Modelling respiration of packaged fresh-cut ‘Rocha’ pear as affected by oxygen concentration and temperature

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ABSTRACT

Respiration rates were measured in fresh-cut ‘Rocha’ pear (Pyrus communis L.) stored at four temperatures (0, 5, 10 and 15 °C) and with oxygen partial pressures ranging from 0 to 18 kPa. Respiratory quotients and ethanol production were used to determine the fermentation threshold. The oxygen concentration effect on the respiration rate was accurately described using Michaelis–Menten kinetics, without non-competitive inhibition by CO2, and the effect of temperature on the respiration rate was well modelled by exponential functions. The oxygen level at which respiration was half its maximum (apparent KmCO2) was similar to or only slightly greater than the fermentation threshold. The narrow range of oxygen between KmCO2 and the fermentation threshold, suggests that modified atmosphere packaging technology has a limited applicability toward extension of the shelf-life of fresh-cut ‘Rocha’ pear.

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1. Introduction

Respiration is a central process in fruit metabolism: it provides energy and carbon skeletons for the anabolic reactions that occur during maturation and ripening, and for cell maintenance during storage. Besides its physiological importance, knowledge of fruit respiratory parameters possesses a technological relevance, particularly toward development of modified atmosphere packaging (MAP). Fruit respiration rate is affected by a number of factors, among which temperature and oxygen partial pressure exert strong effects (Fonseca et al., 2002). In fresh-cut fruit wounding enhances the respiration rate as part of the healing and ethylene responses (Brecht, 1995).

Fresh-cut processing often uses barrier technologies to modify atmosphere composition, prevent contamination, and reduce water loss (Kader et al., 1989). Since fresh-cut fruit respires, hermetically-sealed barriers used in these convenience products result in a modified atmosphere inside the package. Since MAP can extend the shelf-life and marketing period of fresh-cut fruit (Bai et al., 2001; Martinez-Ferrer et al., 2002) a great deal of effort has been dedicated to develop and test appropriate packages, and to optimize oxygen and carbon dioxide partial pressures inside the packages (Kader et al., 1989; Fonseca et al., 2002). A number of studies encompassing packaging films and geometries have assessed O2 concentration versus produce quality, in order to empirically find the best package (Bai et al., 2003; Del Nobile et al., 2007; SolivaFortuny et al., 2007; Teixeira et al., 2007). An alternative approach to the trial-and-error, should data be available, is to deduce optimal packaging geometry and film permeability based on the product respiration rate (Lakakul et al., 1999; Jaccobsen et al., 2000), as affected by oxygen concentration and temperature.

Respiration rates of fresh-cut pear were reported for ‘d’Anjou’, ‘Bartlett’, ‘Bosc’ and ‘Red Anjou’ cultivars stored in air and in 2 kPa oxygen (Gorny et al., 2000). Values range between 0.07–0.22 and 0.27–0.53 mmol CO2 kg−1 h−1, respectively at 0 and 10 °C, rising to 0.86–1.63 mmol CO2 kg−1 h−1 at 20 °C. However, detailed information on the effect of oxygen concentration on the respiration of fresh-cut pear at different temperatures is still unavailable.

The impact of MAP on fresh-cut produce has been shown to be effective for some commodities; however benefits have not been demonstrated for fresh-cut pear. Further, the requirement for packaging fresh-cut fruit in sealed packages, to maintain produce safety and quality, will result in a modified atmosphere. Therefore, it is essential to quantify the relationship between oxygen concentration and respiration rate for fresh-cut ‘Rocha’ pear, stored at different temperatures, and to test respiratory models that would allow the design of MAP for fresh-cut pear at temperatures normally found during processing and distribution.
2. Materials and methods

2.1. Plant material

Pears (Pyrus communis L. ‘Rocha’) were harvested in two consecutive years, 2006 and 2007, at commercial maturity (65 N firmness), from orchards located in the Western Region of Portugal (39°11′N, 9°08′W). ‘Rocha’ pear has a long storage life and raw material is available for fresh-cut processing for up to ten months. Pears used in 2006 had been stored for 6 months at ~0.5 °C, 93–95% RH, and 2.5 kPa O2 plus 0.7 kPa CO2. Fruits processed in 2007 had been stored at ~0.5 °C and 93–95% RH in air for 5 weeks. Fruits removed from storage were allowed to ripen and were processed when the flesh firmness reached 47–49 N, since partially ripe pears are the most suitable for fresh-cut processing (Gorny et al., 2000; Soliva-Fortuny et al., 2004).

