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The effects of storage temperature, storage duration, hydro-cooling, and micro-perforated wrap on shelf life of broccoli (*Brassica oleracea* L., Italica Group)

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Abstract

This work was initiated to determine the effects of cold storage duration and storage temperature on the shelf life of wrapped broccoli when subsequently placed into elevated temperatures to simulate retail shelf conditions. In addition, the effect of hydro-cooling prior to wrapping was evaluated. These tests were designed to test the potential for micro-perforated wrap in extending shelf life of broccoli under varied postharvest handling conditions. In the first experiment, broccoli (*Brassica oleracea* L., Italica Group; cv. Mariner) was harvested and the crop divided into 4 treatments; (1) 'hydro-cooled + no wrap', (2) 'hydro-cooled + wrap', (3) 'non-cooled + no wrap', and (4) 'non-cooled + wrap'. Samples of each of the 4 treatments were removed from storage after 3 days at 1°C and placed into 13°C to simulate shelf conditions. Visual quality, weight loss and respiration were monitored over 5 days at 13°C. This shelf evaluation was repeated with broccoli samples that had been stored for 10 and 17 days at 1°C. In a second experiment, broccoli was harvested and hydro-cooled. Half the crop was wrapped with micro-perforated film and the other half left unwrapped. From these two treatments, half of the heads were stored at 1°C and the other half at 5°C. After 10 days, all the heads were placed at 13°C for 5 days. Measurements at 13°C were taken as in the first experiment. In the first experiment, for broccoli which was stored for only 3 days at 1°C, the use of either hydro-cooling or wrap gave good firmness retention at 13°C shelf conditions. However, for broccoli which had been stored for 10 or 17 days, both hydro-cooling and wrapping were required to achieve the best firmness retention. Yellowing during the 5 days at 13°C was found to be the greatest for broccoli that had been stored at 1°C for only 3 days. Broccoli stored for 10 and 17 days developed much less yellowing during 5 days holding at 13°C. In the second experiment, storage at 1°C resulted in much better shelf life than storage at 5°C. Application of wrap gave best firmness retention and least water loss, independently of storage temperature. Only colour retention was affected by storage temperature, with 5°C storage resulting in a significant increase in yellowing. Neither hydro-cooling nor packaging had an effect on yellowing. Therefore, broccoli firmness and colour retention can be easily maintained for 5 days at elevated 'shelf' temperatures with the integration of a system which includes hydro-cooling, application of micro-perforated film wrap and sufficient cold storage duration at 1°C.

Keywords: *Brassica oleracea* L. (Italica Group); Shelf life; Cooling; Storage; Packaging

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1. Introduction

The deterioration of broccoli after harvest has been extensively studied. Much of the work on deterioration of broccoli has been performed at elevated temperatures (King and Morris, 1994; Tian et al., 1994) without consideration for prior cold storage or handling of the product. Brennan and Shewfelt (1989) conducted experiments on cooling delays and their effect on broccoli quality and shelf life. Their work emphasized the importance of rapid temperature pull-down after harvest in maintaining broccoli quality. However their work did not cover packaging, storage temperature or storage duration as factors to shelf life performance. Several workers have shown that modified atmosphere (MA) packaging can help to maintain broccoli quality under non-ideal handling temperatures (Miyazaki, 1985; Ballantyne et al., 1988; Barth et al., 1993); however, if oxygen levels drop too low, off-odour problems can occur (Miyazaki, 1985; Ballantyne et al., 1988).

In our laboratory, we have found that the selection of precooling method with application of micro-perforated film wrapping can significantly extend the shelf life of broccoli (Gillies and Toivonen, 1995). The advantage of this approach as compared to modified atmosphere (MA) packaging is that the atmosphere within the package cannot become overmodified when temperature abuse occurs. As a result, potential for development of off-odours and off-flavours is minimized. However, micro-perforated films must be tested under various storage temperatures and durations to evaluate the effect of these factors on shelf life performance at elevated 'shelf' temperatures.

The main goal of this work was to determine the effect of both duration and temperature of storage on performance of micro-perforated film-wrapped broccoli when placed into 13°C 'shelf' conditions. The effect of hydro-cooling in relation to storage duration was also evaluated.

2. Materials and methods

The data presented is the second repeat of a 2-year experiment, the results were similar in both years. Broccoli (cv. Mariner) was grown at the Pacific Agriculture Research Station at Agassiz, BC, Canada in 1993. At maturity, the crop was harvested and transported to cooling and handling facilities within 2 h of harvest. All heads were trimmed to a 150-mm length, only blemish- and defect-free heads were used.

