The Anaerobic Compensation Point for Fresh-cut Watermelon and Implications for Postprocess Handling

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Abstract. The influence of temperature and O2 concentration on respiration and shelf life of fresh-cut watermelon was investigated. Product stored at selected temperatures from 1 to 30 °C showed increasing respiration and reduced shelf life with increasing temperature. Oxygen depletion and CO2 evolution were measured using a closed system method and rates of O2 consumption and CO2 production were computed. A mathematical model found to predict the CO2 production as function of temperature and O2 showed an elevated rate of CO2 production at about 14% O2 or lower. A modified atmosphere trial that compared product stored at 7 to 9 °C in air with product at either 14% or 8% O2 revealed increased respiration in the latter treatments, suggesting a relatively high anaerobic compensation point (ACP) > 14% O2. Our results suggest limited applicability of modified atmosphere packaging (MAP) for this product. Fresh-cut watermelon had extended shelf life and reduced respiration rate when stored at 1 to 3 °C and in > 14% O2 atmospheres.

The consumption of fresh-cut watermelon (Citrullus lanatus (Thunb. Matsum. and Nakai)) has increased at a rate of 20% to 30% annually, to the extent that ∼25% of all watermelon produced in U.S. is cut in some fashion for retail sale. Most of this processing is done in the supermarket due to the product’s short shelf life (NWFPA, 1998). Previously we determined that shock and vibration stress encountered during transportation is a major problem that must be addressed by regional processors. Packaging that diminishes cube-to-cube friction appeared to be a promising solution (Fonseca et al., 1999).

Refrigeration is the primary method for reducing the respiration rate and quality deterioration of fresh fruits and vegetables (Cameron et al., 1994; King and Bolin 1999). Intact watermelon stored below 10 °C may suffer chilling injury, which may be manifested as water-soaked areas in the pulp (Risse and Maynard 1990). The respiration rate of intact watermelon ranges between 10 and 20 mL CO2 kg−1 h−1 at 18 °C (Elkashif et al., 1989). Watada et al. (1996) and Gorny (1997) reviewed respiration rates and optimum temperatures for storage of fresh-cut products but their reports lacked information on fresh-cut watermelon. The respiration rate of fresh fruits and vegetables depends on such factors as the composition of the surrounding gas, the moisture content of the tissue, wounding, morphology, maturity, and temperature of the product (Kays, 1991; Talasila et al., 1992). The effectiveness of temperature management on extending the shelf life of some fresh produce can be enhanced by using modified atmosphere packaging (MAP) (Kader et al., 1989). Using MAP techniques, aerobic respiration may be reduced by decreasing the available O2, but only to a critical level below which respiration increases due to anaerobic respiration (Knee, 1980). This level was first mentioned as the extinction point (EP) (Platenius, 1942). Since then, four alternative concepts have been proposed, each one determined by a slightly different method. The anaerobic compensation point (ACP) is the O2 concentration at which CO2 production is minimal (Boersig et al., 1988). The respiratory quotient breakpoint (RQB) is defined as the partial pressure of O2 at which the respiratory quotient increases with reduced O2 content (Beaudry et al., 1992). The maintenance oxygen concentration (MOC) is the oxygen concentration at which ATP production is minimal for maintaining cell viability (Peppelenbos and Rabbinge, 1996). Finally, the fermentation threshold (FT) combines the RQB with the O2 level above which fermentation is extinguished (EP) (Yearsley, et al., 1996).

Generally, in comparison to intact fruit, the fresh-cut products can be stored at lower O2 levels such as 1% to 3% (Gorny, 1997; Kays, 1991) in part because they have less cuticle or skin to restrict gas diffusion (Watada, 1999). However, there are several exceptions: orange slices, for example, undergo anaerobic respiration below 14% O2 (Gorny, 1997), and sweetpotatoes may shift to anaerobic conditions at external oxygen concentrations below 7% (Chang and Kays, 1981). Little has been reported addressing the effect of O2 and CO2 on fresh-cut watermelon physiology. Cartaxo et al. (1997) observed unsatisfactory quality of watermelon cubes stored in 3% O2 atmospheres, but higher O2 concentrations were not tested in that study. Perkins-Veazie et al. (1998) measured respiration of watermelon slices (flesh and rind) stored in jars at 10 °C, observing an increase in rate from storage day 1 to 5; these results, however, were not associated with any physiological or environmental factor.

