

## Effect of carbon dioxide on the internal lower oxygen limits of apple fruit

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### Abstract

The effect of elevated CO<sub>2</sub> between 0 and 8 kPa on steady-state lower O<sub>2</sub> limits based on internal atmospheres (LOL<sup>i</sup>) was estimated for postclimacteric ‘Cox’s Orange Pippin’ and ‘Braeburn’ apples at 0 and 20°C. Two types of LOL<sup>i</sup> were estimated: the anaerobic compensation point (ACP<sup>i</sup>), and the internal fermentation threshold based either on the respiratory quotient (FT<sup>i</sup><sub>RQ</sub>) or ethanol (EtOH) accumulation (FT<sup>i</sup><sub>EtOH</sub>). ACP<sup>i</sup>, for both cultivars and temperatures, remained constant at 0.5 kPa O<sub>2</sub> for ‘Cox’s Orange Pippin’ and 0.8–1.0 O<sub>2</sub> for ‘Braeburn’ apples for levels of CO<sub>2</sub> external to the fruit between 0 and 8 kPa. However, for FT<sup>i</sup><sub>RQ</sub> and FT<sup>i</sup><sub>EtOH</sub>, no consistent trend with level of CO<sub>2</sub> was evident at 20°C for either cultivar. In contrast, at 0°C FT<sup>i</sup><sub>RQ</sub> and FT<sup>i</sup><sub>EtOH</sub> were 0.2–0.8 kPa O<sub>2</sub> higher at 8 kPa CO<sub>2</sub> than at 0 kPa CO<sub>2</sub> (with the exception of FT<sup>i</sup><sub>RQ</sub> for ‘Cox’s Orange Pippin’). A small decrease in O<sub>2</sub> uptake (estimated from the difference in external and internal O<sub>2</sub> atmospheres) was observed between 2 and 8 kPa CO<sub>2</sub> at 20°C. Elevated CO<sub>2</sub> slightly lowered the respiratory quotient (RQ<sup>i</sup>, estimated from the ratio of differences between external and internal atmosphere partial pressures of CO<sub>2</sub> and O<sub>2</sub>) of ‘Cox’s Orange Pippin’ in 8 kPa CO<sub>2</sub> and ‘Braeburn’ in 2 to 8 kPa CO<sub>2</sub> at 20°C, and more markedly in 8 kPa CO<sub>2</sub> at 0°C. The RQ<sup>i</sup> of ‘Cox’s Orange Pippin’ and ‘Braeburn’ apples was slightly and markedly higher respectively at 0°C compared to 20°C. The lower RQ<sup>i</sup> of ‘Braeburn’ at 20°C compared to ‘Cox’s Orange Pippin’ apples indicated ‘Braeburn’ had a higher permeance to CO<sub>2</sub> relative to O<sub>2</sub> compared to ‘Cox’s Orange Pippin’. This study indicates the tolerance of ‘Cox’s Orange Pippin’ and ‘Braeburn’ apples to low O<sub>2</sub> levels may be affected by levels of CO<sub>2</sub>. © 1997 Elsevier Science B.V.

**Keywords:** *Malus domestica*; ‘Cox’s Orange Pippin’; ‘Braeburn’; Internal atmosphere; Anaerobic compensation point; Fermentation threshold; RQ breakpoint; Skin permeance

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## 1. Introduction

Substantial benefits in maintaining quality of harvested fresh crops can accrue from storage in low O<sub>2</sub> and/or elevated CO<sub>2</sub> atmospheres at levels within the fruit's tolerance limits, as an adjunct to low temperature storage (Kader et al., 1989). Tolerance of crops to modified atmospheres can vary markedly depending on cultivar, strain, physiological age, initial quality, temperature, rate of establishment of atmospheres and duration of exposure. In general, under severe stress, fermentative metabolism is enhanced by low external partial pressure of O<sub>2</sub> ( $p_{O_2}^e$ , Pa) and elevated external partial pressure of CO<sub>2</sub> ( $p_{CO_2}^e$ , Pa), long exposure to hypoxia and more advanced developmental stages (Ke et al., 1993). Effects of modification of individual atmosphere components interact: tolerance to elevated  $p_{CO_2}^e$  decreases as  $p_{O_2}^e$  is decreased whilst tolerance to low  $p_{O_2}^e$  decreases as  $p_{CO_2}^e$  is increased (Beaudry and Gran, 1993; Kader et al., 1989).

Lower O<sub>2</sub> limits (*LOL*s) of crops are typically reported in relation to external or package atmospheres (*LOL*<sup>e</sup>). However, those based on internal atmospheres (*LOL*<sup>i</sup>), are likely to more accurately estimate the true *LOL* as they account for variation in respiration rate ( $r_{CO_2}$ , mol kg<sup>-1</sup> s<sup>-1</sup>) and skin permeance to O<sub>2</sub> and CO<sub>2</sub> ( $P'_{O_2}$ ,  $P'_{CO_2}$ , mol s<sup>-1</sup> m<sup>-2</sup> Pa<sup>-1</sup>) of individual fruit (Yearsley et al., 1996a,b). Two types of *LOL*<sup>i</sup> have been described (Yearsley et al., 1996a). The internal anaerobic compensation point (*ACP*<sup>i</sup>) is the internal O<sub>2</sub> partial pressure ( $p_{O_2}^i$ , Pa) at which internal CO<sub>2</sub> partial pressure ( $p_{CO_2}^i$ ) from aerobic and anaerobic respiration is minimal. The internal fermentation threshold (*FT*<sup>i</sup>) is the  $p_{O_2}^i$  below which anaerobic respiration is initiated. The *FT*<sup>i</sup> can be established as the  $p_{O_2}^i$  at which the respiratory quotient based on internal atmospheres (*RQ*<sup>i</sup>) rises 10% above the asymptotic value obtained at higher  $p_{O_2}^i$  (*FT*<sup>i</sup><sub>*RQ*</sub>) or alternatively, the  $p_{O_2}^i$  below which ethanol accumulation increases with initiation of fermentation (*FT*<sup>i</sup><sub>EtOH</sub>). Optimum internal atmosphere (IA) composition at which maximum benefits arising from reduced aerobic respiration accrue without inducing fermentation, lies just above the *FT*<sup>i</sup> (Banks et al., 1993a).

The *LOL*<sup>e</sup>s of apples (Gran and Beaudry, 1993a,b), blueberries (Beaudry et al., 1992; Beaudry, 1993; Cameron et al., 1994), and raspberries (Joles et al., 1994), show strong curvilinear dependence on temperature. In contrast, the *LOL*<sup>i</sup>s of apples only increased marginally with temperature, and the effects were not always consistent (Yearsley et al., 1996b).

Direct and/or indirect suppression of aerobic respiration by low levels of O<sub>2</sub> has been widely reported, and effects on the enzymatic rate of O<sub>2</sub> uptake ( $r_{O_2}$ ) has been modelled using the Michaelis-Menten equation (Dadzie et al., 1993; Peppelenbos et al., 1993; Cameron et al., 1995). Effects of elevated CO<sub>2</sub> on O<sub>2</sub> uptake (and its effects on *LOL*<sup>e</sup>s) have often been studied but the results have been contradictory, with either no effect, an increase or a decrease in respiratory activity reported depending on the crop and the partial pressure of CO<sub>2</sub> (Kidd, 1916; Kidd and West, 1927, 1933; Thornton, 1933; Young et al., 1962; Kerbel et al., 1988; Li and Kader, 1989; Kubo et al., 1990; Cameron et al., 1995). Evidence that elevated CO<sub>2</sub> has little or no effect in reducing  $r_{CO_2}$  or  $r_{O_2}$  has been reported for bananas by Young et al. (1962), and for mushrooms by Peppelenbos et al. (1993). Joles et al. (1994) reported that  $p_{CO_2}^e < 17$  kPa did not affect  $r_{O_2}$  or the *LOL*<sup>e</sup> for raspberries, and Beaudry (1993) that  $p_{CO_2}^e > 20$  kPa resulted in only a small reduction in  $r_{O_2}$  of blueberries. Gran (1993) investigated the effect of temperature on *LOL*<sup>e</sup>s of apples using modified-atmosphere packages and found that 12–15 kPa CO<sub>2</sub> in package headspace increased  $r_{O_2}$  compared to packages where CO<sub>2</sub> had been diminished by a CO<sub>2</sub> absorber, but the effect on *LOL*<sup>e</sup>s was not discussed.