2.2. Fresh-cut preparation and packaging

Whole fruits were sanitized with 150 μL L−1 NaClO for 2 min, rinsed with tap water and air dried. Pears were cut in wedges with 5–10 mm of thickness, without skin removal. The fresh-cut pear wedges (30–265 g) were packed in low-density polyethylene (LDPE) (Dow Chemical Company, Midland, MI) pouches (15 × 15 cm or 20 × 20 cm), that were hermetically-sealed using an impulse heat sealer. The film thickness used in the experiments ranged from 27.0 to 102.1 μm.

Package size, film thickness, and fruit mass were varied to intentionally develop a range of oxygen concentrations (18–210 mmol kg−1 h−1), to purge receiver chamber of oxygen. Once receiver chamber oxygen levels were reduced below detection limits, a mixture of 72 kPa O2 and 29 kPa CO2 was supplied to the donor chamber. The receiver gas composition was sampled (100 μL), using a glass syringe, at 10 min intervals and analysed via sequential O2 and CO2 analyzers, using a paramagnetic O2 detector (Series 1100, Servomex Co., Sussex, UK) and an infrared CO2 detector (ADC 255-MK3, Analytical Development Co., Hoddesdon, UK), respectively, and connected in series (Lakakul et al., 1999). The diffusion rate of gases through the film was used to calculate the permeability coefficients, at 0.6, 10, 21 and 23 °C, in controlled temperature chambers. The Arrhenius equation was used to fit the temperature influence on permeability (Beaudry et al., 1992; Joles et al., 1994; Cameron et al., 1995; Lakakul et al., 1999). The pre-exponential factor and activation energies were estimated via regression analysis, after linearization (Beaudry et al., 1992): ln (P) = −Ea/RT + ln (P0), where P is permeability to O2 or CO2 (mmol cm−2 h−1 kPa−1), P0 is the pre-exponential factor, Ea is activation energy (kJ mol−1), T is temperature (K).

2.3. Respiration rate determination

A permeable system using LDPE of known permeability was used to obtain the respiratory rate of the enclosed produce. Rates of O2 uptake (R02) and CO2 production (RCO2), and respiratory quotient (RQ = RCO2/R02), were calculated once steady-state O2 and CO2 partial pressures were achieved inside the packages, using the following equations (Beaudry et al., 1992; Joles et al., 1994; Lakakul et al., 1999):

\[ R_{O_2} = \frac{P_{O_2, atm} \times (p_{O_2, atm} - p_{O_2, pkg})}{M} \]  \hspace{1cm} (1)

\[ R_{CO_2} = \frac{P_{CO_2, atm} \times (p_{CO_2, pkg} - p_{CO_2, atm})}{M} \]  \hspace{1cm} (2)

where \( R_{O_2} \) and \( R_{CO_2} \) denote respiration rates (mmol kg−1 h−1) of O2 and CO2, \( P_{O_2} \) and \( P_{CO_2} \) denote O2 and CO2 permeabilities (mmol cm−2 h−1 kPa−1), \( A \) is the film area (cm2), \( l \) is the film thickness (cm), \( M \) is the produce mass (kg), \( P \) is partial gas pressure (kPa), \( p \) is partial gas pressure (kPa), \( p_{O_2} \) and \( p_{CO_2} \) denote O2 and CO2 partial pressures (kPa) outside (atm) and inside (pkg) the package, and \( M \) is the produce mass (kg).

Three replicates of each combination of film thickness, film area, and fruit mass were stored at 0, 5, 10 and 15 °C.