Several models have been used to predict aerobic respiration of produce under closed systems. A linear model (Hayakawa et al., 1975) and an enzymatic model (Lee et al., 1991), both based on O2 concentration as an inhibitor of fermentative CO2 production, are noted. Another model uses the ATP concentration as an inhibitor of fermentative CO2 production (Peppelenbos et al., 1996). However, our interest was to find a model that could predict the CO2 production (KCO2) as a function of two factors—temperature and O2. Such a model could be useful to define the ACP and thus predict the onset of anaerobic respiration in products stored at different temperatures.

The objective of this work was to determine the influence of temperature and oxygen on the respiration and shelf life of fresh-cut watermelon. We provide evidence for defining the anaerobic compensation point and discuss implications for postprocess handling.

Materials and Methods

Fruit source. Seeded ‘Royal Sweet’ watermelon was used for the gas sampling and storage temperature components of the study and seedless cultivar 2524 was used to test the effects of initial O2 concentrations. The fruit originated from Mexico and were procured from a local Ingles Supermarket in Clemson, S.C.

Processing protocol and storage temperatures. Intact watermelon fruit were sliced in 1 min in a 200 mg·L−1 chlorine solution. The peel was removed with a sharp knife and pulp cubes (>2.5 cm each dimension) were cut.

Shelf life determination. Cubes (250 g) were placed into a polypropylene (PP) cup-and-lid packages (340 g capacity, Fabri-Kal Corporation, Kalamazoo, Mich.), currently used commercially for fresh-cut watermelon. The packages were filled with air. To avoid contamination, the packages were previously irradiated with UV light (250 nm) for 3 min (420 mJ·cm−2) using a bulb-lamp (Germicidal lamp, American Ultraviolet Co., Beaufort, S.C.) placed 15 cm above the containers. The treatments consisted of experimental units of two packages of product with five replicates placed at six different storage temperatures: 1, 3, 7, 11, 15, and 30 °C. Each day, the stored product was evaluated for color fading, dark-
ening, off-odor, and overall visual quality by a group of 10 untrained panelists simulating typical retail customers. The quality of each watermelon cube was rated according to the following criteria: 10 = excellent; 7 = lowest point of salability; 5 = definite consumer rejection point; and 0 = extremely poor. The maximum shelf life of cubes in each container was defined as the period of time before the overall quality fell below level 7 according to its hedonic scale. Aerobic total count (ATC), using Petrifilm aerobic count plates (3M Co., St Paul, Minn.), was monitored on additional samples stored at temperatures 7 °C and below, to estimate any effect of microbial population on quality judging and respiration results. A sample of 25 g of the product was prepared according to the film manufacturer’s recommendations. Dilutions from 10 to 10 per g sample were incubated at 30 °C for 72 h. Two cubes from each of three containers were sampled on the second, fifth and seventh day of the trial.

**Respiration rate in a closed system.** Watermelon cubes (200 g) processed as previously described were placed into 480-mL glass jars with tight-fitting lids equipped with a rubber sampling port. The jars had been cleaned with 500 mg·L⁻¹ chlorine and irradiated with UV light as described. For each treatment, three fruit were used, each to fill five jars (replicates). The jars were stored at five different temperatures: 3, 7, 11, 15, and 30 °C. A closed system method was used for the measurement of O₂ and CO₂ concentrations (Yam and Lee, 1995; Zhu et al., 2001), with jars being sealed until the CO₂ level reached above 20%, as this was reported to be a safe level for fresh-cut watermelon (Cartaxo et al., 1997). At least twice per day, 1-mL samples were withdrawn from the headspace of each jar. Samples were analyzed for O₂ and CO₂ with a Tracor 540 capillary gas chromatograph fitted with a double column Altech CTR-1 and thermal conductivity detector. The first sample was taken within the first hour after cutting. Respiration rate, as CO₂ production (KCO₂) in mL·kg⁻¹·h⁻¹ at a given O₂ level and CO₂ level, was then calculated according to Kays (1991):

\[
KCO_2 = \left( \% CO_2 \times 10 \right) \frac{V}{W} \left( \frac{t}{h} \right)
\]

where \( \% CO_2 \) = change in carbon dioxide concentration (\%), \( V \) = free space volume of container (L), \( W \) = product weight (kg), and \( t \) = time container was closed (h). The trial was repeated 14 d later.

