reconnues comme telles par la plupart des auteurs. Les autres espèces citées dans la littérature seront considérées comme des formes et si possible rapportés aux espèces sélectionnées.
Pour la désignation des espèces, on a utilisé les règles principales du Code International de nomenclature zoologique, dont certaines sont rappelées dans Daget (1988).

Genre *Acipenser*.

Acipenser baeri Brandt, 1869.

Synonyme : *A. stenorhynchus* Nikolskij, 1939.
(E) siberian sturgeon, (F) esturgeon sibérien, (R) sibirskij osetr.

Le dos est de couleur gris clair à marron foncé et la coloration du ventre varie du blanc au jaune clair (Lebedev et al. 1969 dans Charlon et Williot, 1978). Ecussons : 10-19D 32-59L 7-16V (Lebedev et al., 1969 dans Charlon et Williot, 1978); 12-19D 37-56L 9-15V (Berg, 1962). Le nombre de branchiospines varie de 24 à 45, le plus souvent on en compte entre 30 et 37 (Berg, 1962). Le caryotype est de $2n = 248 \pm 5$ chromosomes (Vasil'ev et al., 1981; Burtzev et al., 1976). La forme du rostre varie selon les individus et les régions. Quelques petites plaques en forme d'étoile sont disséminées entre les rangées d'écussons (Berg, 1962). La lèvre inférieure est très distinctement fendue (Lebedev et al., 1969 dans Charlon et Williot, 1978). LTmax > 3 m, Pmax > 100 kg exceptionnellement 200 kg (Berg, 1962). La longévité maximale est d'au moins 60 ans (Kozhin, 1964). Il existe différentes formes dont une assez caractéristique des rivières Léna, Ienissei et Kolyma. Elle est considérée par certains (Menshikov, 1947 dans Berg, 1962; Semenkova, 1980) comme une sous-espèce *A. baeri stenorhynchus* Nikolskij, 1939; pour d'autres il s'agit d'un hybride de *A. baeri* et de *A. ruthenus*. Entre autres différences ces animaux ont un rostre généralement plus long que les *A. baeri* typiques. Il existe également une forme typique inféodée au lac Baïkal : *A. baeri baicalensis* (Egorov, 1948 dans Kozhin, 1964).

Bien que certaines formes fréquentent les estuaires, notamment celui du Khatanga (Berg, 1962), cette espèce est essentiellement dulçaquicole. On la trouve dans les fleuves sibériens, de l'Ob à la Kolyma ainsi que dans les lacs Baïkal et Zaïsan.

Elle semble également être acclimatée dans le nord de la Chine (Berg, 1962). La maturité sexuelle dans le milieu naturel est atteinte pour les mâles vers 11-13 ans (0,75-0,80 m) dans les rivières Ob, Irtysh et Ienissei (Berg, 1962), 15 ans (0,7 m) lieu non précisé (Kozhin, 1964), 9 ans (0,6 m et 0,73 kg) dans l'Aldan (Sokolov et al., 1986), 0,6 m et 1 kg dans la Léna (Sokolov et al., 1986) et pour les femelles vers 17-18 ans (0,85-0,90 m) dans les rivières Ob, Irtysh et Ienissei (Berg, 1962), 16-20 ans lieu non précisé (Kozhin, 1964), 11-12 ans (0,6 m et 1,2 kg) dans la Léna (Sokolov et Malyutin, 1977), 10 ans (0,6 m et 0,73 kg) dans l'Aldan (Sokolov et al., 1986). La reproduction a lieu pour les mâles tous les 1-2 ans (Kozhin, 1964), 2-4 ans (Sokolov, 1965) et pour les femelles 3-4 ans (Kozhin, 1964), 3-6 ans (Sokolov, 1965). Les géniteurs migrent au printemps vers l'amont des rivières pour frayer en juin-juillet dans des zones de rapides (1,4 ms^{-1}) relativement profondes (6-8 m) et à fond rocheux (Berg, 1962; Sokolov et Malyutin, 1977).

Les oeufs de 2,4 à 2,9 mm de diamètre adhèrent aux galets du fond. Après l'éclosion, les alevins se dirigent progressivement vers les zones de nourricerie situées à l'aval. Selon les zones, les *A. baeri* se nourrissent de larves d'insectes (*Chironomidae, Simulidae, Ephemeridae*) ou de crustacés, amphipodes et isopodes (Lebedev, 1961; Berg, 1962; Kozhin, 1964).

Acipenser brevirostrum Lesueur, 1818

Synonymes : *A. brevirostris* Richardson, 1836 (Jordan et al., 1930; Magnin, 1963; Vladykov-Greeley, 1963); *A. lesueurii* (Valenciennes-Duméril, 1870); *A. microrhynchus* (Dum., 1870); *A. dekayii* (Dum., 1870); *A. rostellum* (Dum., 1870); *A. sinus* (Valenciennes-Duméril, 1870).
(Canada) esturgeon à museau court, (E) : shortnose sturgeon, bottlenose sturgeon.

Le dos est noirâtre avec des reflets olive, les flancs sont gris avec des nuances rougeâtres, le ventre blanc et les viscères noires. Le museau est court et la bouche large. Ecussons : 8-11D 22-33L 6-9V (Leim et Scott, 1972), 7-13D 21-35L 6-11V (Dadswell et al., 1984). Le nombre de branchiospines sur le premier arc est en moyenne de 25,5 et le plus souvent entre 22 et 29 (Vladykov et Beaulieu, 1951). LTmax $\geq$ 0,9 m (Leim et Scott, 1972), 1,35 m (Gorham et Mc Allister, 1974 dans Smith et al., 1986), Pmax $\geq$ 45 kg. La longévité de cette espèce peut atteindre 67 ans (Dadswell et al., 1984).

Principalement dulçaquicole cette espèce effectue parfois des incursions estuariennes voire marines (Dadswell, 1979). Elle fréquente la côte est de l'Amérique du nord du fleuve St Laurent à la Caroline du sud et remonte les rivières Hudson, Savannah, St Jean et Connecticut (Vladykov et Greeley, 1963; Snyder, 1988). Il existe des populations bloquées en eau douce à l'amont de barrage (Buckley et Kynard, 1985). Les mâles atteignent la maturité sexuelle vers 5 ans (0,65 m) dans l'Hudson (Greeley, 1937 dans Magnin, 1962), 11 ans (0,45-0,50 m) dans la rivière St John (Dadswell, 1979) et les femelles vers 6 ans (0,65 m) dans l'Hudson (Greeley, 1937 dans Magnin, 1962) et 13 ans (0,45-0,50 m) dans la rivière St John (Dadswell, 1979. La reproduction ne surviendrait qu'une fois tous les 3-8 ans (Taubert, 1980; Smith et al., 1982). Au printemps les géniteurs se déplacent vers les zones de frayères situées à l'amont (Leim et Scott, 1972), elles se caractérisent par un courant rapide (0,4-0,6 ms^{-1}) et un fond de graviers (Taubert, 1980). La fraie a lieu de février à mai, (Dadswell et al., 1984), les ovules brun-sombres à noirs mesurent de 3,0 à 3,2 mm de diamètre (Dadswell 1979), les oeufs sont adhésifs et démersaux (Meehan, 1910 dans Dadswell et al., 1984). Durant l'été, les adultes redescendent les fleuves jusqu'à la limite de salinité mais les juvéniles de l'année restent en eau douce. La croissance est lente, 0,51-0,66 m à 5 ans et 0,63-0,75 m à 14 ans (Leim et Scott, 1972). L'alimentation se compose d'insectes, crustacés, mollusques et annélides. Les juvéniles d'un an et plus et les adultes passent l'hiver dans

des zones profondes des estuaires à des profondeurs de plus de 10 m. Au printemps ils retournent dans les fleuves pour se nourrir (Dadswell, 1976 dans John et al., 1978).

Cette espèce, actuellement sur la liste des espèces menacées, est protégée dans 13 états américains (Johnson, 1987).

Acipenser dabryanus **Duméril, 1868**

(E) Yangtze sturgeon, (F) petit esturgeon du Yangtze, (Japonais) chōzen chōzame.

Les plaques osseuses du crane sont très nettes et les écussons sont recouverts de denticules osseux. Ecussons : 10-12D 33-54L (Huang Hongjin et al., 1982).

Cette espèce exclusivement dulçaquicole fréquente les zones à fort courant du bassin amont du Yangtze (Province du Szechwan) et du Houang-ho.

Les animaux atteignent la maturité sexuelle à 5 kg pour les mâles et 10 kg pour les femelles (Huang Hongjin et al., 1982).

Acipenser fulvescens **Rafinesque, 1817**

Synonymes : *A. maculosus* Lesueur 1818; *A. rubicundus* Lesueur, 1818; *A. serotinus* Raf., 1820; *A. ohiensis* Raf., 1820; *A. macrostomus* Raf., 1820; *A. truncatus* Raf., 1820; *A. rupertianus* Richardson, 1836; *A. laevis* Agassiz, 1850; *A. carbonarius* Agassiz, 1850; *A. rhynchaeus* Agassiz, 1850; *A. liopeltis* Günther, 1870; de nombreux noms d'espèces ont été attribué par Duméril en 1870 selon les zones d'origine, on trouve ainsi : *A. anthracinus* (lac Erie); *A. megalaspis* (Lac Champlain); *A. lamarii* (lac Erie ?); *A. atelaspis* (Rivière Saskatchewan); *A. rosarium* (lac Erie ?); *A. kirtlandi* (Lac Erie); *A. buffalo* (Lac Erie); *A. anasimos* (Missouri); *A. paranasimos* (Huntsville, Alabama); *A. rafinesquii* (rivière Ohio); *A. rosarium*, (lac Erie); *A. copei* (partie amont du Missouri ?); *A. platyrhinus* (partie amont du Mississipi); *A. rauchi* (rivière Osage, Missouri); *A. nertinianus* (Michigan); *A. cincinnati* (rivière Ohio); *A. richardsoni* (partie amont du Missouri); *A. sturio* Eigemmann, 1895.
(Canada) esturgeon de lac, esturgeon jaune, camus, rock sturgeon (E) lake sturgeon, ripe sturgeon, smoothback (F) esturgeon lacustre, esturgeon jaune.

Couleur jaune pour les jeunes individus, rougeâtre ou verdâtre pour les plus vieux. Ecussons : 9-17D 29-42L 7-12V (Scott et Crossman, 1974), 12-15D (Vladykov et Beaulieu, 1946), 30-38L (Conte et al., 1988). Le premier arc branchial présente en moyenne 33 branchiospines et le plus souvent de 30 à 37 (Vladykov et Beaulieu, 1951). LTmax 2,41 m (Scott et Crossman, 1974), Pmax 140 kg (Madson, 1987).

Cette espèce, de comportement grégaire (Dumont et al., 1987) est strictement dulçaquicole; bien qu'on trouve parfois certains individus en eau saumâtre au Québec (Conte et al., 1988). Elle fréquente les lacs et les zones calmes des rivières de la partie orientale de l'Amérique du nord, notamment les lacs Champlain, Huron, Michigan, Nippissing, St Pierre, St Louis, St François, Supérieur, Winebago, Winnipeg, et les rivières Ottawa, Ohio, Moose, Richelieu, Tennessee, Fox et Wolf. Elle affectionne particulièrement les zones peu profondes (< 4,5 m) avec des fonds de vase ou de graviers. La maturité sexuelle est atteinte pour les mâles vers 18-22 ans (0,85-0,95 m et 3,9-5,2 kg) dans la rivière Nottaway (Magnin, 1966), 16 ans dans le Wisconsin (Priegel, 1973), 25 ans dans l'Ontario (Threader et Brousseau, 1986), 16 ans au Québec (Goyette et al., 1987) et pour les femelles vers 20-23 ans (0,9-1,0 m et 4,3-9,0 kg) dans la rivière Nottaway (Magnin ,1966), 24-26 ans dans le Wisconsin (Priegel, 1973), 20 ans dans l'Ontario (Threader et Brousseau, 1986), 27 ans (1,33 m) au Québec (Goyette et al., 1987).La plupart des mâles ne frayent que tous les 2-3 ans et les femelles tous les 4-6 ans (Magnin, 1966 dans Mongeau et al., 1982); Goyette et al. (1987) indiquent même un intervalle de 9-10 ans pour les femelles du lac St Louis. D'après Dumont et al. (1987)

certains individus, le plus souvent des mâles, peuvent même frayer deux années de suite.