In this study we report on the effect of elevated CO<sub>2</sub> on *LOL*<sup>i</sup>s for postclimacteric 'Cox's Orange Pippin' and 'Braeburn' apples and using a controlled-atmosphere (CA) method (Yearsley et al., 1996a). If the time to steady-state between  $p_{O_2}^i$  and  $p_{O_2}^e$  is short and fruit surface area and skin permeance do not change, a relative estimate of  $r_{O_2}$  for a fruit at steady-state in CA can be determined from the difference between the external and internal partial pressures of O<sub>2</sub> ( $\Delta p_{O_2}$ , Pa). The  $r_{O_2}$  would be proportional to  $\Delta p_{O_2}$  (Dadzie et al., 1996), as follows:

$$r_{O_2} = k_f \Delta p_{O_2} \quad (1)$$

where:  $k_f$  is a fruit constant ( $\text{mol kg}^{-1} \text{s}^{-1} \text{Pa}^{-1}$ ). Thus, in this study we used  $\Delta p_{O_2}$  to give some indication of the effect of elevated  $\text{CO}_2$  on  $r_{O_2}$ .

## 2. Materials and methods

### 2.1. Fruit supply, initial measurements, treatments and storage

Freshly harvested, commercially graded, preclimacteric 'Cox's Orange Pippin' and 'Braeburn' apples (*Malus domestica* Borkh.; mean mass 0.15 kg) were obtained from a commercial orchard in Hawkes Bay, NZ. The fruit were held at 20°C overnight and fruit mass, fruit firmness (0–12 kgf Effegi press-mounted penetrometer fitted with an 11 mm head), soluble solids content (0–20% Atago refractometer) measured for 20 fruit as described by Yearsley et al. (1996b). Fruit were treated with approximately 10 Pa (100 ppm) ethylene at 20°C for 12 h ( $p_{\text{CO}_2}^e$  was maintained below 0.1 kPa using hydrated lime) in a 1.3 m<sup>3</sup> constant temperature chamber, then ventilated in air at 20°C for a further 48 h during which time, respiration and ethylene production rates increased, indicating fruit had become climacteric (data not shown). Trays of fruit were then randomly allocated to temperature and  $\text{CO}_2$  treatments and stored in cartons, at either 2°C for 'Cox's Orange Pippin' (enclosed in perforated polyethylene bags), or 0°C for 'Braeburn' (without bags) as in commercial practice.

Fruit used for each temperature/ $\text{CO}_2$  treatment were removed from cool storage as randomly allocated, and equilibrated in air at  $20 \pm 0.5^\circ\text{C}$  for 24 h before blemish-free fruit were randomly allocated to treatments, six fruit to each of ten CA treatments. Postclimacteric fruit were used for experiments to minimise variation resulting from differences in fruit respiration.

### 2.2. CA and elevated $\text{CO}_2$ treatments

Experiments were conducted in a 1.0 m<sup>3</sup> con-

trolled temperature cabinet (Spaceline, Muller-McAlpine, Auckland, NZ). Each experiment used ten CA bags (Tuflex  $\text{O}_2$  barrier film, Sealed Air, Hamilton, NZ), each containing six fruit, and each with a different level of  $\text{O}_2$  but common level of  $\text{CO}_2$ .  $\text{CO}_2$  treatments used for both cultivars were 0, 2, 4, 6, and 8 kPa  $\text{CO}_2$  at 20°C, and 0 and 8 kPa  $\text{CO}_2$  at 0°C.

Metered, humidified gas mixtures flowed through each CA bag (total flow =  $1.7 \times 10^3 \text{ mm}^3 \text{ s}^{-1}$ ), for up to 7 days, with levels of  $\text{O}_2$  in the bags determined as described by Yearsley et al. (1996a). Mean  $p_{\text{O}_2}^e$  and  $p_{\text{CO}_2}^e$  within bags were controlled to within  $\pm 0.2$  or 0.3 kPa respectively of required levels, and ethylene ( $p_{\text{C}_2\text{H}_4}^e$ ) was kept  $< 1.0$  Pa at the highest  $p_{\text{O}_2}^e$  and 20°C, and  $\leq 0.08$  Pa at 0°C.

Mean temperature for four fruit at different positions within the cabinet varied  $\leq 0.3^\circ\text{C}$ , and relative humidity within CA bags was  $\geq 90\%$ , measured as described by Yearsley et al. (1996b).

### 2.3. Gas measurement and analysis

Fruit allocated to CA treatments each had a 1000 mm<sup>3</sup> glass surface chamber adhered at an equatorial position as described by Yearsley et al. (1996a). Steady-state  $p_{\text{O}_2}^i$  and  $p_{\text{CO}_2}^i$  were estimated as the equilibrated  $p_{\text{O}_2}^i$  and  $p_{\text{CO}_2}^i$  in the surface chambers after 86–90 h exposure to the CA treatments. Gas samples were removed by gas tight syringe (Hamilton 100 mm<sup>3</sup>, Hamilton, NV, USA) from the headspace of each surface chamber through a septum on the CA bags above the chamber and immediately analysed. Composition of  $\text{O}_2$  and  $\text{CO}_2$  were determined using an  $\text{O}_2$  electrode in series with a miniature infra-red  $\text{CO}_2$  transducer (Yearsley et al., 1996a), and percentage values converted to  $p_{\text{O}_2}^i$  and  $p_{\text{CO}_2}^i$  by adjusting for atmospheric pressure (Yearsley et al., 1996b).

Steady-state concentrations of acetaldehyde, ethyl acetate and ethanol ( $c_{\text{Acet}}^i$ ,  $c_{\text{EtAc}}^i$ ,  $c_{\text{EtOH}}^i$ ,  $\text{mol m}^{-3}$ ) in the surface chambers were measured using flame ionisation gas chromatography (Yearsley et al., 1996a).

#### 2.4. Estimation of physiological changes as a function of storage time

As the different CO<sub>2</sub> treatments were randomised in order over time, physiological changes of fruit in storage were quantified for a separate subsample of fruit at each removal from cool storage. Fruit firmness and soluble solids content were measured on a random sample of 20 fruit, and cortical tissue porosity ( $\epsilon$ , m<sup>3</sup> m<sup>-3</sup>) and density ( $\rho$ , kg m<sup>-3</sup>) on 15 fruit using a water displacement / infiltration method (Yearsley et al., 1996a). Additionally, 15 fruit were stored in a perforated polyethylene bag at the treatment temperature for estimating  $r_{\text{CO}_2}$  and C<sub>2</sub>H<sub>4</sub> production ( $r_{\text{C}_2\text{H}_4}$ , mol kg<sup>-1</sup> s<sup>-1</sup>) approximately 68 and 120 h after CA treatments were imposed as described by Yearsley et al. (1996b). Permeance of the fruit's skin to ethane ( $P'_{\text{C}_2\text{H}_6}$ , mol s<sup>-1</sup> m<sup>-2</sup> Pa<sup>-1</sup>) was also estimated on 15 fruit equilibrated to 20°C at the beginning and end of experiments with each cultivar, as described by Yearsley et al. (1996a).

#### 2.5. Estimation of $LOL^i$

The  $LOL^i$ s ( $ACP^i$ ,  $FT^i_{RQ}$  and  $FT^i_{\text{EtOH}}$ ) used in this study were estimated from steady-state  $p^i_{\text{O}_2}$  and  $p^i_{\text{CO}_2}$  of surface chambers as described by Yearsley et al. (1996a), as was the 'bootstrap' statistical procedure used to estimate mean  $LOL^i$ s and their bias-corrected (BCa-type) 95% confidence intervals. As the bootstrap procedure did not assume data to be normally distributed, confidence intervals which were generated were not necessarily symmetrical about means.  $FT^i_{RQ}$  was estimated from  $RQ^i$  calculated as the ratio of the difference in partial pressures of CO<sub>2</sub> ( $\Delta p_{\text{CO}_2}$ , Pa) and O<sub>2</sub> ( $\Delta p_{\text{O}_2}$ , Pa) between internal and external atmospheres ( $(p^i_{\text{CO}_2} - p^e_{\text{CO}_2}) / (p^i_{\text{O}_2} - p^e_{\text{O}_2})$ ).