**Results and Discussion**

**Shelf life determination.** The shelf life of fresh-cut watermelon decreased with increasing temperatures (Fig. 1). The cubes stored at 1 or 3 °C had significantly longer shelf life (>10 d) than cubes stored at above 7 °C (<6 d). Moreover, the predicted data yielded by a nonlinear regression (\( r^2 = 0.96 \)) revealed a maximum inflection between 4 and 6 °C, supporting the importance of storing this product below this range. Microbial population, which was monitored for temperatures 7 °C and below, increased from \( 3 \log_{10} \text{CFU/g} \) on the second day to near \( 6 \log_{10} \text{CFU/g} \) on the seventh day. The results comparing each temperature, however, were statistically non significant (data not shown). Within this temperature range, the microbial count did not appear to be affecting the judging of quality of the product. Quality parameters, including color fading and darkening, off-odor, and juice leakage also confirmed the benefits of storage at the lower temperatures (data not shown). In all cases, rejection of cubes was associated with unpleasant off-odor. Occurrence of random microbial growth was observed in the 1 to 7 °C range. In other studies, this had been associated with the section of the fruit from which cubes were cut (Fonseca, 2000). Although the rigid plastic containers used in retail stores and in the previous experiments are not designed for MAP due to leakage, we observed in a separate study that the headspace oxygen in some filled containers did decrease to as low as 15.8% \( O_2 \) after 5 d of storage at 2.8 °C (Fonseca, 2000). Due to variability in container leakage rates the internal atmospheric modification was not consistent and these containers were not considered to be suitable for MAP studies.

**Respiration rate in a closed system.** The temporal pattern of \( CO_2 \) production of fresh-cut watermelon obtained using Eq. [1] was similar to those of other fresh-cut products such as broccoli florets (Rushing, 1990) and cut iceberg lettuce (Smyth et al., 1998). The rate was initially higher immediately after cutting, ranging between 25 to 50 mL \( CO_2 \) per kg⁻¹·h⁻¹, an apparent result of wound metabolism, then dropping to a plateau of about one-eighth the initial level in <10 h when stored at temperatures below 15 °C (Fig. 2). The rate at this stable stage was higher at higher temperatures. An increase in \( CO_2 \) evolution was observed at all temperatures after a period of time, but the delay in the increased \( CO_2 \) was longer and the increased level was lower at lower temperatures.

Considerable variability was noted in respiration rate during the first 6 h after processing (Fig. 2). This likely was due to the stress caused by cutting the product (Smyth et al., 1998). The resulting respiration data had a quadratic shape. Thus, we fitted the respiration data initiating with the 6-h sample into the following quadratic model:

\[
KCO_2 = b + m_1 (O_2) + m_2 (T) + m_3 (O_2) T + m_4 (T)^2 \quad [2]
\]

where \( T \) = temperature (°C). The data adequately fit the proposed quadratic model (\( r^2 = 0.94 \)). The constant \( b \) and the parameters \( m_1 \), \( m_2 \), \( m_3 \), and \( m_4 \) in the Eq. [2] were \( KCO_2 = 7.507 \pm 0.871 \text{ (O}_2\text{) } - 0.056 \text{ (T) } + 0.026 \text{ (O}_2\text{T) } + 0.028 \text{ (T)}^2 \). Excluding data from the first 5 h, the model allowed the prediction of \( CO_2 \) production as a function of \( O_2 \) and temperature, indicating a distinct increase in respiration rate at a relatively high \( O_2 \) concentration. The model predicted the minimal \( KCO_2 \) at exactly 16.7% \( O_2 \) for all temperatures evaluated. The measured increase
in respiration rate, however, was not substantial until ≈14% (Fig 3). A similar ACP, which has been suggested as a minimum for the storage of orange slices at 0 to 5 °C, was accompanied by an increase in the evolution of ethanol and acetaldehyde (Gorny, 1997).