Au printemps les géniteurs migrent vers les zones de frayères généralement situées à l'amont des rivières dans des zones peu profondes (< 3,5 m) sur des fonds de pierre ou de gravier au pied de rapides ou de chutes d'eau (Magnin, 1966). La période de fraie semble assez étalée, elle se situe selon les zones de début mai à la fin juin, les géniteurs ne semblent pas se nourrir pendant cette période (Magnin, 1962). Les alevins ont une croissance lente et les jeunes sont sédentaires. Les *A. fulvescens* se nourrissent d'invertébrés benthiques : larves d'insectes, mollusques, crustacés (surtout amphipodes), annélides, exceptionnellement de petits poissons; en proportion variable selon les zones (Mongeau, 1982). Durant la période hivernale qu'ils passent dans des zones profondes mais bien oxygénées leur activité est ralentie (Conte et al., 1988). La longévité de cette espèce est au moins de 55 ans pour les mâles et de 80 ans pour les femelles (Magnin, 1962; 1966). Cette espèce est actuellement protégée dans 12 états américains dont le Michigan (Johnson, 1987).

Acipenser gueldenstaedti **Brandt, 1833**

Synonymes: *A. güldenstädti* Brandt, 1833.
(Bulgare) essetra, (D) waxdick, russischer stör, (E) osetr, russian sturgeon, (Estonien) venetuur, (Hongrois) obicna jesetra, (Iranien) tass mahi, (Italien) storione danubiano, (Lithuanien) rusiskasis ersketas, (Roumain) nisetrul, nisetru, (R) ruskij osetr, osetr, (Slovaque) jeseter rusky', (Tchèque) jeseter rusky', (Turc) rusmersini, karaca, rusmersin baligi.
Certaines sous espèces sont parfois distinguées : *A. gueldenstaedti persicus* Borodin, 1897, cité par Kozhin (1964) et élevé par certains au rang d'espèce : *Acipenser persicus* cité par Artyukhin (1988). Celui-ci distingue également une sous espèce *Acipenser persicus colchicus* (Artyukhin et Zarkua, 1986 dans Artyukhin, 1988) appelée : (E) persian sturgeon, (R) kurinskij, lersidskij.
On trouve aussi les appellations : *A. gueldenstaedti colchicus* Marti, 1940 (mer noire) et *A. gueldenstaedti colchicus natio tanaicus* Marti, 1940.

Ecussons : 5-19D 24-50L 6-14V, ils sont pourvus de petits épaississement radiaux (Lebedev et al., 1969 dans Charlon et Williot, 1978). Le museau est court et massif, les barbillons sont non frangés et placés très en avant et la lèvre inférieure n'est pas fendue. Le caryotype est de 2n = 250 ± 8 chromosomes (Birstein et Vasil'ev, 1987). LTmax 2,15 m (Berg, 1962; Pavlov, 1964), 2,35 m (Tsepkin et Sokolov, 1979), Pmax $\geq$ 80-100 kg (Kostomarov, 1933; Lebedev et al., 1969 dans Charlon et Williot, 1978). Certains auteurs distinguent plusieurs formes à l'intérieur de cette espèce, alors que Artyukhin (1988) considère que les animaux qui frayent de juin à septembre notamment dans les rivières Rioni et Inguri ne sont pas des *A. gueldenstaedti* mais des *A. persicus*.

Cette espèce amphihaline fréquente les bassins des mers d'Azov, Noire et Caspienne. Elle remonte les fleuves Volga, Oural, Kura, Terek, Sulak (Caspienne), Danube, Dniepr, Bug, Dniestr, Rioni, Inguri (mer Noire), Don, Kuban (mer d'Azov). Dans les fleuves Oural et Volga on rencontre des formes d'eau douce (Lebedev et al., 1969 dans Charlon et Williot, 1978). La maturité sexuelle est atteinte pour les mâles vers 8-9 ans dans le Don (Kozhin, 1964), 8-10 ans (100 cm et 3 kg) dans la Volga (Veshev et Novikova, 1986 a), 13-14 ans (10-20 kg) dans la Kura (Kozhin, 1964, Lebedev et al., 1969 dans Charlon et Williot, 1978) et pour les femelles 10-14 ans dans le Don (Kozhin, 1964), 12-13 ans (120 cm et 9 kg) dans la Volga (Veshev et Novikova, 1986 a), 10-20 ans (18-30 kg) dans la Kura (Kozhin, 1964; Lebedev et al., 1969 dans Charlon et Williot, 1978). Comme pour d'autres espèces d'*Acipenseridae*, on distingue parmi les géniteurs des migrants de printemps (février à mai) et des migrants d'automne (août à octobre) (Manea, 1966). La ponte a lieu au printemps, l'année de la migration pour les migrants de printemps et l'année suivante pour les migrants d'automne. Les frayères sont dans des zones à fort courant, sur des fonds de 4 à 10 m et à une température de 8 à 15 °C

(Vlasenko et al., 1989). Il y a de 50 à 80 ovules au gramme (Kozhin 1964), les oeufs sont adhésifs (Detlaf et al., 1981).

Après la fraie les adultes repartent rapidement vers la mer et les jeunes restent en rivière un certain temps. L'alimentation des *A. gueldenstaedti* consiste en petits crustacés et annélides, les adultes consomment également des mollusques et des poissons du genre *Clupeonella* (Carausu, 1952 dans Manea 1966; Polyaninova, 1979). La longévité est de plus de 50 ans (Carausu, 1952 dans Manea 1966; Lebedev et al., 1969 dans Charlon et Williot, 1978), voire plus de 100 ans (Berg, 1962; Pavlov, 1964).

Acipenser kikuchii **Jordan et Snyder, 1901**

(E) japanese sturgeon, (Japonais) kikuchii chōzame.

On le trouve dans le sud du Japon (Rostami, 1961; Doroshov, 1985). Cette espèce est considérée comme rare, voire même très rare (Berg, 1904 dans Magnin, 1959 a).

Acipenser medirostris **Ayres, 1854.**

Synonyme : *A. acutirostris*
(E) green sturgeon, japanese sturgeon, Sakhalin sturgeon, (F) esturgeon vert, esturgeon de Sakhaline, esturgeon du pacifique, (Japonais) chōzame, (R) Sakalinskij osetr, Tikhookeasikij osetr.

De couleur vert noir à vert olive. Ecussons : 7-10D, 27-36L ((Lindberg et Legeza, 1965; Okada, 1960) dans Gritsenko et Kosmyunin, 1979), 20-30L (Conte et al., 1988). LTmax $\geq$ 2,3 m, Pmax $\geq$ 160 kg en Amérique (Clemens et Wilby, 1961 dans Conte et al., 1988), $\geq$ 60 kg (Asie). Certains auteurs font de la forme asiatique "mikadoi" une sous espèce *Acipenser medirostris mikadoi* Hilgendorf 1892, voire même une espèce *Acipenser micadoi* (ou *mikadoi*) Hilgendorf 1892 (Magnin, 1959; Gritsenko et Kosmyunin, 1979). La forme asiatique a plus de rayons aux nageoires anale et dorsale, plus d'écussons latéraux et moins d'écussons ventraux que la forme américaine (Rostami, 1961).

Ce migrateur amphihalin ne fréquente les eaux douces que durant très peu de temps, lors de ses migrations anadromes, il se cantonne généralement aux estuaires (Houston, 1987). On le trouve sur la côte pacifique de l'Amérique du nord, du golfe de l'Alaska au sud de la Californie (Conte et al., 1988) et sur les côtes asiatiques : Corée, Chine, Japon, URSS (Berg, 1962). Il fréquente les fleuves japonais Ishikari et Teshio (Okada, 1955) et parfois le bassin du fleuve Amour (Rostami, 1961). Cette espèce atteint la maturité sexuelle à une taille de 1 m et un poids de 8-10 kg. Les géniteurs remontent les rivières en automne et en hiver pour frayer au printemps (Conte et al., 1988) à une température allant de 9°C à 17 °C (Houston, 1987). Les frayères sont dans des zones de courants vifs sur des fonds rocheux, à l'aval de rapides ou de chutes d'eau (Houston, 1987). L'esturgeon vert se nourrit essentiellement d'invertébrés benthiques, les plus grands individus mangent parfois des poissons (Houston, 1987). Cette espèce ne semble faire l'objet d'aucune protection légale en Amérique du nord.

Acipenser multiscutatus **Tanaka, 1898**

(Japonais) sennin chōzame.

Cette espèce vit dans le sud du japon (Rostami, 1961).

Acipenser naccarii **Bonaparte, 1836**

Synonymes : *A. nasus* Heckel, 1851; *A. nardoi* Heckel, 1851.
(F) esturgeon de l'Adriatique, esturgeon italien, (E) adriatic sturgeon, (Croate) jadranska jesetra, (Hongrois) jesetra tuponoska, (Italien) storione cobice, storione italiano, (Slovaque) jadranski jeseter.

Son museau est court et arrondi, la lèvre inférieure est fendue, les barbillons ne sont pas frangés et les écussons sont plus de deux fois plus haut que large (Muus et Dahlström, 1981). Son caryotype est de 2n = 239 ± 7 chromosomes (Fontana et Colombo, 1974).

Probablement surtout dulçaquicole il fréquente le Pô mais on le trouve parfois dans l'Adriatique. La reproduction aurait lieu en avril-mai dans le cours moyen des fleuves (Maitland, 1987). Les jeunes se nourrissent d'invertébrés benthiques, les adultes consomment également des petits poissons (Maitland, 1987).

Acipenser nudiventris **Lovetzky, 1828**

Synonymes : *A. shypa* Eichwald, 1831; *A. glaber* Fitzinger et Heckel, 1836.
(Bulgare) ship, (Croate) sim, (D) glattdick, glatte stör, schip, (E) spiny sturgeon, (F) esturgeon nu, ship, (Hongrois) szintok, (Italien) storione nudo, (Lithuanien) pleistasnùkis ersketas, (Polonais) jesiotr, kustén, kostera, szyp, szypa, (R) ship, bastard, osetr, osetr ship, (Roumain) viza, bogzar, (Slovaque) jeseter hladky', (Tchèque) hladky'jeseter, (Turc) sip , sip baligi, (Ukrainien) ship.

Ecussons : 11-26D 52-74L 11-17V, 11-17D 55-66L 12-16V (Elanidze et al., 1970). LTmax ≥ 2 m, Pmax 65-80 kg (Kostomarov, 1933). La lèvre inférieure est non fendue et les barbillons sont frangés (Lebedev et al., 1969 dans Charlon et Williot, 1978). Son caryotype est de 2n = 118 ± 3 chromosomes (Aref'ev, 1983) ou 118 ± 2 (Sokolov et Vasil'ev, 1989).

Cette espèce dont la plupart des populations sont amphihalines se trouve dans les bassins des mers d'Aral et Caspienne, plus rarement dans les mers Noire (elle remonte le Danube jusqu'à Budapest) et d'Azov. Il existerait des populations dulçaquicoles dans le Danube (Manea, 1966) et acclimatées dans le lac Balkhach (Lebedev et al., 1969 dans Charlon et Williot, 1978). Elle affectionne les zones de fonds durs et de courant rapide et elle hiberne dans des fosses profondes (Manea, 1966). Les mâles atteignent la maturité sexuelle vers 6-9 ans et les femelles vers 12-14 ans. La reproduction a lieu sur des fonds de galets dans le cours moyen des fleuves, en avril dans le Syr-Daria et l'Amou-Daria et en mai-juin dans la Kura. Les ovules mesurent environ 3mm de diamètre et les oeufs sont adhésifs. Les individus venant de frayer et les jeunes de l'année repartent en mer rapidement (Lebedev et al., 1969 dans Charlon et Williot, 1978), ils se nourrissent d'invertébrés benthiques (larves d'insectes, mollusques, crustacés), tandis que les adultes consomment également des poissons (Maitland, 1987). La croissance de cette espèce est très variable selon les zones géographiques (Elanidze et al., 1970).

Acipenser oxyrhynchus **(Mitchill, 1814)**

Synonymes : *A. sturio* L., 1758; Günther, 1870; Ryder, 1890; Dean 1894; Cox, 1896; Monpetit, 1897; Halkett, 1913; Smith et Bean, 1899; Kendall, 1914; Borodin, 1925; Bigelow et Schroeder, 1936; La Gorce, 1939; Bridge et Boulenger, 1958; Rostami, 1961. *A. Cayennensis* Dum., 1867; *Antaceus hallowellii* Dum.; *Antaceus lecontei* (New york)

Dum., 1851; *Huso milberti* (New york) Dum., 1822; *Huso macrorhinus* (New york) Dum., 1820;
Huso mitchilli (New york) Dum.; *Huso kennicotti* (Rivière James) Dum.; *Huso. girardi* (Maryland) Dum.; *Huso bairdi* (Maryland) Dum.; *Huso storeri* (Boston) Dum.; *Huso holbrooki* (Charleston) Dum..
(Canada) esturgeon noir, esturgeon de mer (D) atlantischer stör, (E) atlantic sturgeon, sea sturgeon, common sturgeon, american atlantic sturgeon, (F) esturgeon américain, (R) Ostrorinij osetr.
On distingue généralement une forme *Acipenser oxyrhynchus desotoi* Vladykov, 1955, sur les côtes de Floride et dans le golfe du Mexique, (E) gulf of Mexico sturgeon, gulf sturgeon, common sturgeon.