2.4. Film permeability assessment

Film permeability to O2 (\( P_{O_2} \)) and CO2 (\( P_{CO_2} \)) was measured using an isostatic method (Beaudry et al., 1992; Gavara et al., 1996). The permeation cell contained two external receiver chambers separated from a middle donor chamber by film samples. The middle chamber was initially flushed (100 ml min−1) with pure N2 to purge receiver chamber of oxygen. Once receiver chamber oxygen levels were reduced below detection limits, a mixture of 72 kPa O2 and 29 kPa CO2 was supplied to the donor chamber. The receiver gas composition was sampled (100 μL), using a glass syringe, at 10 min intervals and analysed via sequential O2 and CO2 analyzers, using a paramagnetic O2 detector (Series 1100, Servomex Co., Sussex, UK) and an infrared CO2 detector (ADC 255-MK3, Analytical Development Co., Hoddesdon, UK), respectively, and connected in series (Lakakul et al., 1999). The diffusion rate of gases through the film was used to calculate the permeability coefficients, at 0.6, 10, 21 and 23 °C, in controlled temperature chambers. The Arrhenius equation was used to fit the temperature influence on permeability (Beaudry et al., 1992; Joles et al., 1994; Cameron et al., 1995; Lakakul et al., 1999). The pre-exponential factor and activation energies were estimated via regression analysis, after linearization (Beaudry et al., 1992): ln (P) = −Ea/RT + ln (P0), where P is permeability to O2 or CO2 (mmol cm−2 h−1 kPa−1), P0 is the pre-exponential factor, Ea is activation energy (kJ mol−1), T is temperature (K).

2.5. O2, CO2, and ethanol determination

Headspace gas composition (O2 and CO2) in individual packages was monitored daily using a gas analyzer, as described above, until steady-state was attained. The ethanol present in the headspace was sampled at steady-state and determined using a gas chromatograph system (HP-6890, Hewlett Packard, USA), equipped with a mass spectrometry detector.

2.6. Experimental data modelling

Respiration rate (\( R_{O_2} \)) was described as a function of O2 partial pressure by a Michaelis–Menten model (Eq. (3)), as suggested by Lee et al. (1991):
\[ R_O = K_{m,O_2} + pO_2 \text{pkg} \]

where \( K_{m,O_2} \) is the Michaelis–Menten constant for the non-competitive CO\(_2\) inhibition.

Estimation of \( R_{O_2}^{m,x,T} \) and \( K_{m,O_2} \), based on experimental data, was performed by non-linear regression using the Levenberg–Marquardt method. The estimates obtained at each temperature were used to describe the temperature dependence of \( R_{O_2}^{m,x,T} \) and \( K_{m,O_2} \). In addition, the Michaelis–Menten models were fitted to the whole data set at all temperatures \((n = 129 \text{ in 2006 and } n = 154 \text{ in 2007). In both models, } R_{O_2}^{m,x,T} \text{ was an exponential function of temperature, and } K_{m,O_2} \text{ could be modelled as either an exponential function of temperature or a constant, whereas } K_{m,CO_2} \text{ was always a constant (Table 1).}

The predictions of respiratory parameters, \( R_{O_2}^{m,x,T} \) and \( K_{m,O_2} \), at 0, 5, 10 and 15 °C, generated by solving the equations depicted on Table 4, were used to calculate activation energies, through an Arrhenius plot (Eqs. (5) and (6)), and the temperature coefficient \( (Q_10) \).

\[ \ln \left( \frac{R_{O_2}^{m,x,T} \text{kg}}{R_{O_2}^{m,x,T} \text{kg}} \right) = \ln \left( \frac{R_{O_2}^{m,x,T} \text{kg}}{R_{O_2}^{m,x,T} \text{kg}} \right) - \frac{E_a}{R} \left( \frac{1}{T} - \frac{1}{T} \right) \]

\[ \ln \left( K_{m,O_2} \text{kg} \right) = \ln \left( K_{m,O_2} \text{kg} \right) - \frac{E_a}{R} \left( \frac{1}{T} - \frac{1}{T} \right) \]

where \( R_{O_2}^{m,x,T} \) is the maximal value of \( R_{O_2} \) (mmol kg \(^{-1} \) h \(^{-1} \)), \( R_{O_2}^{m,x,T} \) and \( K_{m,O_2} \) are the pre-exponential factors, \( K_{m,O_2} \) (apparent \( K_{m,O_2} \)) is the \( pO_2 \) kg at half of \( R_{O_2}^{m,x,T} \) (kPa), \( E_a \) is activation energy (kJ mol \(^{-1} \)), \( R \) is ideal gas constant (kJ mol \(^{-1} \) K \(^{-1} \)) and \( T \) is temperature (K).

