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'ROCHA' PEARS STORED UNDER CONTROLLED ATMOSPHERE WITH ULTRA-LOW AND LOW O_2 ASSOCIATED WITH DIFFERENT CO_2 LEVELS[1]

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ABSTRACT – The storage of 'Rocha' pears under controlled atmosphere (CA) preserves fruit quality for long periods. However, inadequate CA conditions might impair ripening and lead to flesh browning. This research was carried out to assess the effects of CA with ultra-low (*ULO*), and low O_2 (*LO*) associated with different CO_2 levels on ripening and occurrence of flesh browning in 'Rocha' pears. Treatments evaluated were: pO_2 = 0.5 kPa (*ULO*) and pCO_2 < 0.03 kPa; pO_2 = 1.0 kPa (*LO*) and pCO_2 < 0.03 kPa; pO_2 = 1.0 kPa and pCO_2 = 1.0 kPa; pO_2 = 1.0 kPa and pCO_2 = 2.0 kPa; and pO_2 = 1.0 kPa and pCO_2 = 3.0 kPa. A completely randomized experimental design was used, with four replicates. Fruits were harvested in Vacaria, RS, and stored under five CA conditions during 270 days (-0.5±0.1 ºC and relative humidity of 96±2%). Fruits were assessed after CA storage for respiratory rate, ethylene production, skin color, flesh firmness, texture, titratable acidity (TA), soluble solids content (SSC), sensory attributes, flesh browning incidence and severity and flesh color. Fruits stored under *LO* with pCO_2 < 0.03 kPa had higher flesh firmness and more yellow skin color than fruits stored under other CA conditions. Sensory attributes, SS and TA of fruits were not affected by CA conditions. Fruit stored under *LO* with pCO_2 = 3.0 kPa had the highest incidence (48%) and severity of flesh browning. The pO_2 = 0.5 kPa and pCO_2 < 0.03 kPa and pO_2 = 1.0 kPa and pCO_2 = 1.0 kPa conditions are the most suitable for the CA storage of 'Rocha' pears.
Index terms: *Pyrus communis*, flesh browning, physiological disorder, ripening, texture.

PERAS 'ROCHA' ARMAZENADAS EM ATMOSFERA CONTROLADA COM ULTRABAIXO O_2 E COM BAIXO O_2 ASSOCIADO A DIFERENTES TEORES DE CO_2

RESUMO - O armazenamento de peras 'Rocha' em atmosfera controlada (AC) mantém a qualidade dos frutos por períodos prolongados. Contudo, condições inadequadas de AC podem favorecer a perda da capacidade de amadurecimento e a ocorrência de escurecimento de polpa nos frutos. Este trabalho teve como objetivo avaliar os efeitos de AC com ultrabaixo O_2 (*UBO*) e com baixo O_2 (*BO*), associado a diferentes teores de CO_2 sobre o amadurecimento e a ocorrência de escurecimento de polpa em peras Rocha. Os tratamentos avaliados foram: pO_2 = 0,5 kPa (*UBO*) e pCO_2 < 0,03 kPa; pO_2 = 1,0 kPa (*BO*) e pCO_2 < 0,03 kPa; pO_2 = 1,0 kPa e pCO_2 = 1,0 kPa; pO_2 = 1,0 kPa e pCO_2 = 2,0 kPa; e pO_2 = 1,0 kPa e pCO_2 = 3,0 kPa. O delineamento experimental utilizado foi o inteiramente casualizado, com quatro repetições. Os frutos foram colhidos em Vacaria-RS, e armazenados nas cinco diferentes condições de AC durante 270 dias (-0,5±0,1 ºC e umidade relativa de 96±2%). Os frutos foram avaliados após o armazenamento em AC, em relação às taxas respiratória e de produção de etileno, cor da casca, firmeza de polpa, atributos de textura, acidez titulável (AT), teor de sólidos solúveis (SS), análise sensorial, incidência e severidade de escurecimento de polpa, e cor da polpa. Os frutos armazenados em *BO* com pCO_2 < 0,03 kPa apresentaram menor perda de firmeza de polpa, além de uma coloração da casca mais amarela. Não houve diferença entre as condições de armazenamento para a análise sensorial, teor de SS e AT. A condição de *BO* com pCO_2 = 3,0 kPa apresentou maior incidência e severidade de escurecimento de polpa. As condições de pO_2 = 0,5 kPa e pCO_2 < 0,03 kPa) e pO_2 = 1,0 kPa e pCO_2 = 1,0 kPa são as mais indicadas para o armazenamento de peras 'Rocha'.
Termos para indexação: *Pyrus communis,* escurecimento de polpa, distúrbio fisiológico, amadurecimento, textura.