De couleur brun-olive avec des taches rouges et blanches sur le ventre, ses viscères sont très pâles (Leim et Scott, 1972). Ecussons : 10D 29L 10V (Magnin 1962), 10-16D 26-34L 9-14V (Leim et Scott, 1972), 7-16D 24-35L 8-12V (Scott et Crossman, 1974). LTmax $\geq$ 4,25 m (Leim et Scott, 1972), Pmax $\geq$ 367 kg (Leim et Scott, 1972). Le nombre de branchiospines sur le premier arc branchial varie de 15 à 27 (John et al., 1978). La forme *Acipenser oxyrhynchus desotoi* à une tête plus large et plus longue, ses nageoires sont également plus longues et les écussons ont une teinte très claire.

Cet amphihalin fréquente les côtes atlantiques de l'Amérique du nord, de la baie d'Hudson (Prince, 1898) à la Floride (Jordan et Everman, 1896, 1900 dans Magnin, 1959 a). Il remonte notamment les fleuves St Laurent, Delaware, Apalachicola (Wooley et Crateau, 1985) et Mississipi (Vladykov, 1955 dans Magnin, 1959 a). La forme *A. oxyrhynchus desotoi* se rencontre du delta du Mississipi à la rivière Suwanee en Floride (Conte et al., 1988). Les mâles atteignent la maturité sexuelle vers 22-34 ans (1,65 m) dans le St Laurent (Scott et Crossman, 1974), 11-20 ans (1,2-1,8 cm) dans l'Hudson (Dovel, 1979 dans Smith, 1985), 12-28 ans au Québec (Therrien, 1988), 7-9 ans dans la Suwanee (Hoff, 1975 dans John et al., 1978), 5-13 ans en Caroline du sud (Smith, 1985) et les femelles vers 27-28 ans (1,9 m) dans le St Laurent (Scott et Crossman, 1974), 20-30 ans (1,8-2,45 m) dans l'Hudson (Dovel, 1979 dans Smith, 1985), 7-19 ans en Caroline du sud (Smith, 1985), 23-36 ans au Québec (Therrien, 1988). Les poissons mâtures quittent la mer au printemps pour se reproduire en rivière (Leim et Scott, 1972). La reproduction a lieu tous les 1 à 5 ans pour les mâles et 3 à 5 ans pour les femelles (Smith, 1988) à des périodes variables selon les régions et ceci en fonction de la température (Smith, 1985). Elle a lieu par exemple en mai-juin dans le fleuve St Laurent. Les zones de frayères se trouvent à l'aval des fleuves, selon Smith (1985) parfois en limite de salinité, dans des zones profondes (11-13 m) à courant modéré (0,5-0,8 ms^{-1}) (Crance, 1987) et à fond dur (Conte et al., 1988) recouvert de blocs rocheux (Smith, 1985). Les ovules, de couleur brun-foncé, presque noire mesurent de 2,1 à 2,5 mm de diamètre d'après Leim et Scott (1972), de 2,5 à 3 mm d'après Smith (1985). Les oeufs démersaux, fortement adhésifs collent au substrat, blocs, plantes, racines (Smith, 1985). Les géniteurs retournent en mer en été (Bigelow et Schroeder, 1953). Les juvéniles dont les déplacements en eaux continentales semblent complexes quittent les eaux douces pour la mer vers 4-6 ans, ils y restent jusqu'à leur maturité sexuelle (Conte et al., 1988). En eau douce les jeunes individus se nourrissent de larves d'insectes et de cladocères, puis lorsqu'ils sont plus gros d'amphipodes et d'isopodes. En mer les *A. oxyrhynchus* mangent des mollusques, des annélides et quelques poissons, notamment des lançons, *Ammodytes sp.* (Leim et Scott, 1972). Les adultes ne se nourrissent pas au cours de la migration de reproduction (Bigelow et Schroeder, 1953).

Acipenser ruthenus Linné, 1758

(Bulgare) chiga, (Croate) keciga, (Danois) sterlet, (D) sterlet, (E) sterlet, (F) sterlet, (Hongrois) kecsege, (Polonais) sterled, czeczuga, (Roumain) cega, (R) sterlyad', (Slovaque) jeseter maly', (Tchèque) jeseter maly', (Turc) çuka, çiga baligi.

Le dos et les flancs sont de couleur gris-beige, les écussons latéraux ainsi que le ventre sont blancs.

Ecussons : 12-17L 57-71L 10-19V (Lebedev et al., 1969 dans Charlon et Williot, 1978). LTmax > 0,9 m (Maitland, 1987), 1,10 m (Tsepkin et Sokolov, 1979), 1,25 m et Pmax > 16 kg (Lebedev et al., 1969 dans Charlon et Williot, 1978). Son museau est étroit et pointu, il possède quatre barbillons longs et frangés. Son caryotype est de 2n = 118 ± 2 chromosomes (Birstein et Vassiliev, 1987) ou 118 ± 4 (Rab, 1986). Certains distinguent une forme *A ruthenus marsigli* qui fréquente les rivières de Sibérie (Manea, 1966).

Cette espèce est principalement dulçaquicole; on la trouve dans les bassins de la mer Noire, de la mer d'Azov et de la mer Caspienne. Elle fréquente les rivières Dvina du nord, Ob, Irtych, Ienisseï, Volga, Niemen, Dvina occidentale, Onega, Mezen et Danube. Il existe des formes migratrices allant en eaux saumâtres (Lukin 1949) et parfois signalées dans la Baltique. La maturité sexuelle est atteinte pour les mâles vers 3-7 ans dans le Danube (Carausu, 1952 dans Manea, 1966), 4-5 ans (0,35 m) en URSS (Lebedev et al. 1969 dans Charlon et Williot 1978),et pour les femelles vers 7-9 ans (0,4-0,45 m) (Lebedev et al. 1969 dans Charlon et Williot 1978). Sa longévité serait d'une trentaine d'année (Lebedev et al. 1969 dans Charlon et Williot 1978) et la reproduction aurait lieu tous les ans (Manea 1966). Les géniteurs jeûnent durant l'hiver, au printemps ils migrent légèrement vers l'amont et se reproduisent en avril-juin, dans des zones profondes (10 m) à fond de graviers et à courant rapide avec des températures d'eau variant de 12 à 17 °C (Manea 1966). Les oeufs de 1,9 à 2,5 mm de diamètre sont adhésifs (Detlaf et al., 1981). Les jeunes dévalent vers le cours inférieur des rivières.

Acipenser schrenki **Brandt, 1869**

(E) Amur sturgeon, (F) schrenki, esturgeon de l'Amour, (Japonais) Amūru chōzame, (R) Amurskij osetr.

Ecussons : 11-17D 32-47L. LTmax > 2,9m (Berg 1962), Pmax > 200 kg (Berg, 1962). Son rostre est pointu, les barbillons sont frangés et la lèvre inférieure est fendue. Son caryotype est de 2n = 240 chromosomes (Vasil'ev et al., 1981).

On le trouve avec le *Huso dauricus* dans le bassin de l'Amour; c'est une espèce principalement dulçaquicole avec des populations amphihalines dans le liman * de l'Amour (Rostami, 1961; Kozhin, 1964). Il fréquente le lac Khanka (Berg, 1962) et a été signalé dans la mer d'Okhotsk (Berg 1904 dans Magnin; 1959 a), mais il semble ne plus y être présent. Les mâles atteignent la maturité sexuelle vers 9-10 ans (1,08-1,16 m pour 6 kg) et les femelles vers 13 ans. La période de reproduction se situe de mai à la fin juin. La croissance des individus est plus rapide dans le liman que dans le fleuve Amour.

Acipenser sinensis **Gray, 1834**

(E) chinese sturgeon, zonghua sturgeon (F) esturgeon chinois, (Japonais) kara chōzame.

La peau est lisse entre les rangées d'écussons. Ecussons : 12-14D, 14-28L, LTmax > 2 m, Pmax > 500 kg (Huang Hongjin et al., 1982).

Cette espèce amphihaline se trouve dans les bassins du Yang-tse-kyang de la rivière Min et du Liao-ho dans le nord-est de la Chine (Magnin, 1959 a; Doroshov 1985), elle fréquente la mer Jaune et le sud de la Corée (Huang Hongjin et al., 1982). Les animaux atteignent la maturité sexuelle à 20 kg pour les mâles et 120 kg pour les femelles (Huang Hongjin et al., 1982); ils ne s'alimentent pas durant la période de reproduction qui va de début octobre à début novembre. Les jeunes se nourrissent de

* estuaire externe de certains fleuves asiatiques présentant un cordon littoral.

petits invertébrés benthiques et les adultes d'insectes et de diatomées (Huang Hongjin et al., 1982).

Acipenser stellatus Pallas, 1771

Synonymes : *A. helops* Pallas, 1811.
(Bulgare) pastruga, (D) sterg, sternhausen, sternstör, (E) stellate sturgeon, (Estonien) sevrjuuga, (F) esturgeon étoilé, sevruga, (Hongrois) pastruga, sõregtok, (Iranien) ouzoun-bouroun, (Italien) storione stellato, (Lithuanien) zvaigzdétasis ersketas, (Polonais) cego, siewruga, (Roumain) pâstruga, (R) sevryuga, (Serbo-croate) dunavska jesetra, pachtrouga, sevrljuga, zvjezdan, (Slovaque) jeseter hvieznaty', (Tchèque) jeseter hvezdnaty', (Turc) mersin baligi.

De nombreuses plaques claires en losange sont disséminées entre les rangées d'écussons. Ecussons : 9-16D 26-43L 9-14V (Lebedev et al., 1969 dans Charlon et Williot, 1978), 30-38L (Kostomarov, 1933). Le museau est extrêmement allongé (>60% de la longueur de la tête), la lèvre inférieure est fendue, les barbillons sont courts et peu frangés. LTmax > 1,7 m d'après Kostomarov (1933), 2,2 m (Lebedev et al., 1969 dans Charlon et Williot, 1978), Pmax 30-32 kg d'après Kostomarov (1933), > 80 kg d'après Lebedev et al., 1969 dans Charlon et Williot, 1978. Son caryotype est de 2n = 118 ± 2 chromosomes (Birstein et Vasil'ev, 1987). Certains auteurs distinguent plusieurs formes géographiques : *A. stellatus illyricus* Brusina, 1902, en Dalmatie; *A. stellatus danubialis* Brusina, 1902, dans le Danube; *A. stellatus donensis* Lovetzky, 1834, dans le Don; *A. stellatus cyrensis*. dans l'Adriatique (Manea, 1966). Ces distinctions reposent essentiellement sur des critères phénotypiques.

Cet amphihalin fréquente les bassins de la mer d'Azov, de la mer Caspienne et de la mer Noire. Il remonte notamment les fleuves Volga, Kura, Oural, Terek et Safi-Rud (Caspienne), Dniestr, Dniepr, Danube et Rioni (mer Noire), Kuban et Don (mer d'Azov). Cette espèce est également acclimatée en mer d'Aral (Kozhin, 1964; Lebedev et al., 1969 dans Charlon et Williot, 1978). La maturité sexuelle est atteinte pour les mâles vers 7 ans dans le Don et le Kuban (Tarasyuk, 1964), 9-13 ans, lieu non précisé (Lebedev et al., 1969 dans Charlon et Williot, 1978), 7 ans (1,05 m et 3-4 kg) dans la Volga (Veshev et Novikova, 1986 b) et pour les femelles vers 8-9 ans dans le Don et le Kuban (Tarasyuk, 1964), 11-17 ans lieu non précisé (Lebedev et al., 1969 dans Charlon et Williot, 1978), 9 ans (1,2 m et 9-10 kg) dans la Volga (Veshev et Novikova, 1986 b). La reproduction a généralement lieu de mai à août dans la Volga, de mi avril à mi septembre dans la Kura, de fin avril à fin août dans le Kuban et de mai à juin dans le Don. On distingue deux périodes de migration des géniteurs dans le Danube. Les migrants de printemps (mars-mai) qui se reproduisent au printemps de la même saison et les migrants d'automne (août-septembre) qui passent l'hiver en rivière dans des fosses et se reproduisent seulement au printemps suivant (Manea, 1966). Les zones de frayères se situent plus à l'aval et dans des zones de courant plus faible que les autres espèces d'esturgeons (Vlasenko, 1974). Le diamètre des ovules est de 2,8-3,0 mm (Lebedev et al, 1969 dans Charlon et Williot, 1978), il y en a de 60 à 80 au gramme (Kozhin, 1964), les oeufs sont adhésifs (Detlaf et al., 1981). Les jeunes se nourrissent de larves d'insectes d'annélides, de mollusques et de crustacés (Manea, 1966).