#### 2.6. Statistical analysis

Regression analysis of the relationships between  $LOL^i$ s of fruit from both cultivars at 20°C, and mean  $p^e_{\text{CO}_2}$  and time in storage were performed using PROC REG of the SAS system (SAS, 1990). Regressions were weighted with the inverse of the bootstrap standard error of means, either

with or without adjustment for time in storage. When adjusted for time, limited degrees of freedom prevented fitting polynomial models beyond quadratic effects. Consequently, regression models were not adjusted for time when storage time effects were not significant. Despite high  $r^2$  values, polynomial effects models were not always significant, indicating there was not enough evidence to make statistically justifiable inferences about the relationship between  $LOL^i$ s and  $p^e_{\text{CO}_2}$ . Analyses of variance of fruit maturity,  $\epsilon$ ,  $\rho$ ,  $r_{\text{CO}_2}$ ,  $r_{\text{C}_2\text{H}_4}$  and  $P'_{\text{C}_2\text{H}_6}$  data were performed using PROC GLM of the SAS system (SAS, 1990). Parameters were estimated for functions fitted to  $\Delta p_{\text{O}_2}$  and  $RQ^i$  using Fig. P software (Fig. P, 1991).

### 3. Results

#### 3.1. Physiological changes as a function of storage time

Fruit firmness of 'Cox's Orange Pippin' and 'Braeburn' apples at harvest was 74 and 75 N respectively and decreased rapidly to 46 and 59 N, respectively, after C<sub>2</sub>H<sub>4</sub> treatment and the first 12 weeks of storage. Soluble solids content of 'Cox's Orange Pippin' and 'Braeburn' was 11.4 and 11.2%, respectively at harvest, and increased slightly to 12.8 and 11.8%, respectively, during storage. Differences through storage in fruit firmness and soluble solids content were considered small and unlikely to influence  $LOL^i$ s (data not shown).

On average, 'Cox's Orange Pippin' apples had slightly higher  $\epsilon$  than 'Braeburn' apples (0.153 and 0.137 m<sup>3</sup> m<sup>-3</sup>, respectively) and lower  $\rho$  (886.7 and 900.8 kg m<sup>-3</sup>, respectively). The relationship between  $\rho$  and  $\epsilon$  for both cultivars was linear ( $\rho = -914.4\epsilon + 1026.4$ ,  $r^2 = 0.87$ ). At different storage times,  $\epsilon$  and  $\rho$  differed significantly for 'Cox's Orange Pippin' but not 'Braeburn' fruit, but the trend over time for 'Cox's Orange Pippin' fruit was not consistent (data not shown).

Mean  $P'_{\text{C}_2\text{H}_6}$  was approximately 1.7 times higher for 'Cox's Orange Pippin' than 'Braeburn' fruit (0.286 and 0.168 nmol s<sup>-1</sup> m<sup>-2</sup> Pa<sup>-1</sup> at 20°C, respectively), and values did not change

Table 1

Estimates of parameters and standard errors (SE) for Eq. (2) describing the relationship between the difference in external and internal (chamber) steady-state partial pressures of O<sub>2</sub> ( $\Delta p_{O_2}$ ) and internal partial pressure of O<sub>2</sub> for 'Cox's Orange Pippin' (COP) and 'Braeburn' apples at 20 and 0°C ( $T$ ) at various external CO<sub>2</sub> partial pressures ( $p_{CO_2}^e$ )

| $T$ (°C)   | $p_{CO_2}^e$ (kPa) | $k_1$ | SE    | $k_2$ | SE    | $r^2$ |
|------------|--------------------|-------|-------|-------|-------|-------|
| COP        |                    |       |       |       |       |       |
| 20         | 0                  | 4.11  | 0.227 | 0.55  | 0.112 | 0.748 |
| 20         | 2                  | 4.27  | 0.231 | 0.90  | 0.166 | 0.777 |
| 20         | 4                  | 3.91  | 0.204 | 0.86  | 0.177 | 0.776 |
| 20         | 6                  | 3.53  | 0.205 | 0.58  | 0.134 | 0.668 |
| 20         | 8                  | 3.58  | 0.177 | 0.45  | 0.089 | 0.712 |
| 0          | 0                  | 0.89  | 0.040 | 0.31  | 0.064 | 0.747 |
| 0          | 8                  | 1.00  | 0.053 | 0.33  | 0.077 | 0.610 |
| 'Braeburn' |                    |       |       |       |       |       |
| 20         | 0                  | 6.75  | 0.517 | 1.54  | 0.333 | 0.731 |
| 20         | 2                  | 5.47  | 0.347 | 0.65  | 0.168 | 0.512 |
| 20         | 4                  | 5.98  | 0.370 | 1.35  | 0.286 | 0.710 |
| 20         | 6                  | 6.76  | 0.525 | 2.32  | 0.502 | 0.710 |
| 20         | 8                  | 5.56  | 0.323 | 1.54  | 0.289 | 0.734 |
| 0          | 0                  | 0.98  | 0.059 | 0.34  | 0.093 | 0.614 |
| 0          | 8                  | 0.84  | 0.074 | 0.36  | 0.161 | 0.291 |

respectively, and of 'Braeburn', 0.07 and 0.93 nmol kg<sup>-1</sup> s<sup>-1</sup> respectively.

### 3.2. $LOL^i$ s as a function of $p_{CO_2}^e$

Though statistically significant polynomial fits of  $LOL^i$ s as a function of  $p_{CO_2}^e$  were found for some data, the amount of variation accounted for was difficult to explain physiologically.

#### 3.2.1. $ACP^i$ as a function of $p_{CO_2}^e$

$ACP^i$  remained essentially constant (approximately 0.52 kPa O<sub>2</sub>) for 'Cox's Orange Pippin' apples at 20°C with  $p_{CO_2}^e$  between 0 and 8 kPa, but at 0°C,  $ACP^i$  was slightly lower at 8 kPa  $p_{CO_2}^e$  than at 0 kPa  $p_{CO_2}^e$  (Fig. 1). For 'Braeburn' apples at 20°C,  $ACP^i$  varied between 0 and 8 kPa (Fig. 2; mean over all levels of  $p_{CO_2}^e$  was 0.92 kPa O<sub>2</sub>) but differences were not significant, and at 0°C was similar (0.88 kPa O<sub>2</sub>) at 0 and 8 kPa  $p_{CO_2}^e$  (Fig. 2). On average,  $ACP^i$  was 0.31 and 0.40 kPa O<sub>2</sub> higher for 'Braeburn' than 'Cox's Orange Pippin' apples at 0 and 20°C respectively (Figs. 1 and 2).

#### 3.2.2. $FT_{RQ}^i$ as a function of $p_{CO_2}^e$

$FT_{RQ}^i$  of 'Cox's Orange Pippin' fruit at 20°C did not vary significantly between 0 and 8 kPa  $p_{CO_2}^e$  (Fig. 1; overall mean = 1.21 kPa O<sub>2</sub>). 'Braeburn' apples at 20°C had a more complex relationship between  $FT_{RQ}^i$  and  $p_{CO_2}^e$  than 'Cox's Orange Pippin' (Fig. 2). There was an indication that  $FT_{RQ}^i$  decreased between 0 and 2 kPa  $p_{CO_2}^e$  then increased again with increasing  $p_{CO_2}^e$ . At 0°C, values of  $FT_{RQ}^i$  at 8 kPa  $p_{CO_2}^e$  were higher than those at 0 kPa of both cultivars (Figs. 1 and 2). When averaged over all CO<sub>2</sub> treatments,  $FT_{RQ}^i$  for 'Cox's Orange Pippin' (0.69 kPa O<sub>2</sub> higher, Fig. 1), but not 'Braeburn', apples at 20°C were higher than  $ACP^i$ .