2.1. Experiment 1: Effect of hydro-cooling and storage duration

Half of 192 harvested and trimmed heads were hydro-cooled and the other half left non-cooled. Hydro-cooling was accomplished using a custom-made pilot scale recirculating shower-type hydrocooler. The core temperature of the broccoli came down to between 4–5°C with hydro-cooling. One half of the heads, from each of these two treatments, was wrapped with SM60 micro-perforated wrap (polyolefin with 0.5 mm diameter perforations, 0.03 mm thick and 0.2% total perforated area; Cryovac Division, W.R. Grace and Co., Duncan, SC) and the other half was left unwrapped. All of the heads were then placed into a storage room set at 1 ± 0.5°C, 95 ± 2% RH.

After 3 days of storage, 16 heads from each treatment were removed from 1°C and placed into a storage room set at 13 ± 1°C, 95 ± 2% RH. One hour after removal from 1°C, three sets comprised of three heads were selected from each treatment and respiration measurements were made. These same heads were used for further measurements over 3 subsequent days at 13°C. The remaining 7 heads of each treatment were weighed to determine weight loss and also assessed for firmness and colour. These same heads were monitored over 5 days at 13°C. The same protocol for measurement and assessment was applied to heads removed from 1°C after 10 and 17 days of storage.

2.2. Experiment 2: Effect of storage temperature

Forty trimmed and hydro-cooled heads were selected. Half were wrapped with micro-perforated film and the other half left unwrapped. From each of these two treatments (wrapped and unwrapped), half were placed into 1 ± 0.5°C, 95 ± 2% RH storage and the other half into 5 ± 0.7°C, 95 ± 2% RH storage for a period of 10 days. The heads were removed from storage and placed into 13°C, 95%

RH conditions. Respiration, weight loss and quality were monitored over 5 days at that temperature as described in Experiment 1.

2.3. Measurements

Respiration was measured as reported previously (Toivonen, 1992). The heads were placed into 12-l polyethylene pails with tight-fitting lids. Heads which were wrapped remained so during the respiration measurements, since the SM60 film does not restrict gas exchange between the broccoli and the surrounding atmosphere (unpublished data). The pails were flushed with CO_2-free air before sealing. Initial samples for CO_2 analysis were taken at time zero and a subsequent sample was taken approximately 0.5 h later (duration times were recorded). Each sample was measured twice. The CO_2 was determined on a Shimdazu GC-9A gas chromatograph (TekScience, Oakville, Ont.), fitted with a 8×0.0032 m Porapak Q (Supelco, Oakville, Ont., 80/100 mesh) column. The oven temperature was 75°C and the helium carrier flow set at 50 ml min^{-1}. A methanizer converted the CO_2 to methane (at 350°C) which was then measured with an FID detector (at 250°C).

On the day of removal from 1°C storage, the colour and firmness were evaluated, using a 5-point scale as reported by Gillies and Toivonen (1995). Heads were removed from packages for only a few minutes to allow quality evaluation. On the colour scale, 1 represented heads of which all buds had turned yellow and 5 represented heads of which all buds were of fresh, dark green appearance. On the firmness scale, 1 represented heads which were completely limp and 5 represented heads which were very firm, with no signs of wilting. Evaluations were repeated for the same heads on each of 4 subsequent days at 13°C.

Weight of individual heads was taken on the day of harvest, after hydro-cooling and wrapping had been completed. This was considered to be the 'original weight' when computing weight loss. Weights were then taken on the day of removal from 1°C and on a daily basis when the broccoli was put into 13°C conditions. Weight losses are expressed as percentage loss of original weight on the day of harvest (i.e., they are cumulative).

2.4. Analysis

The experiments were of randomized complete block design. In the first experiment, there were 3 factors, precooling, wrap, and storage duration. Precooling consisted of two levels, storage duration 3 levels and packaging 2 levels. In the second experiment, there were two factors, wrap and storage temperature. Wrap consisted of two levels and storage temperature two levels. Samples were repeatedly sampled for respiration, weight loss, firmness and colour and so the data were analysed using GLM procedure of SAS (SAS Institute, Cary, NC), with a repeated measures option. For weight loss, firmness and colour, there were 7 replicates measured over 5 days at 13°C, and for the respiration, 3 replicates were measured over 5 days at 13°C. Correlation analysis was also performed to evaluate relationships of respiration and weight loss against changes in colour and firmness of the broccoli.