Acipenser sturio Linnaeus, 1758

Synonymes : *Acipe esturgeon* Daubenton, *Antaceus yarrellii* Dum., *Acipenser loevissimus* Val., *Huso valenciennii* Dum..
(Albanais) blini, (Basque) gaizkata, (Bulgare) nemska esetra, atlantichka esetra, (Danois) stǿr, (D) stör, baltischer stör, gemeiner stör, (E) common sturgeon, sea sturgeon, sturgeon, (Espagnol) esturión, gaizcata, garz cata, marión, sollo, sollo real, (Estonien) tuur, (Feröe) styrja, (Finlandais) sampi, (F) esturgeon commun, esturgeon

d'europe, (Gaelic) bradán fearna, (Grec) akipissios, mouroúna, stourioni, xirichi, (Hongrois) szintok, közönséges tok, (Italien) storione, storione comune, storione reale, (Islandais) styrja, (Lethonien) stohre, store, stuhre, (Lithuanien) asetras, atlánto ersketas, ersketras, sturys, (Néerlandais) steur, (Norvégien) stφr, (Polonais) jesiotr zachodni, czezuga, (Portugais) esturjão, peixe rei, sôlho, (Roumain) sipul, sip, vizã galbena, (R) atlantiskij osetr, nemetskij osetr, baltiskij osetr, obiknovenij osetr, (Serbo-croate) atlantska jesetra, jesetric', storijun, strljun, (Slovaque) jeseter vel'ky, atlanski jeseter, (Suédois) stör, (Tchèque) jeseter velky', (Turc) kolan, açl mersine, kolan baligi, (Ukrainien) atlantiskij osetr, zbiouainij osetr.

Noms vernaculaires français : (Bretagne) créac'h, (Noirmoutier) étrugeon, (Vendée, Gironde) créac, créa, (Côte basque) gaizkata, (Port Vendres) astouriou, esturio, (Sète) esturjoun, esturien, (Provence) esturioun, (Nice) sturion.

Dos de couleur gris rosé à gris-beige et ventre blanc, les viscères sont légèrement grisâtres (Magnin, 1964). Ecussons : 12-13D 35L 11V en Gironde (Magnin, 1962), 9-15D 24-36L 10-12V dans le Rioni (Lebedev et al., 1969 dans Charlon et Williot, 1978), 10-15D 29-38L 10-12V en Gironde (Rochard, données. pers.).Le nombre de branchiospines sur le premier arc varie de 16 à 26 (Magnin 1962) Le museau est allongé et pointu, les barbillons sont non frangés, la lèvre inférieure est nettement fendue au milieu et les écussons ne sont pas plus de deux fois plus haut que large. LTmax 5 m (Laporte, 1853) (actuellement plutôt 2,5 m), Pmax $\geq$ 200 kg (Lebedev et al., 1969 dans Charlon et Williot, 1978). Le caryotype est de 2n = 116 ± 4 chromosomes pour les *A. sturio* issus de l'Adriatique (Fontana et Colombo, 1974).

Cette espèce amphihaline est actuellement présente dans deux aires de répartition relictuelles disjointes : la première s'étend des côtes portugaises aux côtes scandinaves en passant par les îles britanniques, la seconde est constituée par une partie de la mer Noire : le bassin de la rivière Rioni (Ninua, 1976) et la zone marine proche. Une forme dulçaquicole habitant le lac Ladoga (URSS) est parfois citée dans la littérature (Lebedev et al., 1969 dans Charlon et Williot, 1978; Wheeler, 1978). Autrefois on rencontrait *A. sturio* en méditerranée dans le bassin du Rhône et près des côtes marocaines (Furnestin et al., 1958), tunisiennes (Heldt, 1934) et espagnoles (Classen, 1944). Elle a également été signalée dans l'Adriatique (Paccagnella, 1948 dans Magnin, 1959 a; Fontana et Colombo, 1974) et dans le bassin du Danube (Antipa, 1934). Les mâles deviennent mâtures vers 14-18 ans (145 cm et 20 kg) en Gironde (Magnin, 1962), 7-12 ans dans le Rioni (Elanidze et al., 1970); et les femelles vers 16-18 ans (165 cm et 30 kg) en Gironde (Magnin, 1962), 8-14 ans dans le Rioni (Elanidze et al., 1970). Les adultes ne se nourrissent pas au cours de la migration de reproduction, ils quittent la mer au printemps et remontent les fleuves vers les zones de frayères; la reproduction a lieu au mois de mai-juin (animaux de l'Atlantique et de la Mer noire) ou en juin-juillet (animaux de la Baltique). Les oeufs de 2,6 à 3 mm de diamètre sont adhésifs (Lebedev et al., 1969 dans Charlon et Williot, 1978; Elanidze et al,. 1970; Williot, données pers.). Les alevins restent en estuaire au moins un an avant de partir en mer. Les juvéniles reviennent en zone estuarienne au cours de migrations estivales (Magnin, 1962; Castelnaud et al., 1990). Cette espèce se nourrit d'invertébrés benthiques, annélides (*Aphroditidae*), crustacés (*Crangon crangon, Carcinus maenas*) (Magnin, 1962; Rochard, Observ. pers.).

Acipenser transmontanus **Richardson, 1836**

Synonymes : *A. aleutensis* Heckel et Fitzinger, 1836.
(D) weißer stör, (E) white sturgeon, pacific sturgeon, (F) esturgeon blanc.

Cette espèce est de couleur claire de gris à gris brunâtre. Ecussons : 11-14D 38-48L 9-12V (Scott et Crossman, 1974). LTmax $\geq$ 3 m d'après Scott et Crossman (1974), 6,1 m d'après Malm (1980), Pmax 816,5 kg (Malm, 1980).

Cette espèce migratrice présente des populations réellement amphihalines et d'autres euryhalines qui restent dans les fleuves et les estuaires, par exemple la partie inférieure du fleuve Columbia (Carl et al., 1967; Haynes et al., 1978). Il existe également plusieurs populations bloquées à l'amont de barrages sur les rivières Snake (Cochnauer et al., 1985) et Columbia (Haynes et al., 1978). Elle fréquente les côtes pacifiques de l'Amérique du nord de l'Alaska à la Californie centrale et les fleuves Fraser, Columbia, Snake et Sacramento-San Joaquin. La maturité sexuelle est atteinte vers 12 ans (1,2 m) pour les mâles et 15-20 ans (1,5 m) pour les femelles (Galbreath, 1985; Anderson, 1988) et la reproduction a lieu tous les 2 à 8 ans (Conte et al., 1988). Les déplacements de cette espèce sont complexes mais semblent liés à la taille des individus et à la température de l'eau (Haynes et al., 1978). Les adultes quittent l'océan et les estuaires pour remonter les fleuves, la fraie a lieu dans des zones profondes à fond de graviers et de blocs (Conte et al., 1988), les oeufs de 3,8 mm de diamètre (Lutes et al., 1987) sont adhésifs (Doroshov, 1983). Cette espèce se nourrit d'invertébrés benthiques, *Crangon sp, Palaemon macrodactylus* (Kohlhorst et al., 1980), les plus grands individus se nourrissent parfois de poissons (Conte et al., 1988). Les populations sont bien suivies dans certaine zones notamment la Californie mais la période alevin reste relativement mal connue. Cette espèce est protégée dans l'Idaho et le Montana (Johnson, 1987).

Genre *Huso*

Huso dauricus Georgi, 1775

Synonyme : *A. orientalis* Pallas.
(Chinois) sinkhuan-you, (E) siberian great sturgeon, (Japonais) dauria chōzame, (R) kaluga.

LTmax 5,6 m, Pmax > 1000 kg (Kozhin, 1964). Son caryotype est de 2n = 120 chromosomes (Burtzev et al., 1976)

Essentiellement dulçaquicole cette espèce fréquente le bassin de l'Amour; il y aurait comme pour l'*A. schrenki* des populations strictement dulçaquicoles et d'autres amphihalines qui fréquentent le fleuve et son liman (Rostami, 1961, Lukyanenko et al., 1978). Certains individus ont été capturés sur les côtes japonaises d'Hokaïdo (Kunio et Kazuhiro, 1975 dans Gritsenko et Kosmyunin, 1979). Les mâles atteignent la maturité sexuelle vers 18 ans selon Soldatov (1915 dans Magnin, 1962), 14-21 ans selon Krykhtin (1987), et les femelles 20 ans (2,3 m) selon Soldatov (1915 dans Magnin, 1962), 17-23 ans selon Krykhtin (1987). Kozhin (1964) indique l'acquisition de la maturité sexuelle vers 16-17 ans pour les deux sexes. La longévité de cette espèce dépasserait 55 ans (Kozhin, 1964).

Huso huso (Linnaeus, 1758)

Synonymes : *H. ichthyocolla* Bonaparte, 1846; *A. huso* Linnaeus, 1758; *A. vallisnerii* Molin, 1853.
(Bulgare) morouna, (D) hausen, europaïscher hausen, (E) russian beluga, giant sturgeon, great sturgeon, (Estonien) beluuga, (F) bélouga, grand esturgeon, (Grec) akipïssios, (Hongrois) viza, (Italien) storione ladano, (Iranien) fil mahi, (Lithuanien) ersketas, sturys, (Norvégien) beluga fisk, (Polonais) wiz, wyz, (R) bélouga, (Roumain) hurion, morunul, morun, (Serbo-croate) beljuga, bjeluga, moruna, sip, viza, (Slovaque) vyza, (Suédois) hussblosstōr, (Tchèque) vyza velká, (Turc) mersinmorinasi baligi.

Ecussons : 60L (Kostomarov, 1933), 11-14D 41-52L 9-11V (Lebedev et al., 1969 dans Charlon et Williot, 1978). LTmax 8,5 m d'après Marx (1987), $\geq$ 5 m d'après Lebedev et al. (1969, dans Charlon et Williot, 1978), Pmax > 1000 kg d'aprés Lebedev et al. (1969, dans Charlon et Williot, 1978),

1500 kg d'après Frank (1973), 1300 kg d'après Marx (1987). Le museau est assez court et en pointe, les barbillons non frangés sont aplatis, la bouche en forme de demi lune est grande et la lèvre inférieure est fendue au milieu. Il y a au minimum 60 rayons sur la nageoire dorsale (Lebedev et al. 1969 dans Charlon et Williot, 1978). Son caryotype est de 2n = 118 ± 2 d'après Birstein et Vasil'ev (1987) ou 2n = 116 ± 2 d'après Fontana et Colombo (1974) pour des individus de l'Adriatique. Certains auteurs distinguent plusieurs formes, le plus souvent selon des caractères méristiques ou morphologiques parfois de coloration... On trouve ainsi *H. huso maeoticus*, *H. huso ponticus natio occidentalis*, *H. huso ponticus natio orientalis* Salnikov et Maliatski, 1934 et *H. huso caspicus natio kurensis* Babouchkine, 1942.

On trouve cet amphihalin dans les Mers Caspienne, d'Azov et Noire, rarement dans l'Adriatique (Svetovidov, 1973). Il remonte les fleuves Volga, Oural, Kura, Terek (Caspienne), Dniepr, Dniestr, Bug, Danube, Rioni (mer Noire), Don, Kuban (mer d'Azov) (Lebedev et al., 1969 dans Charlon et Williot, 1978). Parfois il fréquente l'Adriatique et remonte le Pô (Magnin, 1959 a; Fontana et Colombo, 1974). Les mâles atteignent la maturité sexuelle vers 12-14 ans dans le Don, 13 ans dans la Kura, 12 ans dans la Volga, 14 ans dans l'Oural et les femelles vers 16-18 ans dans le Don, 18 ans dans la Kura, 15 ans dans la Volga et 18 ans dans l'Oural (Lebedev et al., 1969 dans Charlon et Williot, 1978), la taille à la maturité sexuelle est de 1,8 m pour les mâles et 2,0 m pour les femelles de la mer d'Azov (Tchugunova, 1940 dans Magnin, 1962). Les animaux qui pénètrent en rivière en automne passent l'hiver dans des fosses profondes et vont pondre au printemps, ceux qui remontent au printemps se reproduisent au cours de la même saison. La reproduction à lieu tous les 3-4 ans (Elanidze et al., 1970) dans des zones profondes (8-20 m selon Manea, 1966), à courant rapide et à fond de galets. Il y a de 35 à 50 ovules au gramme et les oeufs sont adhésifs (Lebedev et al., 1969 dans Charlon et Williot, 1978). Après la fraie les adultes retournent en mer rapidement, les alevins de l'année dévalent plus lentement. Leur croissance peut atteindre 45 cm durant la première année (Lebedev et al., 1969 dans Charlon et Williot, 1978). Les jeunes se nourrissent surtout de crustacés alors que les adultes consomment principalement des poissons *gobiidae* (Manea, 1966), le reste de leur alimentation est composé de crustacés *Crangonidae* et de mollusques (Romanycheva et Barybina, 1979 dans Williot, 1984). Généralement pélagiques ces animaux se situent en mer dans des zones de 50-60 m de profondeur. La longévité des animaux de cette espèce peut dépasser les 100 ans (Lebedev et al., 1969 dans Charlon et Williot, 1978).