#### 3.2.3. $FT_{EtOH}^i$ as a function of $p_{CO_2}^e$

In general, estimates of  $FT_{EtOH}^i$  were higher than  $ACP^i$  (Figs. 1 and 2), but the change in  $FT_{EtOH}^i$  with  $p_{CO_2}^e$  was quite different to that for  $ACP^i$  and  $FT_{RQ}^i$ . The relationship between  $FT_{EtOH}^i$  and  $p_{CO_2}^e$  for fruit at 20°C was complex, particularly for 'Cox's Orange Pippin'. There was evidence that for 'Braeburn'  $FT_{EtOH}^i$  was higher

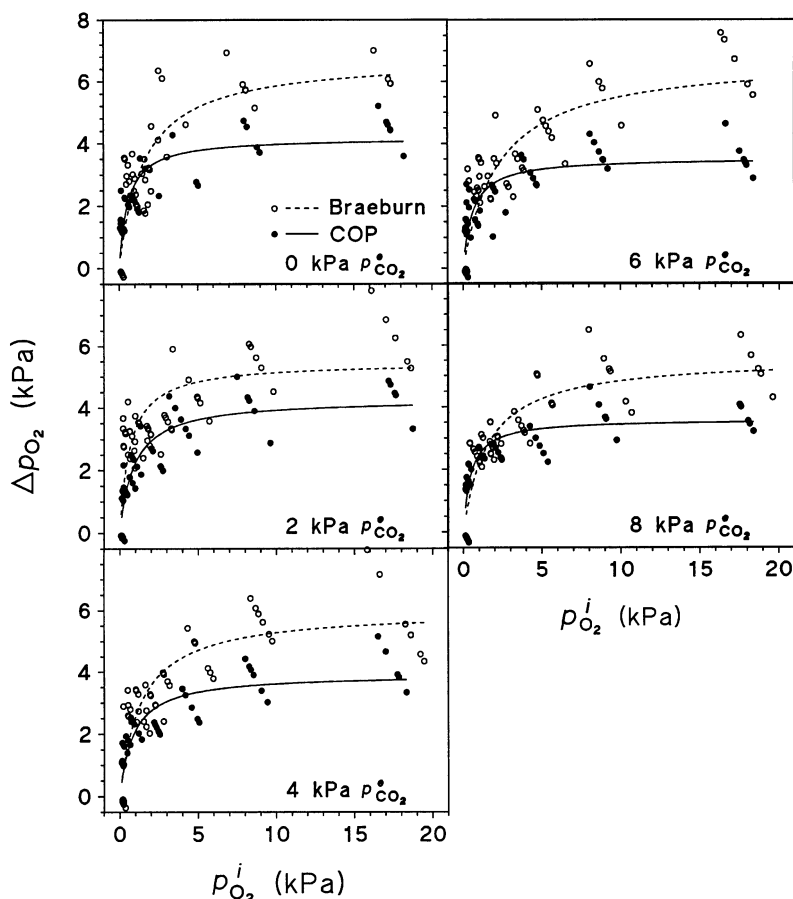


Fig. 3. Difference between external and internal (chamber) steady-state partial pressure of  $O_2$  ( $\Delta p_{O_2} = p_{O_2}^e - p_{O_2}^i$ ) as an estimate of differences in respiration rate in 'Cox's Orange Pippin' (COP) and 'Braeburn' apples at  $20^\circ\text{C}$ , stored in various external partial pressures of  $O_2$  and either 0, 2, 4, 6, or 8 kPa external partial pressures of  $CO_2$  ( $p_{CO_2}^e$ ), balance  $N_2$ . For estimates of parameters of the curves see Table 1.

for apples in 2–8 kPa  $p_{CO_2}^e$  compared to 0 kPa. Estimates of  $FT_{EtOH}^i$  at  $20^\circ\text{C}$  for 'Braeburn' apples were higher than for 'Cox's Orange Pippin'. As with  $FT_{RO}^i$  at  $0^\circ\text{C}$ ,  $FT_{EtOH}^i$  was higher for fruit at 8 than 0 kPa  $p_{CO_2}^e$ , particularly for 'Braeburn' apples. For both cultivars,  $c_{Acet}^i$  and  $c_{EtAc}^i$  accumulation in surface chambers was highly variable (data not shown).

### 3.3. $\Delta p_{O_2}$ as a function of $p_{O_2}^i$ and $p_{CO_2}^e$

The relationship between  $\Delta p_{O_2}$  and  $p_{O_2}^i$  was modelled using the Michaelis–Menten equation:

$$\Delta p_{O_2} = \frac{k_1 p_{O_2}^i}{k_2 + p_{O_2}^i} \quad (2)$$

Estimates and standard errors of  $k_1$  and  $k_2$  are presented in Table 1. There was a large range in  $\Delta p_{O_2}$  for fruit at higher values of  $p_{O_2}^i$  but the range decreased as  $p_{O_2}^i$  decreased (Figs. 3 and 4). Over all treatments, there was a small decrease in fitted values for  $\Delta p_{O_2}$  as  $p_{O_2}^i$  decreased from approximately 18–5 kPa; below approximately 5 kPa  $p_{O_2}^i$ ,  $\Delta p_{O_2}$  rapidly decreased.

There was little difference in  $\Delta p_{O_2}$  of 'Cox's Orange Pippin' apples at  $p_{CO_2}^e$  of 0 or 2 kPa at  $20^\circ\text{C}$  and  $p_{O_2}^i >$  approximately 5 kPa and (Fig. 3). However, as  $p_{CO_2}^e$  increased from 4 to 8 kPa, there was a small decrease in  $\Delta p_{O_2}$  compared to fruit in 0 kPa  $p_{CO_2}^e$ . For 'Cox's Orange Pippin' at  $0^\circ\text{C}$ , at both 0 and 8 kPa  $p_{CO_2}^e$ ,  $\Delta p_{O_2}$  values were

similar for fruit with  $p_{O_2}^i$  between approximately 20 and 5 kPa.

The  $\Delta p_{O_2}$  of 'Braeburn' apples at 20°C and  $p_{O_2}^i >$  approximately 5 kPa, were markedly higher than for 'Cox's Orange Pippin' (Fig. 3). The substantial decrease in  $\Delta p_{O_2}$  at low  $p_{O_2}^i$ , tended to commence at higher  $p_{O_2}^i$  for 'Braeburn' compared to 'Cox's Orange Pippin' apples (Figs. 3 and 4), as generally seen in higher  $k_2$  for 'Braeburn' apples (Table 2). 'Braeburn' apples at 20°C and 0 kPa  $p_{CO_2}^e$  had the highest  $\Delta p_{O_2}$ ; values for fruit in 2, 4, 6 and 8 kPa  $p_{CO_2}^e$  were similar but lower than for fruit in 0 kPa  $p_{CO_2}^e$  (Fig. 4). Similarly,  $\Delta p_{O_2}$  for 'Braeburn' apples at 0°C was higher in fruit at 0 than at 8 kPa  $p_{CO_2}^e$  but similar in 0 and 8 kPa  $p_{CO_2}^e$  for 'Cox's Orange Pippin' (Fig. 4).

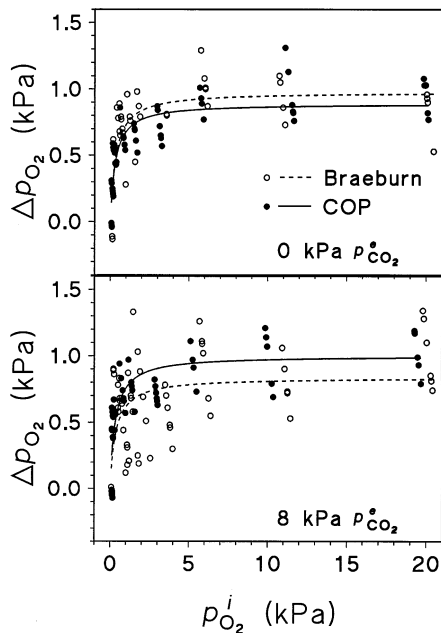


Fig. 4. Difference between external and internal (chamber) steady-state partial pressure of  $O_2$  ( $\Delta p_{O_2} = p_{O_2}^e - p_{O_2}^i$ ) as an estimate of differences in respiration rate in 'Cox's Orange Pippin' (COP) and 'Braeburn' apples at 0°C, stored in various external partial pressures of  $O_2$  and either 0 and 8 kPa external partial pressures of  $CO_2$  ( $p_{CO_2}^e$ ), balance  $N_2$ . For estimates of parameters of the curves see Table 1.