The accuracy of parameters estimation was given by the ratio of the standard error to value estimate. The likelihood of the fit was ascertained by the root mean square error (RMSE) (Yang and Chinnan, 1988).

All statistical analyses were performed using the software package SPSS for Windows v.16.0 (SPSS, Chicago, IL).

### 3. Results and discussion

#### 3.1. Film permeability

Permeability to \( O_2 \) and \( CO_2 \) and their temperature dependence are depicted in Fig. 1. The predicted permselectivity \((\beta = P_{CO_2}/P_{O_2})\) at 0 °C was 4.7 and slightly lower (4.4) at 20 °C. These permeabilities are similar to previous published results by Joles et al. (1994) (4.8 at 20 °C and 4.9 at 0 °C) and by Beaudry et al. (1992) (4.4 at 25 °C and 5.2 at 0 °C). The activation energy for \( P_{O_2} \) was 36.8 kJ mol \(^{-1} \) (Table 2), a value similar to those reported elsewhere (Beaudry et al., 1992; Joles et al., 1994; Lakakul et al., 1999), but slightly below the 40–50 kJ mol \(^{-1} \) range claimed for LDPE films used in MAP (Cameron et al., 1995). Activation energies for \( P_{CO_2} \) were 34.5 kJ mol \(^{-1} \) (Table 2), which were essentially identical to the values 35.0 kJ mol \(^{-1} \) (Lakakul et al., 1999) and 35.5 kJ mol \(^{-1} \) (Beaudry et al., 1992; Joles et al., 1994).

MAP design requires knowledge of permeability coefficients in the range of temperatures found during processing and marketing. Film suppliers usually provide \( O_2 \) transmission rate determined at 0 °C and 25 °C, but without correction of film \( O_2 \) permeability, would lead to gross overestimation.

#### 3.2. Effect of oxygen concentration and temperature on respiration rate

Fruit at the same maturity stage were harvested in two consecutive years and stored for 6 months or 5 weeks, in 2006 and 2007, respectively. Despite the differences in storage durations, respiratory parameters did not differ significantly between years, likely because fruit were processed and analysed with similar maturities, as assessed by flesh firmness.

### Table 1
Models used to describe the effects of oxygen concentration and temperature on the respiration rate of fresh-cut ‘Rocha’ pear.

<table>
<thead>
<tr>
<th>Model label</th>
<th>Model equation</th>
<th>Model parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>MM</strong></td>
<td>( R_O = \frac{R_{O_2}^{m,x,T} \times pO_2 \text{pkg}}{K_{m,O_2} + pO_2 \text{pkg}} )</td>
<td>( a \times e^{b \times T} ) ( q \times e^{r \times T} ) ( \text{Constant} ) ( \text{Constant} )</td>
</tr>
<tr>
<td><strong>MMk</strong></td>
<td>( R_O = \frac{R_{O_2}^{m,x,T} \times pO_2 \text{pkg}}{K_{m,O_2} + pO_2 \text{pkg}} )</td>
<td>( a \times e^{b \times T} ) ( \text{Constant} ) ( \text{Constant} )</td>
</tr>
<tr>
<td><strong>MMNC</strong></td>
<td>( R_O = \frac{R_{O_2}^{m,x,T} \times pO_2 \text{pkg}}{\left</td>
<td>K_{m,O_2} + pO_2 \text{pkg} / K_{m,CO_2} \right</td>
</tr>
<tr>
<td><strong>MMNCg</strong></td>
<td>( R_O = \frac{R_{O_2}^{m,x,T} \times pO_2 \text{pkg}}{\left</td>
<td>K_{m,O_2} + pO_2 \text{pkg} / K_{m,CO_2} \right</td>
</tr>
</tbody>
</table>

* MM: Michaelis–Menten equation (Eq. (3)); MMNC: Michaelis–Menten equation with non-competitive inhibition by \( CO_2 \) (Eq. (4)); \( MM_k \) and MMNCg have constant \( K_{m,O_2} \).

### Table 2
Equations used to predict permeability as a function of temperature \((K)\) and activation energies for the films used in the experiments.