Genre *Pseudoscaphirhynchus*.

Pseudoscaphirhynchus kaufmanni Bogdanov, 1874

(E) Amu Daria shovelnose, large shovelnose, (F) grand nez-pelle de l'Amou daria, (R) bolchoi Amudarinskij lopatonoss

LTmax 0,75 m, Pmax 2,5 kg. Cette espèce possède un très long filament caudal et de 1 à 5 épines à la partie supérieure du museau; ces épines s'atténuent avec l'âge (Frank, 1973). Les mâles sont une fois et demi plus grands que les femelles et ont un museau plus large (Kotsetov, 1982).

Cette espèce fréquente la mer d'Aral mais surtout ses affluents notamment le cours inférieur de l'Amou-Daria. elle se trouve dans des zones peu profondes (1-2 m) et de substrat sablonneux (Kotsetov, 1982). La maturité sexuelle est atteinte vers 6-7 ans à une taille de 0,4 m (Kozhin, 1964). Les ovules sont de couleur gris noirs et mesurent de 1,8 à 2,4 mm de diamètre (Kotsetov 1982). Les jeunes se nourrissent de larves d'insectes et les adultes de petits poissons (Frank, 1973).

Pseudoscaphirhynchus hermanni Kessler, 1877

(F) petit nez-pelle de l'Amou daria, (R) maliy amudarinskij lopatonoss.

LTmax 0,27 m. Cette espèce possède un très long filament caudal (Kozhin, 1964).

Elle fréquente la mer d'Aral mais surtout ses affluents notamment le bassin de l'Amou-Daria.

Pseudoscaphirhynchus fedtschenkoi Kessler, 1872

(F) nez-pelle du Syr daria, (R) Syr-darinskij lopatonoss.

Ces animaux possèdent un filament caudal pouvant atteindre les deux tiers de la longueur du corps. LTmax 0,65 m (Kozhin, 1964).

On les trouve dans la mer d'Aral et notamment le bassin du Syr-Daria.

Genre *Scaphirhynchus*

Scaphirhynchus albus Forbes et Richardson, 1905

(E) Pallid sturgeon.

De couleur brun clair sur le dessus et blanche sur le ventre. LTmax 1,8 m (Migdalski et Fichter, 1979), en moyenne 0,6 m pour les adultes. Il est plus grand que *S. platorhynchus*, Pmax 130 kg.

Strictement dulçaquicole il fréquente le bassin supérieur du Mississipi, rivières Mississipi et Cairo. Son alimentation en plus des larves d'insectes comprend beaucoup de poissons (*cyprinidae*). Il semble fréquemment s'hybrider avec *S. platorhynchus* (Carlson et al., 1985). La maturité sexuelle survient pour les mâles vers 3-4 ans (0,5-0,6 m) (Kallemeyn, 1983). Cette espèce très mal connue (Kallemeyn 1983) et beaucoup moins abondante que *S. platorhynchus* (Conte et al. 1988) est considérée comme menacée (Nordstrom et al., 1977; Conte et al., 1988), elle est protégée dans sept états américains (Johnson, 1987).

Scaphirhynchus platorhynchus Rafinesque, 1820

(E) shovelnose sturgeon, hackleback.

De couleur brun clair. LTmax 0,85 m, Pmax 4,5 kg (Lee 1980 d dans Conte et al., 1988). Les individus mesurent en général moins de 0,6 m. Son caryotype est de 2n = 112 chromosomes (Ohno et al., 1969 dans Fontana et Colombo, 1974). Les adultes fraient au printemps et au début de l'été sur des fonds rocheux dans des zones de courant rapide (Conte et al., 1988).

Cette espèce est strictement dulçaquicole, elle fréquente le bassin très pollué du Mississipi et notamment le Missouri. Absente des grands lacs, elle affectionne les zones de courants rapides, de 0,2 à 1,5 ms^{-1} (Modde et Schmulbach, 1977). Les animaux de moeurs nocturnes, se nourrissent essentiellement de larves d'insectes et atteignent la maturité sexuelle vers 5-7 ans (Lee, 1980) soit à une taille de 35 cm (Carlander, 1969 dans Conte et al., 1988).

Ils semblent s'hybrider fréquemment avec *S. albus* (Carlson et al., 1985). Cette espèce est protégée au Texas (Johnson 1987).

APPROCHE SYNTHETIQUE

Les tableaux et figures ci aprés résument les principales données écologiques des Acipenseridae. Lorsqu'une espèce n'apparait pas cela signifie que nous ne disposons pas des informations. les points communs, les différences et les particularités des espèces sont mis en évidence. Les références non citées dans les tableaux se retrouvent dans les paragraphes de présentation des espèces.

Les *Acipenseridae* sont largement répandus dans dans tout l'hémisphère nord (Figure 1). On trouve des espèces européennes : *A. sturio, A. nacarrii, A. stellatus, A. gueldenstaedti, A. ruthenus, A. nudiventris, H. huso*; des espèces asiatiques : *A. baeri, A. schrenki, A. sinensis, A. multiscutatus, A. dabryanus, A. kikuchii, H. dauricus, P. kaufmanni, P. hermanni,* et *P. fedtshenkoi*; une espèce à la fois asiatique et américaine (côte pacifique) *A. medirostris*;.des espèces américaines : *A. fulvescens, A. transmontanus, A. oxyrhynchus, A. brevirostrum, S. albus* et *S. platorhynchus*. Tous les *Acipenseridae* effectuent des migrations, certains se cantonnent dans les eaux douces, la plupart s'aventurent dans les eaux saumâtres des estuaires et d'autres vont même passer une partie de leur vie en mer. Cependant il est excessivement difficile de les classer, car à l'intérieur d'une même espèce on trouve souvent différents type écologiques de populations (Tableau 1).

Pour certains le genre *Pseudoscaphirhynchus* est strictement dulçaquicole et endémique dans les tributaires de la mer d'Aral (Nikolskij, 1957 dans Magnin, 1959 a), pour d'autres il fréquente également les eaux salées de cette mer (Gosteeva, 1953 dans Magnin, 1959 a). Vu l'état de dégradation écologique de la mer d'Aral on peut considérer ce genre comme extrêmement menacé, certaines espèces sont peut-être même déja éteintes.

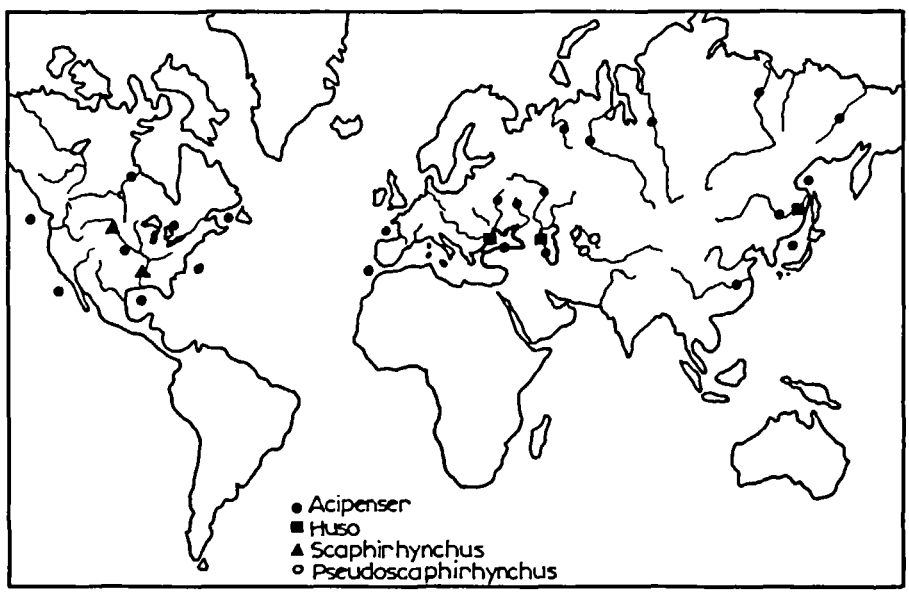


Figure 1 - Carte de répartition des esturgeons, d'après Magnin, 1962.

Tableau 1 - Types écologiques de populations et aires de répartition des espèces d'esturgeons. Pour chaque espèce nous avons indiqué en gras le type écologique le plus fréquent, mais pour la plupart des espèces il existe d'autres types de populations.

Espèces	Type de populations et aire de répartition
A. baeri	Populations dulçaquicoles ou faiblement euryhalines fleuves sibériens (Votinov et Kas'yanov, 1974). Populations dulçaquicoles de lacs (Zaïsan et Baïkal)(Berg, 1962) Introduit en Baltique (Kairov et Kostrichkina, 1970).
A. brevirostrum	Populations dulçaquicoles ou euryhalines Fleuves des côtes est américaines et canadiennes (Dadswell, 1979; Scott et Scott, 1988).
A. dabryanus	Populations dulçaquicoles Fleuves Yang Tsé Kiang et Houang Ho (Magnin, 1959 a; Huang Hongjin et al., 1982).
A. fulvescens	Populations dulçaquicoles Rivières et lacs du nord est de l'Amérique du nord (Magnin, 1959 a; Scott et Crossman, 1974).
A. gueldenstaedti	Populations amphihalines Bassins des mers Noires, Azov et Caspienne (Kozhin, 1964; Manea, 1966). Sous populations migrant à l'automne ou au printemps (Artyukhin, 1988).
A. kikuchii	Japon (Rostami, 1961; Matsuraba, 1955 dans Suzuki et Nishi, 1977).
A. medirostris	Populations amphihalines Pacifique nord, U.S.A., Canada, U.R.S.S., Chine, Japon (Magnin, 1959; Kozhin, 1964; Scott et Crossman, 1974; Conte et al. 1988).
A. multiscutatus	Japon (Rostami, 1961; Matsuraba, 1955 dans Suzuki et Nishi, 1977).
A. naccarii	Populations amphihalines Nord de l'Adriatique (Magnin, 1959 a; Tortonese, 1989).
A. nudiventris	Populations amphihalines Bassins des mers Noire, Azov, Caspienne et Aral. Quelques populations dulcaquicoles et euryhalines (Kozhin, 1964; Manea, 1966).
A. oxyrhynchus	Populations amphihalines Côtes est et nord-est de l'Amérique du nord (Scott et Scott, 1988).
A. ruthenus	Populations dulçaquicoles Fleuves sibériens et tributaires des mers Noire, Azov et Caspienne (Kozhin, 1964; Sokolov et Vasil'ev, 1989).
A. schrenki	Populations dulçaquicoles ou euryhalines Bassin de l'Amour (Kozhin, 1964).

A. sinensis	Populations amphihalines Bassin du Yang Tsé Kyang (Magnin, 1959 a; Huang Hongjin et al., 1982).
A. stellatus	Populations amphihalines Bassins des mers Noire, Azov et Caspienne Introduit en mer d'Aral (Kozhin, 1964; Shubina et al., 1989).
A. sturio	Populations amphihalines Europe occidentale, mers Méditerranée et Noire (Magnin, 1959 a; Ninua, 1976) Il subsisterait une population bloquée dans le lac Ladoga (Lebedev et al., 1969 dans Charlon et Williot, 1978).
A. transmontanus	Populations euryhalines Côtes ouest américaines et canadiennes (Scott et Crossman, 1974). Il existe des populations bloquées, notamment sur le fleuve Columbia (Galbreath, 1985).
H. dauricus	Populations principalement dulçaquicoles, Bassin de l'Amour. (Kozhin, 1964). Quelques incursions en zone estuarienne.
H. huso	Populations amphihalines Bassins des mers Noire, Azov, Caspienne et Méditérranée. Sous populations migrant au printemps et à l'automne (Kozhin, 1964; Pirogovskii et al., 1989).
P. kaufmanni	Populations essentiellement dulçaquicoles. Amou Daria (tributaire de la mer d'Aral). Quelques incursions en zone estuarienne (Kozhin, 1964).
P. hermanni	Population dulçaquicole. Amou Daria (Kozhin, 1964).
P. fedtshenkoi	Population dulçaquicole. Syr Daria (Kozhin, 1964).
S. albus	Population dulçaquicole Bassin Mississipi-Missouri (Carlson et al., 1985).
S. platorhynchus	Population dulçaquicole Bassin Mississipi-Missouri (Carlson et al., 1985).