### 3.4. $RQ^i$ as a function of $p_{O_2}^i$ , temperature and $p_{CO_2}^e$

The relationship between  $RQ^i$  and  $p_{O_2}^i$  was modelled using the empirical equation:

$$RQ^i = (k_3 p_{O_2}^{i k_4}) + k_5 \quad (3)$$

Estimates of parameters and standard errors are presented in Table 2. Difficulties in obtaining significant parameters prevented them being used to calculate  $FT_{RQ}^i$ .

$RQ^i$  remained close to unity for 'Cox's Orange Pippin' apples at 0 and 20°C and 0, 2, 4, and 6 kPa  $p_{CO_2}^e$  until initiation of fermentation at low  $p_{O_2}^i$  (Figs. 5 and 6).  $RQ^i$  was lower for 'Cox's Orange Pippin' fruit at 0°C in 8 kPa  $p_{CO_2}^e$ , compared to 'Braeburn', and compared to 0 kPa  $p_{CO_2}^e$ . Asymptotic levels of  $RQ^i$  of 'Braeburn' apples at 20°C were  $< 1$  and lower than for 'Cox's Orange Pippin'. At 20°C,  $RQ^i$  was marginally lower at 2, 4, 6 and 8 kPa  $p_{CO_2}^e$  compared to 0 kPa.

## 4. Discussion

This study demonstrated that although small variation in  $ACP$ 's occurred for 'Cox's Orange Pippin' and 'Braeburn' apples at 0 and 20°C, and more marked variations in  $FT_{RQ}^i$  and  $FT_{EtOH}^i$ , the lack of clear trends suggested  $LOL$ 's were not markedly affected by levels of  $p_{CO_2}^e$  between 0 and 8 kPa  $CO_2$ . Furthermore, there was no evidence that the levels of  $p_{CO_2}^e$  used in the study caused  $CO_2$ -induced fermentation over the six day period the apples were exposed to CA treatments.

The pattern of changes in  $FT_{EtOH}^i$  at 20°C was the converse of that for  $FT_{RQ}^i$ . Given that these should both be reflections of the same physiological transition point, we can conclude that there was no major effect of  $p_{CO_2}^e$  on fermentation thresholds at 20°C. The difference in response of  $FT_{EtOH}^i$  may also have reflected the likelihood that EtOH levels in the tissue would have been continually accumulating with time. Since the chambers would inevitably have lagged behind changes occurring within the fruit, the concentrations of EtOH sampled from the chambers may not have been at steady-state. The marked increase at 8

Table 2

Estimates of parameters for plots and standard errors (SE) of the respiratory quotient ( $RQ^i$ ) as a function of internal (chamber) steady-state partial pressure of  $O_2$  using Eq. (3), for 'Cox's Orange Pippin' (COP) and 'Braeburn' apples at 20 and 0°C ( $T$ ), and at various external  $CO_2$  partial pressures ( $p_{CO_2}^e$ )

| $T$ (°C)   | $p_{CO_2}^e$ (kPa) | $k_3$ | SE    | $k_4$ | SE    | $k_5$ | SE    | $r^2$ |
|------------|--------------------|-------|-------|-------|-------|-------|-------|-------|
| COP        |                    |       |       |       |       |       |       |       |
| 20         | 0                  | 0.52  | 0.294 | -0.48 | 0.185 | 0.78  | 0.255 | 0.624 |
| 20         | 2                  | 0.18  | 0.136 | -1.02 | 0.388 | 1.03  | 0.123 | 0.628 |
| 20         | 4                  | 0.06  | 0.070 | -1.54 | 0.566 | 1.03  | 0.105 | 0.617 |
| 20         | 6                  | 0.11  | 0.075 | -1.13 | 0.298 | 0.98  | 0.082 | 0.618 |
| 20         | 8                  | 0.20  | 0.170 | -1.06 | 0.420 | 0.74  | 0.153 | 0.528 |
| 0          | 0                  | 0.28  | 0.236 | -0.47 | 0.264 | 0.95  | 0.198 | 0.449 |
| 0          | 8                  | 0.54  | 0.343 | -0.36 | 0.202 | 0.19  | 0.295 | 0.605 |
| 'Braeburn' |                    |       |       |       |       |       |       |       |
| 20         | 0                  | 0.37  | 0.165 | -1.02 | 0.195 | 0.70  | 0.151 | 0.780 |
| 20         | 2                  | 0.03  | 0.010 | -1.94 | 0.146 | 0.69  | 0.024 | 0.921 |
| 20         | 4                  | 0.03  | 0.020 | -2.52 | 0.401 | 0.58  | 0.033 | 0.830 |
| 20         | 6                  | 0.11  | 0.066 | -2.05 | 0.349 | 0.67  | 0.074 | 0.768 |
| 20         | 8                  | 0.10  | 0.027 | -1.94 | 0.164 | 0.66  | 0.023 | 0.923 |
| 0          | 0                  | 0.02  | 0.039 | -1.64 | 0.810 | 1.09  | 0.060 | 0.460 |
| 0          | 8                  | 0.46  | 0.450 | -0.32 | 0.279 | 0.46  | 0.405 | 0.253 |

kPa  $p_{CO_2}^e$  in  $FT_{RQ}^i$  and  $FT_{EtOH}^i$  at 0°C may have been a consequence of higher solubility of  $CO_2$  at the lower temperature, and this might have accentuated the effect of a given  $p_{CO_2}^e$  on fruit tolerance to low  $p_{O_2}^i$ . Alternatively, since the vapour pressure of EtOH above aqueous solutions would be highly temperature sensitive, the onset of fermentation may be more difficult to detect by measuring vapour phase levels of EtOH in fruit kept at the temperatures used in the experiment. Therefore, it is possible that the accumulation of EtOH in chambers did not reflect tissue concentrations because either values for chamber EtOH were not at steady-state or were not measured with great accuracy.

Higher average  $p_{CO_2}^i$  and lower  $p_{O_2}^i$  of 'Braeburn' compared to 'Cox's Orange Pippin' apples (0.7 kPa  $p_{CO_2}^i$  and 1.0 kPa  $p_{O_2}^i$  respectively at 20°C in air) probably resulted from the lower skin permeance of 'Braeburn' fruit. However, both cultivars have high  $p_{CO_2}^i$  compared to cultivars (such as 'Golden Delicious' and 'Splendour' (2.5 and 2.0 kPa  $p_{CO_2}^i$  respectively; Dadzie, 1992) which have similar rates of respiration to 'Braeburn' but markedly greater skin permeance. High  $p_{CO_2}^i$  in 'Cox's Orange Pippin' and 'Braeburn'

fruit contributes to the development of  $CO_2$ -related physiological storage disorders (core flush and internal browning in 'Cox's Orange Pippin' (Meheriuk et al., 1994), and the brown-heart like 'Braeburn' browning disorder in 'Braeburn' (Curry, 1996). Development of 'Braeburn' browning disorder can be induced in 'Braeburn' apples by storage at 0°C in  $p_{CO_2}^e > 2.0$  kPa (balance air), and further exacerbated when  $p_{O_2}^e$  is reduced below (N.H. Banks, personal communication). In the current study no visible external or internal damage was observed in experimental fruit stored in anoxic atmospheres for up to 7 days and with  $p_{CO_2}^e$  of 8 kPa  $CO_2$ . Brief exposures to potentially harmful levels of  $CO_2$  in fruit may be tolerated for short periods (Ke and Kader, 1992). However, it is probable that damage would develop in fruit during longer storage periods in such high  $CO_2$  atmospheres.