<table>
<thead>
<tr>
<th>Predicting equation ((\text{mmol cm cm}^{-2} \text{ h}^{-1} \text{ kPa}^{-1}))</th>
<th>( E_a ) (kJ mol (^{-1} ))</th>
<th>( R^2 )</th>
</tr>
</thead>
<tbody>
<tr>
<td>( P_{O_2} = 0.067 \times e^{-4423(T)} )</td>
<td>36.8</td>
<td>0.930</td>
</tr>
<tr>
<td>( P_{CO_2} = 0.118 \times e^{-4153(T)} )</td>
<td>34.5</td>
<td>0.923</td>
</tr>
</tbody>
</table>

* \( R^2 \) and \( E_a \) are the mean (±SE) of the experimental values measured at oxygen concentrations ranging from 18 kPa to the fermentation threshold (Fig. 3).

### Table 3
Experimental rates of \( O_2 \) uptake and \( CO_2 \) production, respiratory quotient \( \text{(RQ)} = \frac{\text{R}_{CO_2}}{\text{R}_{O_2}} \) and fermentation threshold \( (FT) \) for fresh-cut ‘Rocha’ pear stored at various temperatures.

<table>
<thead>
<tr>
<th>Temperature ((\text{°C}))</th>
<th>( R_O ) range ((\text{mmol kg}^{-1} \text{ h}^{-1}))</th>
<th>( R_{CO_2} ) range ((\text{mmol kg}^{-1} \text{ h}^{-1}))</th>
<th>( \text{RQ}^* )</th>
<th>( \text{FT} ) (kPa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.02–0.06</td>
<td>0.04–0.08</td>
<td>1.37 ± 0.02</td>
<td>0.75</td>
</tr>
<tr>
<td>5</td>
<td>0.03–0.18</td>
<td>0.07–0.22</td>
<td>1.38 ± 0.03</td>
<td>0.25</td>
</tr>
<tr>
<td>10</td>
<td>0.04–0.49</td>
<td>0.13–0.60</td>
<td>1.26 ± 0.02</td>
<td>0.75</td>
</tr>
<tr>
<td>15</td>
<td>0.09–0.95</td>
<td>0.23–1.19</td>
<td>1.18 ± 0.01</td>
<td>0.88</td>
</tr>
</tbody>
</table>

* \( \text{RQ}^* \) are the mean (±SE) of the experimental values measured at oxygen concentrations ranging from 18 kPa to the fermentation threshold (Fig. 3).
These rates, expressed as CO₂ production and obtained between 1.2 and 1.4 (Table 3), consistent with the usage of organic acids cut 'Rocha' pear stored at various temperatures ranged between (Gorny et al., 2000).

Air and in 2 kPa O₂ for other fresh-cut pear varieties at 0 or 10 0.1 and 18 kPa O₂, were similar to the range of value reported in MMNC a manner consistent with saturation kinetics (Fig. 2).

Effect of steady-state O₂ partial pressure and storage temperature on the Fig. 2. Arrhenius plot of gases permeabilities. (open symbols). Lines based on predicting equations presented in Table 2. Inset: Fig. 1. Effect of temperature on film permeability to O₂ (solid symbols) and CO₂ (open symbols). Lines based on predicting equations presented in Table 2. Inset: Arrhenius plot of gases permeabilities.

Respiration rates of fresh-cut 'Rocha' pear are shown in Table 3. The respiratory quotient (RQ) for aerobic respiration of fresh-fruits (Beaudry, 2000), due to absence of skin or less cuticle that may be used to determine the lower limit for aerobic respiration (Joles et al., 1994; Cameron et al., 1995). ‘Fermentation threshold’, the sudden increase in RQ or headspace ethanol concentration occurring when pO₂ dropped below a certain level, ranged between 0.3 and 0.9 kPa (Table 3).

Higher demand for O₂ at elevated temperatures fermentation occurred at higher pO₂ (Beaudry et al., 1992; Lakakul et al., 1999), as observed for 5 up to 15 °C (Table 3). Fermentation threshold of whole 'Bartlett' pears varied between 0.3 and 1.7 kPa, at temperatures between 0 and 25 °C (Kader et al., 1989). Temperature has a small influence on CO₂ and O₂ diffusion in 'Conference' pear as demonstrated by small activation energies obtained by Ho et al. (2006). Since the higher demand for O₂ at elevated temperatures cannot be compensated by diffusion, fermentation occurs at higher pO₂ (Beaudry et al., 1992).