3. Results

3.1. Experiment 1

Changes in firmness, weight losses and colour were significant (Table 1); however, there were significant interaction terms. Therefore data are presented as simple effects in Figs. 1–3. Changes in respiration were only significant over storage duration and so the main effects are presented in Table 2.

Cumulative weight losses at 13°C for wrapped broccoli, whether hydro-cooled or not, were similar after 3, 10 and 17 days of 1°C (Fig. 1). In contrast, cumulative weight loss for the non-wrapped broccoli increased substantially with storage duration. By the third week, weight loss in unwrapped broccoli, hydro-cooled or not, exceeded 10% and the broccoli was rated as being limp (Fig. 2). The rate of water loss at 13°C was similar for all three test weeks as indicated by the similar slopes of water loss (data not shown). Changes in firmness (Fig. 2) parallelled water loss (Fig. 1). The changes in firmness over the 3 weeks were found to be highly correlated with water loss ($r = -0.91$, $P \leq 0.01$).

Hydro-cooling improved the retention of firmness at 13°C for the unwrapped broccoli which had been stored for 3 days at 1°C (Fig. 2). In contrast, hydro-

Table 1
Mean squares of the quadratic comparisons for changes in firmness, colour, weight loss and respiration in 'Mariner' broccoli stored at 1°C for 3, 10 and 17 days prior to placement at 13°C for 5 days

Source	df	Mean square of quadratic comparisons			
		Firmness	Weight loss	Colour	Respiration
Precooling (Pc)	1	2.86[c]	0.07[a]	2.86[c]	142[a]
Wrap (W)	1	1.65[c]	0.005[a]	1.65[c]	39[a]
Storage (S)	2	0.7[c]	0.57[c]	0.75[c]	634[c]
Pc × W	1	0.17[a]	0.15[c]	0.17[a]	114[a]
Pc × S	2	0.04[a]	0.02[a]	0.04[a]	100[a]
W × S	2	0.21[a]	0.06[b]	0.21[a]	68[a]
Pc × W × S	2	0.60[b]	0.16[c]	0.60[b]	10[a]
Error	72 (24)[d]	0.13	0.02	0.13	72

[a] Not significant.
[b] Significant at the 95% level.
[c] Significant at the 99% level.
[d] Degrees of freedom for respiration.

Table 2
Changes in respiration measured at 13°C, 1 h after removal from storage at 1°C, >95% RH

Time in 1°C storage (days)	Respiration rate (mg CO_2 kg^{-1} h^{-1})
3	86.6
10	74.1
17	68.7
$LSD_{0.05}$	14.3

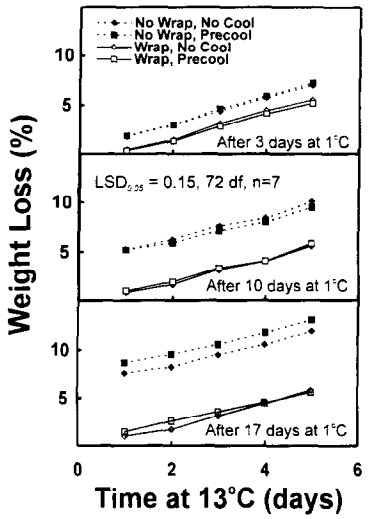

Fig. 1. The effect of storage duration at 1°C, hydro-cooling and packaging on the subsequent percentage weight loss of 'Mariner' broccoli when placed at 13°C for 5 days.

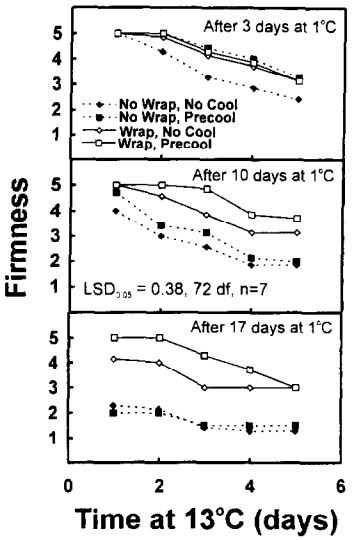

Fig. 2. The effect of storage duration at 1°C, hydro-cooling and packaging on the subsequent changes in firmness of 'Mariner' broccoli when placed at 13°C for 5 days. Firmness rating from 1 to 5, 1 = totally limp head, and 5 = very firm head.

cooling provided a benefit in firmness retention only for the wrapped broccoli when it had been stored for 10 or 17 days at 1°C. Of the four treatments, only the broccoli which was both hydro-cooled and wrapped, maintained a similar firmness retention at 13°C after 3, 10 and 17 days of storage at 1°C. In the first week, either hydro-cooling and/or wrapping provided good