Respiration rate with different initial levels of Oxygen. The trial we conducted with different initial O2 levels in barrier pouches at 7 to 9 °C revealed an increased CO2 production and O2 consumption for the treatments having either 14% or 8% O2 (Fig 4). Although the air treatment had a higher respiration rate for the first sampling (2 h), this dropped to a stable level as was observed in previous experiments, maintaining a level of 1/5 to 1/3 the rate of the O2 modified treatments.

The CO2 accumulation in the pouches with air averaged <2% after 12 h, while pouches flushed with 8% or 14% O2 accumulated ≈4% CO2. This difference was maintained or increased for 74h, the point at which the O2 was depleted to <1% for the reduced O2 treatments (Fig. 5). These results suggested a consistent tendency of the watermelon cubes to accelerate CO2 production at a relatively high O2 level, e.g., 14%, verifying the hypothesis that ACP is above 14% O2 suggested by the data from the prediction model. Several alcohols and aldehydes have been found as major volatile flavor and odor components in watermelons recently harvested (Kemp, 1975; Yahima et al., 1985). The concentration of these volatiles may change at a critical O2 level (Beaudry, 1999), which could partially explain the off-odor at the end of the shelf life of the product in the storage temperature trial.

When using data from the product in barrier pouches, the model approximated the respiration of the air treatment, but underestimated the 8% and 14% initial O2 level treatments when the O2 level was still above 1% (data not shown). This was expected as the model was based on data yielded from a closed system that had air at the starting point, and because increased CO2 may suppress respiration (Watkins, 2000). A study of increased CO2 with a nonmodified level of O2 may reveal whether such a treatment further extends shelf life of fresh-cut watermelon. Cartaxo et al. (1997) previously found positive results with high levels of CO2, which support this hypothesis.

The most important point with developing closed system models, as presented here, is to provide a pattern of respiration of a fresh-cut product at different temperatures and changing O2 or CO2 (if calculating respiration in terms of O2 consumption), and estimate the ACP level as described by Boersig et al. (1988). Accuracy should still be a concern, since our empirical model indicates a fixed lower O2 limit for the 0 to 30 °C range. Beaudry et al. (1992) revealed that the RQB of blueberries increased with increasing temperature. The lower O2 limit also has been reported to increase with increasing temperature in whole apples (Gran and Beaudry, 1993), and raspberries (Joles et al., 1994). Despite this, Yearsley et al. (1997) reported ACP based on internal lower O2 limit and concluded that the effect of temperature on the ACP of whole apples stored between 0 and 28 °C was not significant. Little has been reported on the effect of temperature on the lower O2 limit of fresh-cut products but it appears that the effect is not as marked as with whole products, perhaps due to higher O2 diffusion in fresh cuts. Lakalul et al. (1999) observed an increase of the FT with increasing temperature in apple slices but the increase was only 0.2 kPa in the range of 0 to 15 °C. Our regression estimated 94% of the respiration data and predicted a single ACP that was verified at 7 to 9 °C. Experiments evaluating ACP at higher temperatures are needed to represent other temperatures also encountered during the retailing and consumer’s handling.

Another consideration is that analyses were based on watermelon from a distant production region (Mexico). Transportation stress might have played a role in the observed ACP, as it has been reported that ACP shifts toward higher concentrations with the physiological aging of intact fruits (Boersig et al., 1988).

Since the model depends on changing gases, as in any closed or static system, a subsequent step must involve the use of permanent controlled atmospheres (Yang and Chinnan, 1988).
The addition of CO₂-salt forming bases such as KOH (Cameron et al., 1989) or the testing of different starting gas levels may also give additional insight. In this study, whether with an initial nonmodified atmosphere or with increased CO₂ concentration in the headspace, the product respired more when exposed to an O₂ level of 14% or lower than for air, suggesting limited applicability of MAP systems for extending the shelf life of this product. Our results should be useful for the design of postprocessing handling protocols for fresh-cut watermelon, particularly in selecting storage temperatures and packaging materials.

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