Fresh-cut produce generally need lower O₂ levels than whole fruits (Beaudry, 2000), due to absence of skin or less cuticle that could restrict gas diffusion (Burg and Burg, 1965; Schotsmans et al., 2003; Ho et al., 2006). The minimum O₂ concentration tolerated by mature intact green 'Bartlett' pear at 25 °C is 1.6 to 1.7 kPa, and by 'Passe Crassane' pear cell cultures is 1.1 to 1.3 kPa (Boersig et al., 1988). The lower oxygen limit reported for sliced pear is 0.5 kPa at 0–5 °C (Beaudry, 2000; Gorny, 2001). Our results (Table 3) indicate that fresh-cut 'Rocha' pear should be packaged with pO₂ higher than 0.8 kPa at 0 °C, 0.3 kPa at 5 °C, 0.8 kPa at 10 °C and 0.9 kPa at 15 °C to avoid fermentation. Consistent with the fermentation thresholds reported herein for 'Rocha' pear, 2.5 kPa O₂ was considered acceptable for fresh-cut 'Conference' pear hold at 4 °C, but 0 kPa O₂ induced visual tissue damage (Soliva-Fortuny et al., 2007).

Table 4
Models predictions of \( R_{O_2}^{\text{max,T}} \), \( K_{n,O_2} \), and \( K_{\text{MCO}_2} \) as a function of temperature (K) and models corresponding accuracies.

<table>
<thead>
<tr>
<th>Model</th>
<th>( R_{O_2}^{\text{max,T}} ) (mmol kg⁻¹ h⁻¹)</th>
<th>( K_{n,O_2} ) (kPa)</th>
<th>( K_{\text{MCO}_2} ) (kPa)</th>
<th>R²</th>
<th>RMSE</th>
<th>( Q_{O_2} R_{O_2}^{\text{max,T}} )</th>
<th>( E_t R_{O_2}^{\text{max,T}} )</th>
<th>( E_t K_{n,O_2} )</th>
</tr>
</thead>
<tbody>
<tr>
<td>MM</td>
<td>( 2.40 \times 10^{-26} e^{0.205T} )</td>
<td>( 4.26 \times 10^{-10} e^{0.238T} )</td>
<td>-</td>
<td>0.963</td>
<td>0.043</td>
<td>7.8</td>
<td>134.2</td>
<td>155.8</td>
</tr>
<tr>
<td>MMx</td>
<td>( 3.84 \times 10^{-24} e^{0.187T} )</td>
<td>2.07</td>
<td>-</td>
<td>0.927</td>
<td>0.057</td>
<td>6.5</td>
<td>122.4</td>
<td>-</td>
</tr>
<tr>
<td>MMNC</td>
<td>( 1.38 \times 10^{-24} e^{0.191T} )</td>
<td>( 1.28 \times 10^{-10} e^{0.242T} )</td>
<td>41.99 (77%)</td>
<td>0.966</td>
<td>0.039</td>
<td>6.8</td>
<td>125.9</td>
<td>158.4</td>
</tr>
<tr>
<td>MMNCx</td>
<td>( 2.43 \times 10^{-19} e^{0.140T} )</td>
<td>1.27</td>
<td>-</td>
<td>14.26 (42%)</td>
<td>0.948</td>
<td>0.048</td>
<td>4.4</td>
<td>97.5</td>
</tr>
</tbody>
</table>

a) MM – Michaelis–Menten model; MMNC – Michaelis–Menten model with non-competitive inhibition by CO₂.

Values within parenthesis indicate parameter accuracy (ratio of standard error to estimated value).

Activation energies \( (E_t) \) of respiratory parameters were calculated using Arrhenius plots (Eqs. (5) and (6)).

Gas equilibrium concentration inside packages was reached by 4–5 days at 15 °C, 6–8 days at 10 °C, 7–10 days at 5 °C, and 10–11 days at 0 °C. The oxygen uptake rate increased with pO₂ pkg in a manner consistent with saturation kinetics (Fig. 2).

Respiration rates of fresh-cut 'Rocha' pear are shown in Table 3. These rates, expressed as CO₂ production and obtained between 0.1 and 18 kPa O₂, were similar to the range of value reported in air and in 2 kPa O₂ for other fresh-cut pear varieties at 0 or 10 °C (Gorny et al., 2000).