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INTRODUCTION

Currently, about 140 thousand tons of pears (*Pyrus communis*) are imported each year in order to meet the demand of the consumer market, representing the highest percentage of total fresh fruits imported by Brazil (FAORO AND ORTH, 2010). Among the numerous difficulties for the crop development, the lack of studies and technologies aimed at the storage of pears produced in the country stands out.

'Rocha' pear is a European cultivar adapted to the growing conditions of some regions of southern Brazil (PETINELI et al., 2014). This cultivar has good storage capacity, tolerating up to nine months under controlled atmosphere (CA) storage (GAGO et al., 2013; MARTIN et al., 2015).

The reduction of partial O_2 pressures (pO_2) and the increase of partial CO_2 pressures (pCO_2), during CA storage decrease the respiratory activity and the synthesis and action of ethylene, delaying the ripening and senescence of fruits (WEBER et al., 2013a). However, pears stored under inadequate CA conditions may develop a number of undesirable characteristics, such as loss of ripening ability and incidence of flesh browning (PEDRESCHI et al., 2008; MARTIN et al., 2015).

Flesh browning is a physiological disorder characterized by the development of brown spots and cavities inside the flesh, which can cause great loss in the commercial value of fruits (PEDRESCHI et al., 2009). This disorder can be due to the difficulty of CO_2 diffusion inside the fruit, which presents higher partial pressures inside the flesh and smaller ones in layers closer to the fruit skin (PEDRESCHI et al., 2008). Improper CA conditions, with high CO_2 contents inside the chamber, can lead to an even greater accumulation of this gas in the flesh, leading to an increase in the damage extent (PEDRESCHI et al., 2008). On the other hand, this damage can also occur due to the reduction in partial O_2 pressures in the storage environment (FRANCK et al., 2007).

For 'Rocha' pears produced in western Portugal and harvested with flesh firmness of 60 N, Cavaco et al. (2009) used CA condition with pO_2 = 2.0 kPa and pCO_2 = 0.5 kPa at temperature of -0.5 °C and relative humidity (RH) of 94-96% to store fruits with quality for a period of eight months. However, these conditions are different from those found by Martin et al. (2015) for 'Rocha' pears produced in southern Brazil and harvested with flesh firmness of 67 N, where CA conditions of pO_2 = 1.0 kPa with pCO_2 = 1.0 kPa and pO_2 = 1.0 kPa with pCO_2 = 2.0 kPa (-0.5 °C and 96% RH) maintained fruit quality after eight and a half months of storage, allowing normal ripening without causing the incidence of flesh browning.

Ultra-low O_2 storage (*ULO*) is a technique complementary to CA that aims to minimize losses during the post-harvest period by reducing pO_2 to values below 1.0 kPa (BRACKMANN et al., 2013). Storage under such conditions causes a ripening delay, reducing the loss of acids and sugars during respiration, as well as the ethylene biosynthesis rate in fruits (WEBER et al., 2013b). However, when *ULO* storage is done beyond the tolerable period, or under excessively low partial O_2 pressures, some detrimental effects may occur, such as increased incidence of flesh browning (PEDRESCHI et al., 2009). Until now, the effect of *ULO* on the storage of pears produced in Brazil remains unknown.

Depending on the orchard location, pears may be more or less susceptible to flesh browning, which influences the ability and ideal conditions for fruit storage (FRANCK et al., 2007). Thus, although the conditions used in the storage of 'Rocha' pears produced in the western region of Portugal are already well elucidated, it is necessary to determine the partial pressures of gases suitable for the storage of fruits produced in the southern region of Brazil.

The aim of this work was to evaluate the effects of CA with ultra low O_2 (*ULO*) and with low O_2 (*LO*) associated to different CO_2 levels on ripening and occurrence of flesh browning in 'Rocha' pears.

MATERIALS AND METHODS

The experiment was conducted in 2012/2013 season with 'Rocha' pears from a commercial orchard located in Vacaria, RS (28°30'39 "S and 50°55'47"W and 960m altitude), using six-year-old plants grafted on 'BA29' quince (*Cydonia oblonga*) and spaced 1.5m in line and 4.0m between lines. After harvest, fruits showing rot, lesions, defects or low caliber were excluded.

Fruits were stored under temperature of -0.5 ± 0.1°C and RH of 96 ± 2% for 270 days in experimental mini-chambers with capacity of 233 L. The CA conditions evaluated were ultra-low O_2 (*ULO*) and low O_2 (*LO*) associated with different CO_2 proportions. Treatments were: pO_2 = 0.5 kPa (*ULO*) and pCO_2 < 0.03 kPa; pO_2 = 1.0 kPa (*LO*) and pCO_2 < 0.03 kPa; pO_2 = 1.0 kPa and pCO_2 = 1.0 kPa; pO_2 = 1.0 kPa and pCO_2 = 2.0 kPa; and pO_2 = 1.0 kPa and pCO_2 = 3.0 kPa. The experimental design was completely randomized, with four replicates and an experimental unit consisting of 30 fruits.