Higher  $ACP^i$  for 'Braeburn' compared to 'Cox's Orange Pippin' apples at approximately 0 kPa  $p_{CO_2}^e$  and for fruit at 20°C compared to 0°C (for  $FT_{RQ}^i$  and  $FT_{EtOH}^i$ ), were consistent with those reported for the same cultivars from the previous season (Yearsley et al., 1996b). As suggested in that work, slight increases in  $LOL^i$ s as a



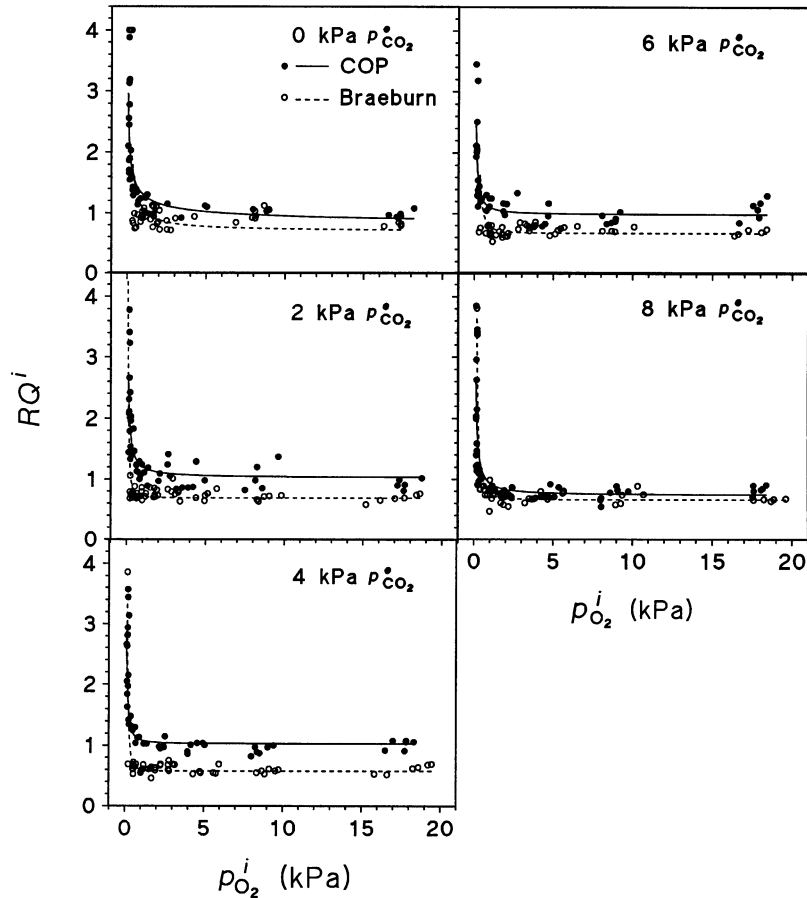


Fig. 5. Respiratory quotient ( $RQ^i$ ) as a function of the internal (chamber) steady-state partial pressure of  $O_2$  ( $p_{O_2}^i$ ) for 'Cox's Orange Pippin' (COP) and 'Braeburn' apples at  $20^\circ\text{C}$ , stored in 0, 2, 4, 6 or 8 kPa external partial pressure of  $CO_2$  ( $p_{CO_2}^e$ ), balance  $N_2$ . For estimates of parameters of the curves see Table 2.

response to increasing temperature, and lower  $LOL$ 's for 'Cox's Orange Pippin' apples could have been influenced by higher tissue porosity for 'Cox's Orange Pippin' as well as the effect of increasing temperature lowering the solubility of gases. As cortical tissue porosity and density did not change significantly with time in storage for 'Braeburn' fruit, and the small changes in porosity and density for 'Cox's Orange Pippin' were not consistent with time, they were unlikely to have influenced  $LOL$ 's in this study. However, lower porosity and higher density of 'Braeburn' fruit may potentially reduce diffusivity of  $O_2$  through the cortex and contribute to increased  $LOL$ 's of 'Braeburn' compared to 'Cox's Orange Pippin'

fruit. The larger gradients in  $O_2$  partial pressure ( $\Delta p_{O_2}$ ) for 'Braeburn' compared to 'Cox's Orange Pippin' fruit at  $20^\circ$  (Fig. 3) were probably a consequence of the markedly lower skin permeance, as the respiration rate as 'Braeburn' was lower than that for 'Cox's Orange Pippin'. Differences in  $\Delta p_{O_2}$  were attenuated at  $0^\circ\text{C}$  because of the lower respiration rate at that temperature. As  $LOL$ 's should be independent of skin permeance, these differences would not have contributed to variation in  $LOL$ 's between cultivars.

In general, elevated  $CO_2$  had a small effect on reduced  $r_{O_2}$  (as reflected by  $\Delta p_{O_2}$ ) for both cultivars at  $20^\circ\text{C}$  compared to fruit in 0 kPa  $p_{CO_2}^e$ . Although the fitted  $\Delta p_{O_2}$  curve for 'Cox's Orange

Pippin' at 0°C, 8 kPa  $p_{\text{CO}_2}^e$  was slightly higher than that for the 0 kPa  $p_{\text{CO}_2}^e$  treatment, the change in  $\Delta p_{\text{O}_2}$  was too small, and the variation in 'Braeburn' data at 8 kPa  $p_{\text{CO}_2}^e$  too variable, to indicate a significant effect of elevated  $p_{\text{CO}_2}^e$  on  $r_{\text{O}_2}$  at 0°C. The change in apparent Michaelis–Menten constant (values of  $k_2$  in Table 1) was not consistent with change in  $p_{\text{CO}_2}^e$ . However, the higher values of  $k_2$  for 'Braeburn' apples indicated depression of  $p_{\text{O}_2}^i$  had a more marked effect on  $r_{\text{O}_2}$  of 'Braeburn' compared to 'Cox's Orange Pippin', suggesting involvement of oxidases with lower affinity in 'Braeburn' than 'Cox's Orange Pippin' or differences in gradients in  $p_{\text{O}_2}^i$  within the flesh.

A number of studies with apples indicate that elevated  $\text{CO}_2$  may reduce  $RQ$  (Fidler and North, 1967; Metlitskii et al., 1972). The slight lowering of  $RQ^i$  at elevated  $\text{CO}_2$  as observed in this study suggested there was a larger effect of  $\text{CO}_2$  on  $r_{\text{CO}_2}$  than  $r_{\text{O}_2}$ , and resulted in smaller  $\Delta p_{\text{CO}_2}$  compared with  $\Delta p_{\text{O}_2}$ . Elevated  $\text{CO}_2$  (5%) has also been reported to reduce  $r_{\text{CO}_2}$  to a larger extent than  $r_{\text{O}_2}$  of postclimacteric 'Cox's Orange Pippin' (Fidler and

North, 1967). This may result from increased fixation of  $\text{CO}_2$  into organic acids, and reduced catabolism of organic acids (Murata and Minamide, 1970). The higher  $RQ^i$  of 'Braeburn' at 0°C compared to 20°C may have resulted from a higher temperature coefficient for  $r_{\text{O}_2}$  compared to  $r_{\text{CO}_2}$  (Fidler and North, 1967). However,  $RQ^i$  is not an absolute measure of  $RQ$ , and depends both on  $RQ$  and the relative permeance of the fruit skin to  $\text{CO}_2$  and  $\text{O}_2$ . Therefore, lower  $RQ^i$  for 'Braeburn' compared to 'Cox's Orange Pippin' apples at 20°C (Fig. 5, and  $k_5$  values in Table 2), indicated 'Braeburn' fruit had a higher permeance to  $\text{CO}_2$  relative to  $\text{O}_2$  compared to 'Cox's Orange Pippin' apples, as has been reported (Yearsley et al., 1996c). This may have indicated a higher permeability of the cuticle to  $\text{CO}_2$  than  $\text{O}_2$ , and/or a larger component of diffusion occurred through the cuticle compared to pores for 'Braeburn' compared to 'Cox's Orange Pippin' apples (Banks et al., 1993b). The slightly lower  $RQ^i$  for COP and particularly 'Braeburn' apples at 20°C compared with 0°C and 0 kPa  $p_{\text{CO}_2}^e$  (compare Figs. 5 and 6), might also be explained by differential effects of temperature on diffusion through cuticles and pores. With increasing temperature, cuticular diffusion would increase more than diffusion through pores which is relatively temperature-independent (Cameron et al., 1994). As  $\text{O}_2$  diffusion in apples is dominated by pores, and diffusion of  $\text{CO}_2$  occurs through pores and cuticle (Banks et al., 1993b), increasing temperature would have a greater effect on increasing  $P'_{\text{CO}_2}$  than  $P'_{\text{O}_2}$ , resulting in decreased  $RQ^i$ .