The respiratory quotient (RQ) for aerobic respiration of fresh-cut 'Rocha' pear stored at various temperatures ranged between 1.2 and 1.4 (Table 3), consistent with the usage of organic acids as major respiratory substrates (Fonseca et al., 2002). RQ values reported in the literature for whole 'Bartlett' pear varied from 0.8 at the initial climacteric to 1.4 at fully ripe stage (Biale, 1950). RQ values pertaining to intact 'Conference' pear lie between 0.7 and 0.8 (de Wild and Peppelenbos, 2001; Lammertyn et al., 2003). The reported RQ for fresh-cut 'Conference' pear are 1.7 after 1 day following processing (Soliva-Fortuny et al., 2007) are likely an analytical artefact or else a clear sign of fermentation.

Respiratory quotients and ethanol levels responded similarly to steady-state oxygen partial pressure (pO₂) (Fig. 3), and either variable may be used to determine the lower limit for aerobic respiration (Joles et al., 1994; Cameron et al., 1995). 'Fermentation threshold', the sudden increase in RQ or headspace ethanol concentration occurring when pO₂ dropped below a certain level, ranged between 0.3 and 0.9 kPa (Table 3). At elevated temperatures fermentation occurred at higher pO₂ (Beaudry et al., 1992; Lakakul et al., 1999), as observed for 5 up to 15 °C (Table 3). Fermentation threshold of whole 'Bartlett' pears varied between 0.3 and 1.7 kPa, at temperatures between 0 and 25 °C (Kader et al., 1989). Temperature has a small influence on CO₂ and O₂ diffusion in 'Conference' pear as demonstrated by small activation energies obtained by Ho et al. (2006). Since the higher demand for O₂ at elevated temperatures cannot be compensated by diffusion, fermentation occurs at higher pO₂ (Beaudry et al., 1992).

3.3. Modelling the effect of oxygen and temperature on respiration rate

The respiratory parameters \( R_{O_2}^{\text{max,T}} \), \( K_{n,O_2} \) and \( K_{\text{MCO}_2} \) were estimated individually at each temperature, and their temperature dependence subsequently studied with the models presented in Table 1.

Exponential functions gave the best fit of \( R_{O_2}^{\text{max,T}} \) in both experiments using the estimates of each temperature \( (R² > 0.98, \text{not shown}) \). The temperature dependence of \( R_{O_2}^{\text{max,T}} \) has been analysed according to the Arrhenius’ law (Hertog et al., 1998; Jacxsens et al., 1999), as observed for 5 up to 15 °C (Table 3)
ported for other fresh-cut pear varieties (Gorny et al., 2000). Similarly, \( Q_{10} \) of fresh-cut vegetables may range from 2.8 to 8.0 (Jacxsens et al., 2000).

The activation energy of common intact produce range between 50 and 89 kJ mol\(^{-1} CO_2\) (Exama et al., 1993; Fonseca et al., 2002), and can be as high as 135.9 kJ mol\(^{-1} CO_2\) in fresh-cut vegetables (Jacxsens et al., 2000).

Hertog et al. (1998) examined the temperature dependence of apparent \( K_m \) according to the Arrhenius’ law and concluded that this variable can be treated as a constant. Non-competitive models published for intact ‘Conference’ pear (de Wild et al., 1999; Lammertyn et al., 2001, 2003; Ho et al., 2006, 2008) did not make \( K_m \) temperature-dependent. A constant \( K_m \) was obtained for ten fresh-cut vegetables (Jacxsens et al., 2000) and packaged raspberry fruit for the temperature range from 0 to 20°C (Joles et al., 1994), while for blueberry a temperature effect on \( K_m \) was found (Song et al., 1992; Cameron et al., 1994). Ho et al. (2006) indicated for cortex tissue of ‘Conference’ pear a \( K_m \) of 1.16 kPa, similar to \( K_m \) found for fresh-cut ‘Rocha’ pear (Table 4). Cytochrome c oxidase, which is believed to be the rate determining enzyme in the respiratory pathways, has a high affinity to oxygen (\( K_m = 0.10–0.15 \) kPa) and its activity does not change during hypoxia, when \( pO_2 < 0.25 \) kPa (Nanos et al., 1994). However, diffusion limitations can lead to a much higher value of \( K_m \) in intact pear (6.2 kPa) than that in protoplasts (\( \approx 0.007 \) kPa), and even higher than in the case of mitochondrial respiration (Lammertyn et al., 2001). Differences in the reported values of \( K_m \) can be accounted for by the joint effect of respiration at the cellular level and gas diffusion through the pear tissue and skin (Lammertyn et al., 2003).