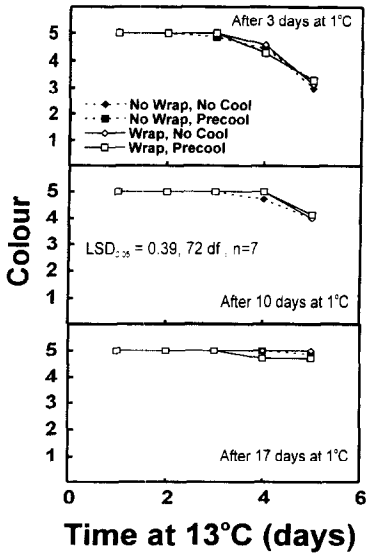

Fig. 3. The effect of storage duration at 1°C, hydro-cooling and packaging on the subsequent changes in colour scores of 'Mariner' broccoli when placed at 13°C for 5 days. Colour score rating from 1 to 5, 1 = fully yellow head, and 5 = dark green head.

firmness retention. By the third test week, only broccoli that had been both hydro-cooled and wrapped maintained superior firmness. Firmness differences were found between hydro-cooled versus non-cooled broccoli despite the fact that no significant differences in weight loss were found. This is due to an approximately 5% water uptake by broccoli during hydro-cooling (Gillies and Toivonen, 1995). However, uptake was not reflected in this work since the 'original weight' was taken after hydro-cooling. A 5% water uptake (by weight) gives the hydro-cooled broccoli a specific advantage over non-cooled broccoli in terms of maintaining firmness, at least for the first 10 days of 1°C storage.

Neither hydro-cooling or wrapping had an effect on the colour changes in the broccoli (Fig. 3). Yellowing became obvious by the 4th day at 13°C for broccoli that had been stored for only 3 days at 1°C. Broccoli stored for 10 days started to show significant colour change on the fifth day at 13°C. Broccoli stored for 17 days did not show significant colour changes, even after 5 days at 13°C. Colour changes were not correlated with weight losses (data not shown). While there were no significant changes in respiration during shelf-life tests, respiration declined over the 17 days of storage at 1°C (Table 2). The losses in colour (i.e., development of yellowing) were highly correlated with respiration rate on the day of removal from storage ($r = -0.85$, $P \leq 0.01$).

3.2. Experiment 2

No interaction between wrap treatment and storage treatment was found and so data are presented as main effects in Table 3. Wrap treatment was found to only have significant effect on respiration rate, firmness and weight loss of broccoli. Unwrapped broccoli showed a transient upsurge in respiration rate on day 2 at 13°C. A similar respiratory upsurge has been reported during the course of senescence of oat leaves (Tetley and Thimann, 1974). In contrast, wrapped broccoli showed a steady decline in respiration with time at 13°C with no such upsurge. Firmness declined much more rapidly for unwrapped broccoli as compared against wrapped broccoli. The decline in firmness was highly correlated with weight loss ($r = -0.85$, $P \leq 0.01$). The respiration rates were not highly correlated with weight loss. As in Experiment 1, visually perceptible colour change of broccoli was not significantly affected by wrap treatment.

Storage temperature had a significant effect on colour change of broccoli and respiration at 13°C (Table 3). Broccoli stored at 5°C for 10 days became fully yellow (rating of 1) by the 4th day at 13°C; whereas broccoli stored at 1°C for 10 days remained fully dark green in colour. Colour change was correlated with respiration ($r = 0.65$, $P \leq 0.01$).

4. Discussion

Storage duration had a significant effect on shelf life, manifested through a reduction in yellowing when placed into 13°C conditions. The application of hydro-cooling and/or micro-perforated packaging improved firmness retention, but had no effect on yellowing for broccoli that was placed into 13°C conditions. As a result the combination of extended storage with wrap at 1°C resulted in the best overall quality after a subsequent 5 days of holding at 13°C. The results are similar to those achieved with MA packaging (Ballantyne et al., 1988; Barth et al., 1993). This micro-perforated packaging does

Table 3
Effect of wrap and storage temperature on hydro-cooled broccoli quality, respiration and weight loss at 13°C, 95% RH