Les principaux paramètres biométriques montrent de très grandes variations, interspécifiques mais également intraspécifiques. De façon générale l'acquisition de la première maturité sexuelle est relativement tardive. En ce qui concerne la taille, on peut distinguer de petites espèces (genres *Pseudoscaphirhynchus* et *Scaphirhynchus*, *A. ruthenus*, *A. dabryanus* et *A. brevirostrum*) et de très grandes espèces (genre *Huso*), entre ces deux extrêmes la plupart des espèces approchent ou dépassent les 2 m et 100 kg (Tableau 2).

Tableau 2 - Données biométriques pour chaque espèce, valeurs minimales lors de la première maturité sexuelle et données maximales (LT : longueur totale).

Espèce	A la 1ère maturité sexuelle			Maximum	
	Age (année)	LT (m)	Poids (kg)	LTmax (m)	Pmax (kg)
A. baeri				3,0	200
Mâles	9	0,6-0,8	0,73		
Femelles	10	0,6-0,9	0,73		
A. brevirostrum				1,3	45
Mâles	5	0,45-0,5			
Femelles	6	0,45-0,5			
A. dabryanus				-	-
Mâles			5		
Femelles			10		
A. fulvescens				2,4	140
Mâles	16	0,85-0,95	4-5		
Femelles	20	0,9-1,2	4-9		
A. gueldenstaedti				2,3	100
Mâles	8	1,0	3		
Femelles	10	1,2	9		
A. kikuchii				-	-
A. medirostris				2,0	160
Mâles-Femelles		1,0	8-10		
A. multiscutatus				-	-
A. naccarii				-	-
A. nudiventris				2,0	80
Mâles	6				
Femelles	12				
A. oxyrhynchus				4,2	367
Mâles	7	1,2			
Femelles	8	1,8			
A. ruthenus				1,1	16
Mâles	3	0,35			
Femelles	7	0,4-0,45			
A. schrenki				2,9	200
Mâles	9	1,1	6		
Femelles	13				
A. sinensis				> 2,0	500
Mâles			40		
Femelles			120		
A. stellatus				2,2	80
Mâles	7	1,05	3-4		
Femelles	8	1,2	9-10		
A. sturio				5,0	500
Mâles	7	1,45	20		
Femelles	8	1,65	30		
A. transmontanus				6,1	816
Mâles	12	1,2			
Femelles	15	1,5			
H. dauricus				5,6	1000
Mâles	14	2,3			
Femelles	16	2,3			

H. huso			8,5	1300
Mâles	12	1,8		
Femelles	16	2,0		
P. kaufmanni			0,7	2,5
Femelles	6	0,4		
P. hermanni			0,3	
P. fedtschenkoi			0,7	
S. albus			1,8	130
Mâles	3	0,5		
S. platorhynchus			0,9	4,5
Mâles-femelles	5	0,35		

Quelque soit le type écologique de la population, la reproduction des *Acipenseridae* a toujours lieu en eau douce, généralement au printemps ou au début de l'été, dans des eaux avoisinant les 15°C. Le substrat est constitué préférentiellement de graviers et de blocs, la profondeur est d'au moins quatre mètres et le courant est vif 1 ms^{-1}.

Tableau 3 -Période de fraye et caractéristiques des frayères d'*Acipenseridae*.

Espèces	Température (°C)	Epoque de reproduction	Nature du fond	Profondeur (m)	Vitesse du courant (ms^{-1})
A. baeri	11-16 (Berg, 1962;	Fin printemps, été	Blocs ou gravier Sokolov	6-8 et	1,4 Malyutin, 1977)
A. brevirostrum	9-15 (Taubert, 1980; Dadswell, 1984; Scott et Scott, 1988)	Fin hiver-printemps	Gravier		0,4-0,6
A. fulvescens	13-18 (Magnin, 1966; Scott et Crossman 1974)	Fin printemps	Blocs ou gravier	< 3,5	Rapide
A. gueldenstaedti	8-15 (Vlasenko et al., 1989)	Printemps	Blocs, gravier sable	4-10	1-1,5 (Tanasijchuk, 1964 dans Vlasenko et al., 1989)
(*A. persicus*)	20-22 (Artyukhin, 1979)	Eté			
A. medirostris	8,9-16,7 (Houston, 1987; Scott et Crossman, 1974; Conte et al. 1988)	Fin printemps	Rocheux		Rapide
A. naccarii		Printemps (Maitland, 1987)			
A. nudiventris		Printemps (Borzenko, 1950 dans Sokolov et Vasil'ev, 1989)	Galets		1-2
A. oxyrhynchus	13,5-18 (Scott et Crossman, 1974; Smith, 1985; Crance, 1987; Scott et Scott, 1988)	Printemps-été	Blocs	11-13	0,5-0,8

A. ruthenus	12-17	Printemps	Galets et gravier	≥ 10	1,5-5
	(Jankovic, 1958; Manea, 1966; Sokolov et Vasil'ev, 1989)				
A. schrenki		Printemps			
		(Kozhin, 1964)			
A. sinensis		Automne			
		(Huang Hongjin et al., 1982)			
A. stellatus	15-26	Printemps-été	Blocs, graviers	2-14	1,2-1,5
	(Tanasijchuk, 1964; Vlasenko, 1981 dans Shubina et al., 1989)				
A. sturio		Printemps-été	Blocs, galets	> 5	1,5-2,0
					(Ninua, 1976)
H. huso	7-12	Printemps	Galets	4-20	1,1-1,9
		(Manea, 1966; Tanasijchuk, 1964; Pirigovskii et al., 1989)			
P. kaufmanni	14-16				
	(Nikolskim, 1938 dans Makeeva et al. 1964)				

L'intervalle de temps entre deux frayes successives est très variable, mais il est généralement plus court chez les mâles que chez les femelles. Les variations intraspécifiques sont très importantes, on remarque notamment d'importants écarts selon les zones géographiques. Il faut noter que cet intervalle de temps est déduit d'observations faites sur les pièces osseuses et que l'interprètation est délicate.

Tableau 4 - Intervalle de temps entre deux frayes successives (en années).

Espèces	Zones géographiques	Mâles	Femelles	Auteurs
A. baeri	Sibérie	1-2	3-4	(Kozhin, 1964)
		2-4	3-6	(Sokolov, 1965)
A. brevirostrum	Côtes américaines	3-8		(Taubert, 1980)
				(Dadswell et al., 1984)
A. fulvescens	Québec	2-3	4-6	(Magnin, 1966)
	Lac St Louis	9-10		(Goyette et al. 1987)
A. gueldenstaedti	Danube		> 6	(Kukuradze et al., 1975 dans Vlasenko et al., 1989)
	Volga	2-3	5	(Krasikov, 1981 dans Vlasenko et al., 1989)
A. oxyrhynchus	Côtes américaines	1-5	3-5	(Smith, 1988)
A. ruthenus	Danube	1	1	(Manea, 1966)
			1-2	(Jankovic, 1958)
A. stellatus	Caspienne	3-4		(Kazancheev,1981 dans Shubina et al., 1989)

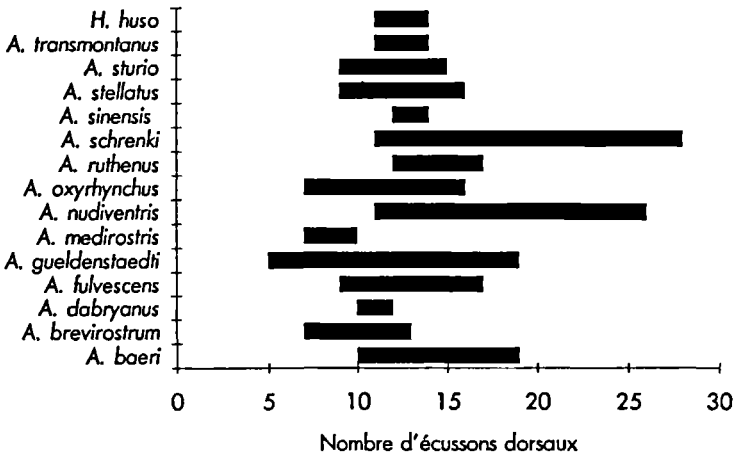
A. transmontanus	Columbia			2-11	(Galbreath, 1979 dans Cochnauer et al. 1985)
	Fraser	4-11		6-8	(Semakula et Larkin, 1968 dans Cochnauer et al., 1985
H. huso	Mers Noire et Caspienne		3-4		(Elanidze et al., 1970)
	Mer d'Azov	4,6	5	5,5	(Makarov, 1970)
P. kaufmanni	Amou Daria			3-4	(Makeeva et al., 1964)

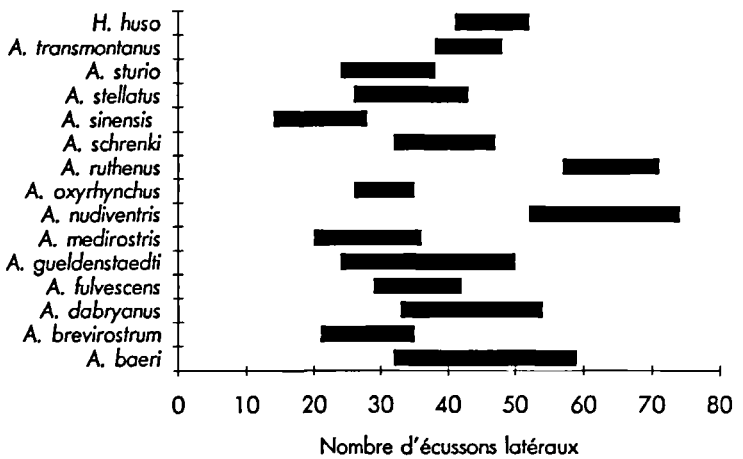
Le diamètre des ovules varie de 1,5 à 4,0 mm selon les espèces d'*Acipenseridae* (tableau 5). La taille des ovules semble être corrélée positivement avec la taille.

Tableau 5 - Diamètre des ovules.

Espèces	Diamètre (mm)	Auteurs
A. baeri	2,5-2,7	(Detlaf et al., 1981)
A. brevirostrum	3,0-3,2	(Dadswell et al., 1979 dans Dadswell et al., 1984)
A. fulvescens	2,7	(Barney, 1924 dans Magnin, 1962)
	3,0-3,5	(Czeskleba et al., 1985)
A. gueldenstaedti	3,0-3,5	(Detlaf et al., 1981)
A. g. colchicus	3,2-3,8	(Detlaf et al., 1981)
A. naccarii (*)	2,2-2,4	(Arlati et al., 1988)
A. oxyrhynchus	2,6	(Ruder, 1890 et Borodon, 1925 dans Magnin, 1962)
	2,1-2,5	(Leim et Scott, 1972)
	2,5-3,0	(Smith, 1985)
A. ruthenus	1,9-25	(Detlaf et al., 1981)
A. stellatus	2,7-3,2	(Detlaf et al., 1981)
A. sturio	2,6-2,8	(Mohr, 1958 dans Magnin, 1962)
	3,0	(Tchalikov, 1949 dans Magnin, 1962)
	2,6	(Williot ,données non publiées)
A. transmontanus	3,8	(Lutes et al., 1987)
H. dauricus	3,6-4,0	(Detlaf et al., 1981)
H. huso	3,6-4,0	(Detlaf et al., 1981)
P. kaufmanni	1,5-2,7	(Makeeva et al., 1964)

(*) : ces ovules ont été pondus par des femelles d'origine sauvage mais élevées plusieurs années en pisciculture.





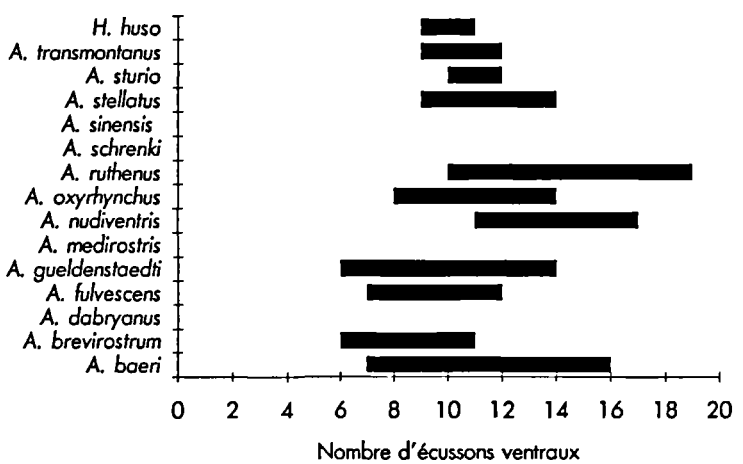


Figure 2 - Nombre d'écussons sur les rangées dorsale, latérales et ventrales.