The models in which there was a temperature dependence of \( K_{m,O_2} \) were only marginally better than those where \( K_{m,O_2} \) was constant (Table 4). Michaelis–Menten and non-competitive Michaelis–Menten models were able to explain more than 93% of the total variability of the data and either model accurately predicted the experimental results (RMSE < 0.06). However, the non-competitive Michaelis–Menten model provided an estimate of \( K_{m,CO_2} \) with low accuracy (Table 4). In addition, the \( K_{m,CO_2} \) values were very high (14.3–42.0 kPa), indicating a low inhibitory effect of \( CO_2 \) on \( O_2 \) consumption (Ho et al., 2008).

To discriminate between different types of inhibition it is necessary to know \( O_2 \) consumption at two or more different \( CO_2 \) concentrations and at three \( O_2 \) concentrations (Peppelenbos et al., 1996). Since our data did not satisfy these requirements, it is not possible to assume an influence of \( CO_2 \) on \( O_2 \) consumption. However, given the accuracy of the simpler Michaelis–Menten model and the indications of minimal \( CO_2 \) influence, the use of simpler model is preferred in this case (Fig. 2).

### 3.4. Respiratory responses to low oxygen and implications for MAP

Respiratory behavior as a function of oxygen concentration provides a basis to deduce the potential benefits of using MAP technologies in a given commodity. Low oxygen atmospheres could be useful if a sizeable reduction in respiration rate (e.g. 50%) can be reached without the induction of fermentation (Beaudry, 2000). In cases where \( K_{m,O_2} \) is much higher than fermentation threshold, a reduction in \( pO_2 \) slows down metabolic activities without an increase in the RQ and fermentative compounds. The range of \( pO_2 \) between \( K_{m,O_2} \) and fermentation threshold is termed ‘safe working atmosphere’ (Beaudry, 2000). On the other hand, if \( K_{m,O_2} \) is lower than the fermentation threshold any attempt to reduce \( O_2 \) could

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**Fig. 3.** Respiratory quotient (A) and headspace ethanol (B) as a function of steady-state \( O_2 \) of packaged fresh-cut ‘Rocha’ pear.
lead to fermentation. Fresh-cut ‘Rocha’ pear had a very narrow (0.01–1.71 kPa) safe working atmosphere at the temperatures tested (Table 5). Low oxygen atmospheres did not offer any reduction in metabolic activity without the danger of inducing anaerobiosis, especially between 0 and 10 °C, temperatures normally found during storage and marketing. These data are consistent to the findings compiled by Gorny (2001), who also reported a poor efficacy of modified atmosphere for fresh-cut pear at 0–5 °C.

In conclusion, respiration of fresh-cut ‘Rocha’ pear as function of oxygen concentration and temperature can be accurately predicted through Michaelis–Menten kinetics without inhibition by CO2. These data provide a foundation whereby sealed packages can be designed to avoid fermentative metabolism of fresh-cut ‘Rocha’ pear. However, given the respiratory kinetics of fresh-cut ‘Rocha’ pear, it is anticipated that low oxygen packaging is of little or no benefit in slowing down metabolism in this convenience produce.

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References


Table 5

Michaelis–Menten model (MM) respiratory parameters for fresh-cut ‘Rocha’ pear stored at various temperatures.

<table>
<thead>
<tr>
<th>Temperature (°C)</th>
<th>Fermentation threshold (kPa)</th>
<th>( K_m \times V_o ) (kPa)</th>
<th>Safe working atmosphere (kPa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.75</td>
<td>0.07</td>
<td>No</td>
</tr>
<tr>
<td>5</td>
<td>0.25</td>
<td>0.24</td>
<td>0.01</td>
</tr>
<tr>
<td>10</td>
<td>0.75</td>
<td>0.79</td>
<td>0.04</td>
</tr>
<tr>
<td>15</td>
<td>0.88</td>
<td>2.59</td>
<td>1.71</td>
</tr>
</tbody>
</table>