	Respiration (mg CO_2 kg^{-1} h^{-1})		Firmness[a]		Colour[b]		Weight loss (% of original weight)	
Wrap treatment:	Wrap	No Wrap	Wrap	No Wrap	Wrap	No Wrap	Wrap	No Wrap
Day 0	73.7	72.3	4.9	4.9	4.2	4.1	1.1	4.2
Day 1	71.4	69.0	4.7	4.2	3.9	3.6	2.2	5.2
Day 2	69.3	83.0	4.4	3.6	3.5	3.1	3.6	6.5
Day 3	61.8	68.6	3.9	2.6	3.1	3.0	4.6	7.5
Day 4	–	–	3.3	2.5	2.6	2.6	6.0	8.8
Significance	c		c		NS		c	
Storage temperature:	1°C	5°C	1°C	5°C	1°C	5°C	1°C	5°C
Day 0	78.0	68.0	4.9	4.9	4.9	3.5	3.0	2.2
Day 1	74.9	65.5	4.2	4.7	5.0	2.6	3.9	3.5
Day 2	80.8	71.6	4.0	4.0	5.0	1.6	5.4	4.7
Day 3	70.8	59.8	3.0	3.4	5.0	1.1	6.2	5.9
Day 4	–	–	2.9	3.0	4.1	1.0	7.8	7.0
Significance	c		NS		d		NS	
LSD	9.6		0.4		0.3		0.8	

[a] 1–5 scale: 1 = totally limp, and 5 = very firm.
[b] 1–5 scale: 1 = fully yellow, and 5 = dark green.

not permit atmospheric over-modification which occurs with MA packaging at elevated temperatures (Ballantyne et al., 1988). This suggests that micro-perforated wrap can be an alternative to MA wrap, provided that the product is stored at 1°C for at least 10 days.

Storage for 10 days at 5°C resulted in much lower shelf life than found for broccoli stored at 1°C, with the differences in quality being associated with degree of yellowing during 5 days at 13°C. Firmness retention and weight losses appeared to be independent of storage temperature. Micro-perforated wrap provided the only significant effect on firmness retention.

The results of these two experiments show that quality retention in broccoli is a consequence of what appears to be two independent factors. The first factor is weight loss which is strongly associated with losses in firmness in both experiments. The second factor is colour retention, which appears to be both positively (Experiment 2) and negatively (Experiment 1) correlated with changes in respiration.

Increased duration of 1°C storage resulted in a decline in respiration which was associated with a reduction in yellowing at simulated shelf conditions. The effect of storage 1°C duration on reduction of respiration reflects a progressive inhibition of metabolic activity of broccoli at low temperatures. An overall reduction of metabolic activity could explain the reduction of yellowing seen subsequently at 13°C. In contrast, storage at 5°C resulted in reduced respiration rates, which was correlated to increase in yellowing at simulated shelf conditions. The decline in respiration at 5°C is likely a consequence of cellular deterioration which then led to enhanced senescence and yellowing of the broccoli at shelf temperatures. These results show that while two different handling conditions can result in similar reductions in respiratory activity of broccoli, the physiological condition of the tissue, with regard to senescence, can be quite different. This is consistent with the views of Romani (1987), who suggested that mitochondrial activity levels during senescence did not determine changes, rather they were a consequence of cellular changes (i.e., a reflection of intracellular conditions).

Yellowing of leafy and green vegetables has been attributed to peroxidase activity (Baardseth and von Elbe, 1989; Yamauchi and Watada, 1991) and lipoxygenase activity (Zhuang et al., 1995). The activities these two enzymes could be responsible for yellowing of broccoli. Low temperatures are known

to enhance phenolic content and metabolism in many plant tissues and organs (Rhodes et al., 1981) and phenolics are known to affect the activity of lipoxygenase and peroxidase (Van Sumere et al., 1975; Oszmianski and Lee, 1990). Therefore, one possible affect of low temperature storage in broccoli could be in enhancing phenolic levels of the tissue, which could then inhibit oxidative injuries leading to chlorophyll loss when the broccoli is placed at elevated 'shelf' temperatures. The extent of phenolic increases in broccoli held at low temperatures has not yet been determined and whether such levels could moderate peroxidase and/or lipoxygenase-mediated yellowing in broccoli is not known. This hypothesis is currently being investigated. Preliminary results indicate that there are some changes in apparent oxidant enzyme activities with low temperature storage of broccoli.

The results presented here show that broccoli shelf life at elevated temperatures improves after 10 or 17 days storage at 1°C. Low-temperature storage can be used as an alternative to MA packaging, in some situations. This approach requires the application of both hydro-cooling and micro-perforated wrap to ensure firmness retention while the broccoli is held for an appropriate duration at 1°C to ensure reduction of yellowing potential. Studies on the mechanisms that are responsible for the low-temperature response reported here are underway.

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