Parmi les espèces pour lesquelles le caryotype a été déterminé (Tableau 6), on distingue deux groupes: un premier groupe de 7 espèces avec 2n ≈ 120 et un second groupe de 4 espèces avec 2n ≈ 240.

Tableau 6 - Nombre de chromosomes.

Espèces	2n	Auteurs
A. baeri	249 ± 5	(Vasil'ev et al., 1981)
A. gueldenstaedti	250 ± 8	(Birstein et Vasil'ev, 1987)
A. naccarii	239 ± 7	(Fontana et Colombo, 1974)
A. nudiventris	118 ± 3 118 ± 2	(Aref'ev, 1983) (Sokolov et Vasil'ev, 1989)
A. ruthenus	118 ± 2 118 ± 4	(Birstein et Vasil'ev, 1987) (Rab, 1986)
A. schrenki	≈ 240	(Vasil'ev et al., 1980)
A. stellatus	118 ± 2	(Birstein et Vasil'ev, 1987)
A. sturio	116 ± 4	(Fontana et Colombo, 1974)
H. dauricus	120	(Burtzev et al., 1976)
H. huso	116 ± 2 118 ± 2	(Fontana et Colombo, 1974) (Birstein et Vasil'ev, 1987)
S. platorhynchus	112 ± 5	(Ohno et al., 1969 dans Birstein et Vasil'ev, 1987.)

DISCUSSION ET PERSPECTIVES

Malgré certaines différences, notamment dans les habitats, on constate une relative homogénéité biologique et écologique à l'intérieur de la famille des *Acipenseridae*. Outre la morphologie très particulière des esturgeons, leurs caractéristiques de reproduction et d'alimentation par exemple, sont proches.

Cependant, il est aisé de remarquer que la quantité et la qualité des données disponibles sont très inégales selon les espèces, pour certaines, il y a un manque flagrant de connaissances biologiques de base, c'est le cas des esturgeons asiatiques en général et de l'esturgeon italien *A. naccarii*. Les espèces qui ont été ou sont encore exploitées, sont évidemment celles qui ont été les plus étudiées (*A. gueldenstaedti*, *A. stellatus*, *H. huso*), mais les données sont parfois anciennes ou parcellaires et demanderaient à être actualisées.

Des informations plus détaillées pourront être obtenues sur les *Acipenseridae* d'Europe dans l'ouvrage récent "General Introduction to Fishes, Acipenseriformes" Holcik J. Ed (1989) et pour certaines espèces dans des monographies spécialisées, par exemple concernant *A. brevirostrum*, la synthèse de Dadswell et al., (1984). Cependant pour la plupart des espèces *d'Acipenseridae* il subsiste des lacunes importantes, c'est un handicap insurmontable pour la mise au point d'une clé permettant l'identification de toutes les espèces.

Tableau 7 - Les hybrides d'esturgeons européens, naturels (en bas à gauche) et artificiels (en haut à droite) les plus fréquents.

	A.baeri	*A.gueldenstaedti*	*A.nudiventris*	*A.ruthenus*	*A.stellatus*	*H.huso*
A.baeri				Ab_f x Ar_m (2)		
A.gueldenstaedti			Artificiel viable (1), stérile (9)	Artificiel viable ◄–► (1) stérile (9)		Artificiel viable ◄–► (1) stérile (9,10)
A.nudiventris		Naturel (1,3,4) stérile (9)			Artificiel, viable fertile (9)	Artificiel Hh_f x An_m viable (9)
A.ruthenus	Naturel Sibérie Ob Ienissei (1,5,6)	Naturel fréquent Volga (1,3,5) stérile (9)	Naturel (4) ◄–►		Artificiel, viable fertile ◄–► (11)	Artificiel, viable F1 = Hh_f x Ar_m = Bester F2 fertile également $F1_m$ x Hh_f, viable $F1_f$ x Ar_m, viable (9)
A.stellatus		Naturel (1,3,7) stérile (9)	Naturel (1,4,7) fréquent Oural, Kura fertile (9)	Naturel (1,5,7) fréquent Volga, Don, Danube		
H.huso		Naturel, (1,3) Kama, Volga stérile (9)	Naturel (1,4) Kama	Naturel (1,8) Volga, Kama	Naturel (1,7) Kama	

(1) Kozhin, 1974. (2) Williot (données non publiées). (3) Vlasenko et al., 1989 a. (4) Sokolov et Vasil'ev, 1989 a. (5) Sokolov et Vasil'ev, 1989 b. (6) Sokolov et Vasil'ev, 1989 c. (7) Shubina et al., 1989. (8) Pirigovskii et al., 1989. (9) Burtzev dans Charlon et Williot, 1986. (10) Nikolyukin, 1966 dans Fontana et Colombo, 1974. (11) Nikolyukin, 1964.
f = femelle, m = mâle, ◄–► indique que les deux types d'hybrides ($_m$ x $_f$ et $_f$ x $_m$) ont été obtenus ou observés

Problèmes de systématique.

La systématique des *Acipenseridae* avec des figures marquantes Richardson, Rafinesque, mais surtout Duméril, date en grande partie du siècle dernier et repose sur la notion fixiste d'espèce typologique. Elle s'accorde très mal avec le polymorphisme, qu'il soit ontogénétique (différences morphologiques liées à l'age, variation dans la forme du rostre, peut être même dans le nombre des écussons), individuel ou intraspécifique (Daget, 1988). Ce polymorphisme intraspécifique, bien décrit par Roussow (1955) dans le cas d'A. fulvescens explique que certaines espèces, notamment celles ayant une vaste répartition géographique aient pu être identifiées sous plusieurs noms. C'est le cas pour *A. fulvescens* décrits sous au moins 17 noms différents par Duméril en fonction des localités d'origine,lors de l'année 1870, avant que des études plus approfondies et faites avec une autre vision de la notion d'espèce aient montré qu'il y avait synonymie! Pour certaines espèces,par exemple A. gueldenstaedti ce travail de différenciation reste à faire

La sous-famille des *Acipenserinae* comprend deux genres *Huso* et *Acipenser*. Pour le genre *Huso* les auteurs s'accordent unanimement à reconnaître deux espèces *Huso huso* et *Huso dauricus*. Par contre en ce qui concerne le genre *Acipenser* les choses sont moins claires.

Selon les auteurs (Berg 1932, Magnin 1959 a, Rostami 1961, Berg 1962, Scott et Crossman 1974, Doroshov 1985) on dénombre de 16 à 20 espèces pour le genre *Acipenser*. Certains font fréquemment de *A. sturio* et *A. oxyrhynchus* une seule et même espèce, d'autres attribuent des niveaux taxonomiques différents. Ainsi par exemple le taxon *A. persicus* Borodin, 1897, est considéré par certains comme une espèce à part entière (Artyukhin, 1988), le taxon *A. primigenius* Chalikov, 1944 n'est quasiment jamais plus utilisé depuis les année soixante (Chalikov 1944, Magnin 1959); le taxon *A. micadoi* Hilgendorf, 1932 est au plus considéré actuellement comme une sous espèce d'*A. medirostris* et le taxon *A. multiscutatus* n'est apparemment pas considéré comme une espèce par Doroshov (1985).

Pour notre part, faute d'informations indiquant à quelle espèce il se rapporte, nous retiendrons *A. multiscutatus* comme une espèce d'*Acipenser*. En ce qui concerne *A. oxyrhynchus et A. sturio*, Magnin (1964) a montré qu'il s'agissait de deux espèces distinctes et nous les considérerons comme telles. Ainsi nous aboutissons à 17 espèces dans le genre *Acipenser* : *A. baeri, A. brevirostrum, A. dabryanus, A. fulvescens, A. gueldenstaedti, A. kikuchii, A. medirostris, A. multiscutatus, A. nacarii, A. nudiventris, A. oxyrhynchus, A. ruthenus, A. schrenki,* A. *sinensis, A. stellatus, A. sturio* et *A. transmontanus*. Il faut cependant savoir que certaines espèces sont douteuses et que d'autres devraient peut-être être retenues.

La sous-famille des *Scaphirhynchinae* comprend pour tous les auteurs les deux genres et les cinq espèces que nous avons citées. Mais vu la situation écologique de la mer d'Aral, il est probable que certaines espèces, si ce n'est l'ensemble des espèces du genre *Pseudoscaphirhynchus* aient disparues.

Nous aboutissons au total à 24 espèces d'Acipenseridae.

Critères de détermination des espèces.

On peut distinguer trois principales catégories de critères d'identification actuellement utilisés pour les *Acipenseridae* :

- des critères écologiques ou géographiques (présence possible ou pas d'une espèce dans une zone ou dans un milieu donné) ;
- des critères morphométriques (proportions relatives de différentes parties de l'animal, nombre de rayons, d'écussons de branchiospines etc...)
- des critères autres (éthologie, coloration du corps ou des viscères, goût de la chair ou du "caviar"...).

Le plus souvent pour déterminer à quelle espèce appartiennent des individus issus du milieu naturel, on procède dans un premier temps par élimination, en fonction du secteur géographique de capture et des espèces susceptibles de s'y trouver. Dans un second temps on utilise certains critères morphométriques et si nécessaire les autres critères.

Cependant ces critères de détermination, valables à l'intérieur d'une zone géographique, ne permettent pas toujours d'identifier l'espèce avec certitude si on se place au niveau mondial avec toutes les espèces d'*Acipenseridae* potentiellement présentes. En effet si certains critères morphométriques comme les barbillons frangés ou non, la lèvre inférieure fendue ou non ne prêtent pas à confusion, ce n'est pas le cas du nombre d'écussons pourtant souvent cités comme caractère distinctif.

Bien que la méthode la plus utilisée soit celle de Lebedev, la façon de compter les écussons, notamment sur les rangées latérales peut varier selon les auteurs. Seules certaines espèces (*A. sinensis*, *A. ruthenus* et *A. nudiventris*) sont assez bien séparées des autres par le nombre d'écussons latéraux, les nombres d'écussons dorsaux et ventraux ne permettent pas une bonne différenciation des espèces, elles ont toutes entre 5 et 19 écussons ventraux et entre 5 et 26 écussons dorsaux (figure 2). Le nombre de branchiospines s'il permet de distinguer certaines espèces américaines (Vladykov et Beaulieu, 1951) ne semble pas non plus être un critère suffisant pour l'ensemble de la famille (figure 3).

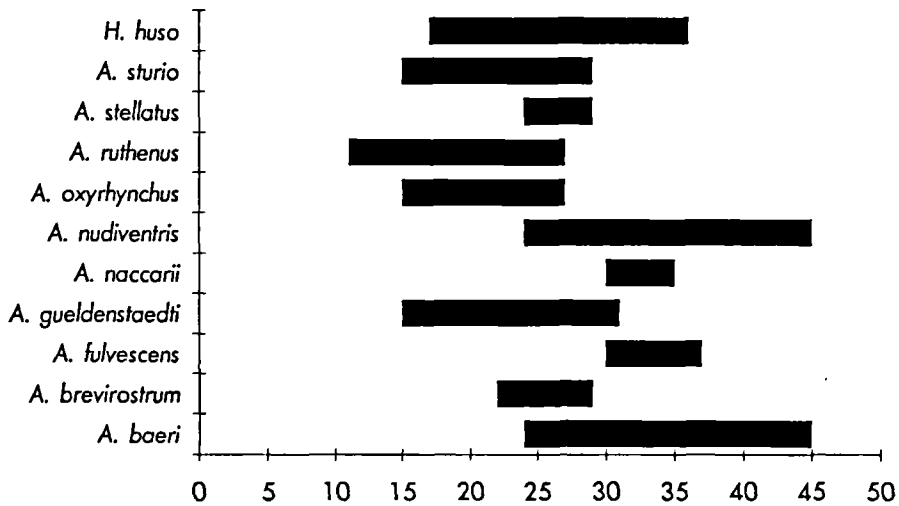


Figure 3 - Nombre de branchiospines sur le premier arc branchial.

L'introduction volontaire ou involontaire (échappement à partir d'élevage), de souches pures ou d'hybrides allochtones, perturbe les aires de répartition naturelles et rend les identifications d'espèces encore plus complexes. C'est pourquoi il serait nécessaire et relativement urgent de se doter de nouveaux outils plus performants, reposant sur des données précises et actuelles.