$\text{CO}_2$ -related disorders include browning and cavities in defined areas of cortex and core tissue injury (Meheriuk et al., 1994), 'scald-like' symptoms purported to result from free-radical-catalysed oxidation of proteins and other macromolecules (Burmeister and Dilley, 1995), and interactions with low temperature injury (Padfield, 1969).  $\text{CO}_2$ -related disorders may develop at  $p_{\text{O}_2}^i$  and  $p_{\text{CO}_2}^i$  above the onset of fermentation, reducing mitochondrial function and enhancing other oxidative processes. As both 'Cox's Orange Pippin' and 'Braeburn' apples are susceptible to  $\text{CO}_2$ -related disorders during cool-storage in elevated  $\text{CO}_2$ , recommendations for

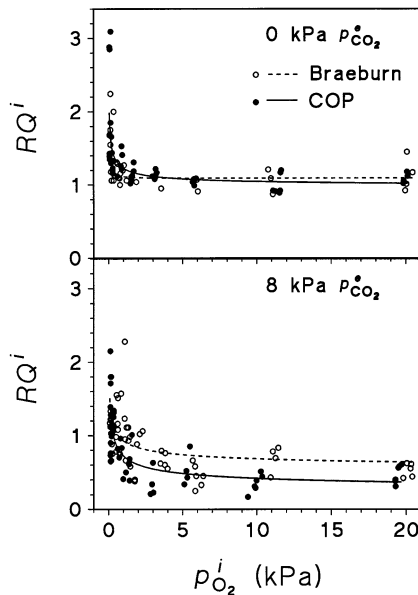


Fig. 6. Respiratory quotient ( $RQ^i$ ) as a function of the internal (chamber) steady-state partial pressure of  $\text{O}_2$  ( $p_{\text{O}_2}^i$ ) for 'Cox's Orange Pippin' (COP) and 'Braeburn' apples at 0°C, stored in 0 and 8 kPa external partial pressure of  $\text{CO}_2$  ( $p_{\text{CO}_2}^e$ ), balance  $\text{N}_2$ . For estimates of parameters of the curves see Table 2.

optimum storage atmospheres based on *LOL*'s would need to be adjusted for the potential to develop disorders.

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## References

- Banks, N.H., Cleland, D.J., Yearsley, C.W., Kingsley, A.M., 1993a. Internal atmosphere composition—a key concept in responses of fruits and vegetables to modified atmospheres. In: Proceedings of the Australasian Postharvest Conference. The University of Queensland, Gatton College, Lawes, Queensland, Australia, pp 137–143.
- Banks, N.H., Dadzie, B.K., Cleland, D.J., 1993b. Reducing gas exchange of fruits with surface coatings. *Postharv. Biol. Technol.* 3, 269–284.
- Beaudry, R.M., 1993. Effect of carbon dioxide partial pressure on blueberry fruit respiration and respiratory quotient. *Postharv. Biol. Technol.* 3, 249–258.
- Beaudry, R.M., Cameron, A.C., Shirazi, A., Dostal-Lange, D.L., 1992. Modified-atmosphere packaging of blueberry fruit: effect of temperature on package O<sub>2</sub> and CO<sub>2</sub>. *J. Am. Soc. Hort. Sci.* 117, 436–441.
- Beaudry, R.M., Gran, C.D., 1993. Using a modified-atmosphere packaging approach to answer some post-harvest questions: factors influencing the lower oxygen limit. *Acta Hort.* 326, 203–212.
- Burmeister, D.M., Dilley, D.R., 1995. A 'scald-like' controlled atmosphere storage disorder of Empire apples—a chilling injury induced by CO<sub>2</sub>. *Postharv. Biol. Technol.* 6, 1–7.
- Cameron, A.C., Beaudry, R.M., Banks, N.H., Yelanich, M.V., 1994. Modified-atmosphere packaging of blueberry fruit: Modelling respiration and package oxygen partial pressures as a function of temperature. *J. Am. Soc. Hort. Sci.* 119, 534–539.
- Cameron, A.C., Talasila, P.C., Joles, D.W., 1995. Predicting film permeability needs for modified-atmosphere packaging of lightly processed fruits and vegetables. *HortSci.* 30, 25–34.
- Curry, E.A., 1996. Maintaining optimum fruit quality of 'Braeburn' apples. *Washington State Univ. Tree Fruit Postharv. J.* 7, 3–7.
- Dadzie, B.K., 1992. Gas exchange characteristics and quality of apples. Ph.D. Dissertation, Massey University, Palmerston North, New Zealand, pp. 339
- Dadzie, B.K., Banks, N.H., Cleland, D.J., Hewett, E.W., 1993. Role of skin resistance to gas diffusion in the response of fruits to modified atmospheres. *Acta Hort.* 343, 129–134.
- Dadzie, B.K., Banks, N.H., Cleland, D.J., Hewett, E.W., 1996. Changes in respiration and ethylene production of apples in response to internal and external partial pressures. *New Zealand J. Crop Hort. Sci.* 9, 297–309.
- Fidler, J.C., North, C.J., 1967. The effect of the conditions of storage on the respiration of apples. I The effects of temperature and concentrations of carbon dioxide and oxygen on the production of carbon dioxide and uptake of oxygen. *J. Hort. Sci.* 42, 189–206.
- Fig. P, 1991. Fig. P Software Corporation, Durham, North Carolina.
- Gran, C.D., 1993. Fruit respiration and determination of low oxygen limits for apple (*Malus domestica* Borkh.) fruit. M.S. Dissertation, Michigan State University, East Lansing, MI, p. 116.
- Gran, C.D., Beaudry, R.M., 1993a. Determination of the low oxygen limit for several commercial apple cultivars by respiratory quotient breakpoint. *Postharv. Biol. Technol.* 3, 259–267.
- Gran, C.D., Beaudry, R.M., 1993b. Modified atmosphere packaging determination of the low oxygen limit for apple fruit using respiratory quotient and ethanol accumulation. Proceedings 6<sup>th</sup> International CA Research Conference, Cornell University, Ithaca, New York. pp. 54–62.
- Joles, D.W., Cameron, A.C., Shirazi, A., Petracek, P.D., Beaudry, R.M., 1994. Modified-atmosphere packaging of 'Heritage' red raspberry fruit: respiratory response to reduced oxygen, enhanced carbon dioxide, and temperature. *J. Am. Soc. Hort. Sci.* 119, 540–545.
- Kader, A.A., Zagory, D., Kerbel, E.L., 1989. Modified atmosphere packaging of fruits and vegetables. *Crit. Rev. Food Sci. Nutr.* 28, 1–30.
- Ke, D., Kader, A.A., 1992. Potential of controlled atmospheres for postharvest insect disinfestation of fruits and vegetables. *Postharv. News Info.* 3, 31N–37N.
- Ke, D., Mateos, M., Kader, A.A., 1993. Regulation of fermentative metabolism in fruits and vegetables by controlled atmospheres. Proceedings 6<sup>th</sup> International CA Research Conference, Cornell University, Ithaca, New York. pp. 63–67.
- Kerbel, E.L., Kader, A.A., Romani, R.J., 1988. Effects of elevated CO<sub>2</sub> concentrations on glycolysis in intact 'Bartlett' pear fruit. *J. Am. Soc. Hort. Sci.* 115, 975–978.
- Kidd, F., 1916. The controlling influence of carbon dioxide. Part III. The retarding effect of carbon dioxide on respiration. *Proc. R. Soc. (London)* 89, 136–156.
- Kidd, F., West, C., 1927. A relation between the concentration of oxygen and carbon dioxide in the atmosphere, rate of respiration, and length of storage of apples. *Rep. Food Invest. Bd.*, for 1925, 1926, DSIR, London, pp. 41–42.
- Kidd, F., West, C., 1933. Effect of ethylene and of apple vapours on the ripening of fruits. *Rep. Food Invest. Bd.* for the year 1932, DSIR, London, pp. 55–58.