A l'intérieur de la famille des Acipenseridae il semble nécessaire d'utiliser successivement plusieurs critères pour arriver à identifier toutes les espèces. Pour cela, il serait très intéressant de pouvoir disposer d'un petit système expert utilisant les trois types de critères précédemment cités (écologiques ou géographiques, morphométriques, divers), auxquels il conviendrait d'adjoindre éventuellement des critères biochimiques (composition de la chair) ou génétiques (caryotype...). Ce système pourrait être utilisé pour les animaux vivants et les produits commerciaux chair et "caviar".

On peut donc souhaiter qu'il y ait un effort de réalisé afin que les données nécessaires à la mise sur pied de cet outil et qui pour une partie existent probablement déja soient disponibles.

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PRODUCTION D'ESTURGEON ET DE CAVIAR, ETAT ACTUEL ET PERSPECTIVES

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La pêche d'esturgeon conduit à deux types de produits commerciaux, la chair et le caviar. C'est une activité ancienne. Plus récemment se développe une nouvelle activité qui commence juste parfois à être mesurable, c'est la production d'esturgeon en pisciculture dans le but de commercialiser la chair. Afin de se faire une idée du devenir possible de ces productions on va d'abord analyser leur évolution récente. On a tout d'abord comparé les productions des principaux pays depuis la fin du siècle dernier ou le début du 20ième aux productions actuelles (Tableau 1).

Table 1- Evolution des quantités d'esturgeon produites par les principaux pays (tonnes) entre la fin du XIXième ou le début du XXième siècle et la période présente

Pays	Captures par pêche			Production par élevage		
	Années	Quantités	Source	Années	Quantités	Sources
URSS						
Caspienne	1900					
	/1910	30000	(1)			
	1987	19674	(2)			
Azov	1987	1317	(2)	1988	~400	(3)
USA						
Atlantique	1890	3300	(4)			
	1900	300	(4)			
Pacifique	1892	2500	(5)			
total	1987	519	(2)	1988	~500	(6)
Roumanie	1920	450	(7)			
	1987	40	(2)			
Canada	1890	200	(4)			
	1987	45	(2)			
Iran	1987	1700F	(2)			
Italie				1987	~150	
Total Monde	1987		23332F			

(1) Shubina (1975)
(2) FAO Annuaires statistiques des pêches
(3) Ivanov (communication durant le symposium),
(4) Smith (1985)
(5) Galbreath (1985)
(6) Doroshov (communication durant le symposium)
(7) Bacalbaça-Dobrovici (1991)
F "Estimation de la FAO d'après les sources d'information

On constate que les captures ont diminué pour tous les pays, que deux d'entre eux dominent largement, ce sont l'URSS avec 90% des captures mondiales et loin derrière l'Iran représentant 7% de ces mêmes captures. L'essentiel des captures de ces

deux pays est réalisé en Mer Caspienne (91,6% du total mondial). Les données publiées par la FAO représentent les captures nominales et doivent inclure pour chaque pays l'ensemble des productions qu'elles proviennent de pêche ou d'élevage. Cependant on a tout lieu de penser que sauf peut-être pour l'URSS, les productions dues à l'élevage ne sont pas déclarées. L'une des raisons pourrait être que ce sont des activités très récentes.

Pour aller encore plus loin dans l'analyse on a retracé l'évolution des productions d'esturgeon et de caviar de 1965 à 1987 pour l'URSS (Tableau 2) et l'Iran (Tableau 3).

Tableau 2- Evolution des captures d'esturgeon par l'URSS (en tonnes)

Année	Zone FAO 37[1]	Zone FAO 07[2]	
		Sibérie[3]	Total
1965	600	760	16200
1966	800	630	14300
1967	700	600	16200
1968	600	400	17500
1969	500	340	17400
1970	600	260	16600
1971	600	250	18500
1972	800	310	19600
1973	1000	230	19700
1974	1000	240	20700
1975	1080	270	23750
1976	1350	-	26820
1977	1150	-	27920
1978	740	-	24150
1979	1130	-	25190
1980	960	-	25740
1981	850	-	25600
1982	1070	-	24630
1983	1330	-	24240
1984	1430	-	22810
1985	1290	-	21480
1986	1370	-	20440
1987	1320	-	19670

[1] Mer Noire et Mer d'Azov
[2] Mer Caspienne et Sibérie
[3] Votinov et Kasyanov, 1974 et 1979

En ce qui concerne l'URSS on a distingué 3 zones, la première correspond aux captures effectuées par ce pays dans la zone 37 de la FAO et donc en Mer Noire et Mer d'Azov cette dernière étant prédominante (Kozhin, 1964). La seconde à l'ensemble des fleuves sibériens, principalement l'Ob et l'Irtysh, l'Ienisseï et enfin la Léna. L'aménagement de ces fleuves et l'industrialisation ont conduit à une diminution importante des captures et bien que nous manquions de chiffres récents les tonnages débarqués dans cette zone n'ont très vraisemblablement pas dû augmenter. Enfin la troisième zone correspond à la Mer Caspienne qui avec la Sibérie représente la zone 07 de la FAO. Les quantités pêchées en Caspienne s'obtiennent dans le tableau 2 en

soustrayant du total de la zone 07 les chiffres relatifs à la Sibérie. En première approximation on peut considérer que la colonne total du tableau 2 représente les quantités débarquées en Caspienne. Ainsi après les 30000 tonnes du début du siècle seulement 15000 à 16000 tonnes sont capturées au début des années 60, puis jusqu'à 25000 à 28000 à la fin des années 70. L'évolution était alors encourageante et pouvait être considérée comme le fruit de tous les efforts consentis par l'URSS, réglementation, exploitation raisonnée, repeuplement massif (Williot, 1984). Par contre depuis le début des années 80 il semble bien que l'on assiste à une nouvelle diminution des prises.

Quant à la production de caviar soviétique on ne peut en avoir qu'une estimation globale. En effet les chiffres contenus dans les annuaires de la FAO sont trop invraisemblables pour être retenus (en 1987, 21960 tonnes de caviar pour 20991 tonnes d'esturgeon !). Gödecken (1986) rapporte une estimation de 500 tonnes pour la production au début des années 60 (saison de pêche 61/62) alors que les quantités d'esturgeon captureés peuvent être estimées à un peu moins de 20000 tonnes; Cela conduirait à un rapport caviar/esturgeon de 2,5%. A la fin des années 70, la production aurait été voisine de 1750 tonnes selon Marti(1979) dans Doroshov and Binkowski (1985), le même rapport serait alors égal à 7%(captures d'esturgeon proches de 25000 tonnes). Si on applique le même rapport (7%) aux dernières productions connues, cela conduit à estimer la production de caviar soviétique à 1400 tonnes en 1987.

Les captures iraniennes sont réalisées en Mer Caspienne. Après être restées en deçà de 1000 tonnes jusqu'en 1956/1957 (Rostami, 1960), la moyenne annuelle a été ensuite égale à 1450 tonnes jusqu'en 1962 (Farid-Pak, 1963). Elles ont été supérieures à 2000 tonnes de 1965 à 1972 (Tableau 3) puis semblent s'être stabilisées entre 1500 et 1700 tonnes. Durant la même période la production de caviar a augmenté de 50%(Tableau 3). Les rapports caviar /esturgeon varient entre 11,8 et 17,9%. Ce rapport avait 12,7% pour valeur moyenne de 1957 à 1962 selon les données de base publiées par Gödecken (1986).

En retenant l'estimation précédente pour l'URSS (1400), le chiffre FAO pour l'Iran (300) et les estimations de Bourguignon (1989) pour les autres pays (50 tonnes environ), on arriverait à une production mondiale de caviar d'environ 1750 tonnes en1987.

Le marché mondial, c'est à dire celui donnant lieu à des flux internationaux matière et financier, est nettement inférieur en tonnage car les deux principaux producteurs sont aussi les principaux consommateurs même si d'après Gödecken (1986) dans le tableau 3, l'autoconsommation iranienne diminuerait au profit des exporations. Selon deux estimations anonymes rapportées par Bourguignon (1989), les exportations réunies de l'URSS et de l'Iran en 1987 seraient de l'ordre de 500 tonnes, chiffre auquel devrait s'ajouter les 50 tonnes correspondant à la production estimée des autres pays, soit donc 550 tonnes.

Quelle confiance peut-on accorder à ces estimations?

Les stocks exploités (*Acipenser stellatus, Acipenser gueldenstaedti* et *Huso huso*) par les soviétiques et les iraniens en Caspienne sont interdépendants (Rostami, 1960; Kozhin et al.,1964) à l'exception peut-être de *Acipenser gueldenstaedti persicus.* Dès lors il paraît surprenant que l'on arrive à des estimations aussi différentes du rapport caviar/esturgeon qui sont au moins deux fois élevées pour l'Iran que pour l'URSS. D'après les données de Rostami (1961) le même type de calcul appliqué uniquement aux femelles des 3 espèces et sur l'ensemble de la période 1927/1928 à 1956/1957 conduit à une valeur de 13% incohérente avec les calculs du tableau 3 puisque ceux-ci doivent normalement tenir compte des mâles dans les captures. Bien sûr cette comparaison est discutable car elle porte sur des périodes différentes, cependant l'écart est tel qu'il justifie d'en faire état.

Selon Bourguignon (1989) les quatre principaux pays importateurs de caviar (France, RFA, Suisse et USA) ne totalisaient que 220 à 230 tonnes en 1987-88, chiffre ne correspondant même pas à la moitié du marché mondial tel que nous l'avons défini. De nombreuses personnes s'accordent pour penser que ce marché est l'objet d'une fraude importante, est-elle suffisante pour expliquer l'enorme décalage entre le chiffre précédent et notre estimation du marché mondial à 550 tonnes? Ne faudrait-il pas remettre en cause cette estimation?

Tableau 3- Captures d'esturgeon, Production et exportation de caviar par l'Iran (en tonnes).

Année	Captures d'esturgeon (C)[1]	Production de caviar (P)[2]	(P/C)%	Exportation de caviar [3]	
				Quantité (Q)	(Q/P)%
1965	2100	-	-	-	-
1966	2200	-	-	-	-
1967	2400	-	-	-	-
1968	2300	-	-	-	-
1969	2500	-	-	-	-
1970	3000	-	-	-	-
1971	2400	-	-	-	-
1972	2200	-	-	-	-
1973	1500	-	-	-	-
1974	1500F	-	-	-	-
1975	1500F	-	-	-	-
1976	1500F	25	-	-	-
1977	1500F	183	-	-	-
1978	1801	212	11,8	82	38,7
1979	1578	211	13,4	79	37,4
1980	1429	233	16,3	70	30
1981	1496	234	15,6	96	41
1982	1450F	204	-	101	49,5
1983	1500F	225	-	113	50,2
1984	1557	238	15,3	130	54,6
1985	1650	267	16,2	-	-
1986	1690	303	17,9	-	-
1987	1700F	300	-	-	-

[1] FAO Annuaires statistiques des pêches
[2] FAO Annuaires statistiques des pêches vol.59 pour 1976 et 1977, vol.65 depuis 1978
[3] Gödecken (1986)
F "Estimation de la FAO d'après les sources d'informations disponibles"

Quelles sont les tendances actuelles?

Les prévisions de production par pêche ne peuvent être que pessimistes. nous avons déjà fait remarquer que les débarquements soviétiques diminuaient depuis quelques années alors qu'aucune information ne laisse supposer que les soviétiques aient profondément modifié leur gestion de ces stocks dans un passé récent. La seule hypothèse à cette chute des captures est une dégradation des milieux fréquentés par les esturgeons, quelques articles récents dont celui de Berg (1990), confirment cette explication. Si la détérioration de l'environnement est effectivement la cause de ce déclin, il faut malheureusement s'attendre à ce que la tendance actuelle se poursuive

pendant plusieurs années avant qu'un ensemble de mesures puisse permettre de modifier cette évolution.

On doit être également inquiet pour les captures iraniennes sur la base des chiffres publiés par la FAO (Tableau 3). On imagine mal en effet que l'augmentatiopn de la productivité de caviar soit sans conséquence à l'avenir.

Il paraît donc très vraisemblable, uniquement sur la base de l'analyse quantitative ci-dessus, que la production d'esturgeon par pêche et donc celle de caviar iront en diminuant, aucun pays ne disposant des stocks suffisants pour pallier cette baisse de productivité du bassin caspien. Ainsi, indépendamment des aspects qualitatifs, il est probable que des tensions apparaissent sur ces marchés.

Les perspectives semblent par contre favorables pour la production d'esturgeon d'élevage. Inexistante il y a seulement quelques années, elle est en train de se développer et de nombreux projets sont soit en cours de réalisation soit à l'étude. Probablement la taille des produits issus d'élevage sera pour un moment inférieure à celle des produits de la pêche. Il est probable que l'on assiste à brève échéance à des tentatives d'élevage pour produire du caviar, ceci surtout si les tensions évoquées plus haut se confirment.

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