- Kubo, Y., Inaba, A., Nakamura, R., 1990. Respiration and  $C_2H_4$  production in various crops held in  $CO_2$ -enriched atmospheres. *J. Am. Soc. Hort. Soc.* 115, 975–978.
- Li, C., Kader, A.A., 1989. Residual effects of controlled atmospheres on postharvest physiology and quality of strawberries. *J. Am. Soc. Hort. Sci.* 114, 629–634.
- Meheriuk, M., Prange, R.K., Lidster, P.D., Porritt, S.W., 1994. Postharvest disorders of apples and pears. *Agric. Canada, Publ. 1737/E*, Ottawa, Ont., Canada.
- Metlitskii, L.V., Sal'kova, E.G., Volkind, N.L., Bondarev, V.L., Yanyuk, V.Y., 1972. Controlled atmosphere storage of fruits. *E'konomika*, Moscow. (English translation, Dhote, A.K., 1983. Amerind, New Delhi.)
- Murata, T., Minamide, T., 1970. Studies on organic acid metabolism and ethylene production during controlled atmosphere storage of apples (*Malus pumila* Miller, cv Rolls). *Plant Cell Physiol.* 11, 857–863.
- Padfield, C.A.S., 1969. The Storage of Apples and Pears. *New Zealand DSIR, Bulletin* 111. pp. 117.
- Peppelenbos, H.W., van't Leven, J., van Zwol, B.H., Tijsskens, L.M.M., 1993. The influence of  $O_2$  and  $CO_2$  on the quality of fresh mushrooms. *Proceedings 6<sup>th</sup> International. CA Research Conference*, Cornell University, Ithaca, New York. pp. 746–758.
- SAS, 1990. SAS Institute Inc. Cary, NC.
- Thornton, N.C., 1933. Carbon dioxide storage. III. The influence of carbon dioxide on the oxygen uptake by fruits and vegetables and flowers. *Contrib. Boyce Thompson Inst.* 5, 371–402.
- Yearsley, C.W., Banks, N.H., Ganesh, S., Cleland, D.J., 1996a. Determination of lower oxygen limits for apple fruit. *Postharv. Biol. Technol.* 8, 95–109.
- Yearsley, C.W., Banks, N.H., Ganesh, S., 1996b. Temperature effects on the lower internal oxygen limits of apple fruit. *Postharv. Biol. Technol.* 11, 73–83.
- Yearsley, C.W., Banks, N.H., Ganesh, S., 1996c. Effect of physiological age on the lower internal oxygen limits of apple fruit. *Postharv. Biol. Technol.* (submitted).
- Young, R.E., Romani, R.J., Biale, J.B., 1962. Carbon dioxide effects on fruit respiration. II. Response of avocados, bananas and lemons. *Plant Physiol.* 37, 416–422.

markedly with time in storage over the experimental period. On average,  $p_{\text{CO}_2}^i$  of 'Braeburn' apples at 20°C in air was 0.7 kPa higher than for 'Cox's Orange Pippin' fruit (5.18 and 4.48 kPa  $\text{CO}_2$  respectively). Conversely,  $p_{\text{O}_2}^i$  of 'Braeburn' apples at 20°C in air was 1.0 kPa lower than for 'Cox's Orange Pippin' fruit (16.47 and 17.49 kPa  $\text{O}_2$  respectively).

There was a tendency for  $r_{\text{CO}_2}$  and  $r_{\text{C}_2\text{H}_4}$  to

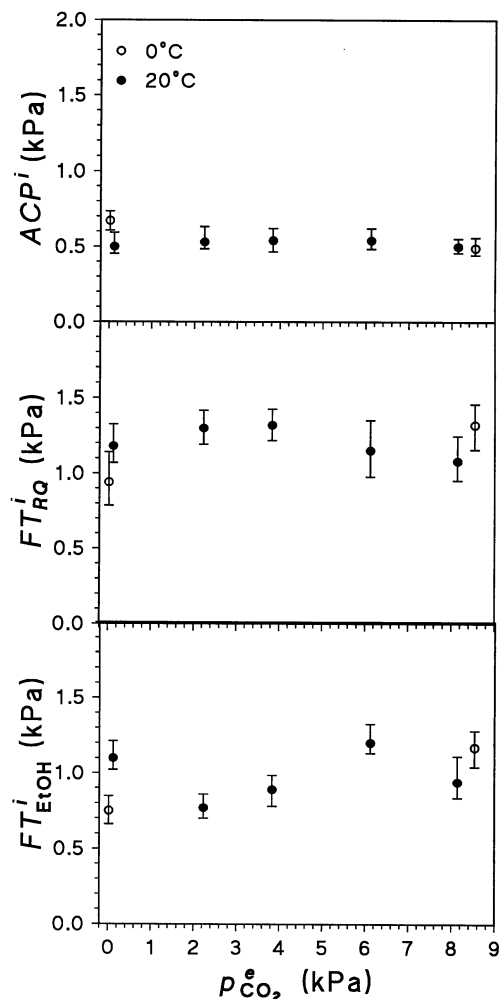


Fig. 1. Bootstrap estimates of steady state internal lower oxygen limits ( $ACP^i$ ,  $FT^i_{RQ}$ , and  $FT^i_{EtOH}$ ) based on partial pressures of surface chambers for 'Cox's Orange Pippin' apples as a function of external partial pressure of  $\text{CO}_2$  ( $p_{\text{CO}_2}^e$ ). Values represent bootstrap means and 95% bias corrected (BCa-type) confidence intervals.

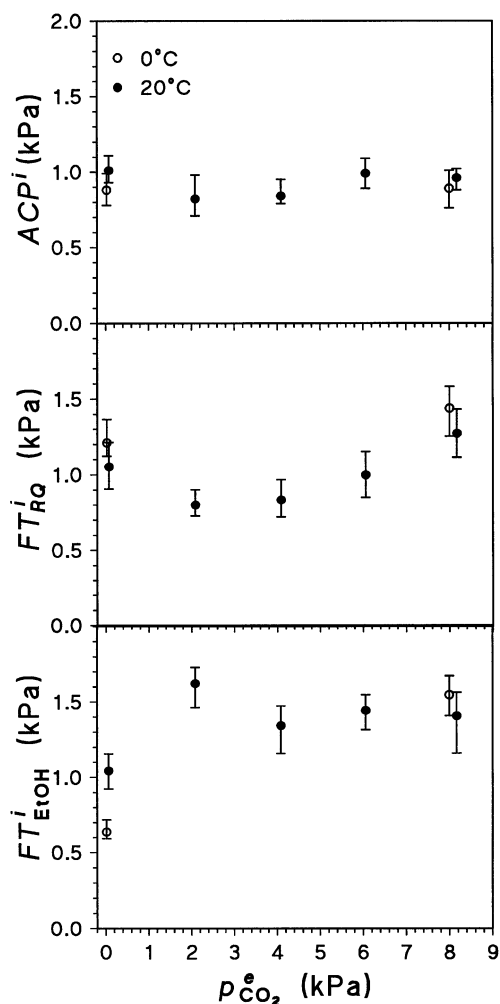


Fig. 2. Bootstrap estimates of steady state internal lower oxygen limits ( $ACP^i$ ,  $FT^i_{RQ}$ , and  $FT^i_{EtOH}$ ) based on partial surface chambers for 'Braeburn' apples as a function of external partial pressure of  $\text{CO}_2$  ( $p_{\text{CO}_2}^e$ ). Values represent bootstrap means and 95% bias corrected (BCa-type) confidence intervals.

increase with time in storage for both cultivars at both 0 and 20°C, but increases were not always significantly different between each assessment time. Changes were consistent with those expected for apples ripening in cool storage. Mean  $r_{\text{CO}_2}$  of 'Cox's Orange Pippin' fruit at 0 and 20°C was 28 and 177  $\text{nmol kg}^{-1} \text{s}^{-1}$  respectively, and of 'Braeburn', 18 and 124  $\text{nmol kg}^{-1} \text{s}^{-1}$  respectively. Mean  $r_{\text{C}_2\text{H}_4}$  of 'Cox's Orange Pippin' fruit at 0° and 20°C was 0.15 and 1.42  $\text{nmol kg}^{-1} \text{s}^{-1}$