CA conditions were established by diluting

O_2 in the storage environment with N_2 injection from a gas generator using the "Pressure Swing Adsorption" (PSA) principle and subsequent CO_2 injection from high pressure cylinders up to reaching the pre-established content in treatments with pCO_2> 0.03 kPa. The maintenance of the desired partial pressures of gases, which varied due to fruit respiration, was daily performed using automatic gas control equipment (Kronenberger / Climasul, Caxias do Sul, Brazil). When CO_2 and O_2 contents were not adequate, the equipment corrected the partial pressures up to the pre-established conditions in treatments. The O_2 consumed by respiration was replaced by the injection of atmospheric air into the mini-chambers, and excess CO_2 was absorbed by a 40% potassium hydroxide solution, through which air from the storage environment was circulated. In treatments with reduced pCO_2 (pO_2 = 0.5 kPa and pCO_2 <0.03 kPa, pO_2 = 1.0 kPa (*LO*) and pCO_2 <0.03 kPa), pCO_2 was maintained by placing hydrated lime inside micro-chambers for continued CO_2 elimination in the storage environment.

At the end of storage, fruits were evaluated after zero, three and six days of shelf life (20 ± 5ºC and RH of 63 ± 2%) in relation to skin color, respiratory rate and ethylene production. After six days in the environmental condition, they were also evaluated in relation to flesh firmness, texture, titratable acidity (TA), soluble solids content (SSC), sensorial analysis, incidence and severity of flesh browning and flesh color.

Respiratory rate (nmol of CO_2 kg^{-1} s^{-1}) and ethylene production (nmol C_2H_4 kg^{-1} s^{-1}) were quantified by gas chromatography. Ten fruits of each replicate were packed in 4.1 L containers, hermetically sealed. Respiratory rate and ethylene production were obtained by the difference in the CO_2 and C_2H_4 content, respectively, inside the container, immediately after its closure and after half an hour. After this period, using a 1.0 mL plastic syringe, two samples were collected from the free space of these containers, which were injected into a Varian® gas chromatograph, model CP-3800 (Palo Alto, USA), equipped with a 3 m long Porapak N® column (80-100 mesh), methanator and flame ionization detector. The column, detector, methanator and injector temperatures were 70, 250, 380 and 130°C, respectively. Nitrogen, hydrogen and synthetic air fluxes used were 70; 30 and 300 mL min^{-1}, respectively.

Skin color was evaluated in terms of 'hue' ($h°$) angle values, using a CR 400 colorimeter (Konica Minolta®, Tokyo, Japan). The h^o values present the following correspondences regarding the colors of the vegetal tissue surface: 0º / red, 90º / yellow, 180º / green and 270º / blue. Readings were performed at two opposite points of the equatorial region of fruits.

Flesh firmness (N) was determined in the equatorial region of fruits, at opposite points, after removal of a small portion of the skin, using an electronic penetrometer (GÜSS Manufacturing Ltd, Cape Town, South Africa) equipped with 7.9 mm diameter probe.

Texture attributes were analyzed with a TAXT-Plus® electronic texturometer (Stable Micro Systems Ltd, Surrey, UK), in terms of strengths required to the skin rupture (N) and flesh penetration (N) using a 2 mm diameter probe, which was introduced into the flesh at a depth of 10 mm, with pre-test, test and post-test rates of 30, 3 and 40 mm s^{-1}, respectively.

TA (% malic acid) values were obtained by diluting a 10 mL juice sample (obtained by processing the fruits in a centrifuge) into 90 mL of distilled water, followed by titration with 0.1 N NaOH to pH 8.1. A TitroLine® Easy automatic titrator from SCHOTT Instruments (Mainz, Germany) was used for titration of samples.

The SSC (ºBrix) were determined in a digital refractometer model PR201α (Atago®, Tokyo, Japan), using an aliquot of juice obtained for TA quantification.

For the sensory analysis, an acceptance test by hedonic scale was used, where the taster expressed the degree of liking or disliking the fruit in relation to two specific attributes: sweetness / acidity relationship and texture. The scale used in the test had seven points, where 1 = disliked very much, 2 = disliked much, 3 = disliked, 4 = neither liked nor disliked, 5 = liked, 6 = liked much, 7 = liked very much. For evaluation purposes, 30 untrained individuals were invited. Each judge was provided with an evaluation worksheet and fruit samples of all tested storage conditions. For this, the randomized samples were composed of a fruit slice (corresponding to ¼ of fruit with no skin and seed), being offered (at 20 ± 5ºC) in plastic containers coded with three-digit numbers. The judges were also questioned regarding the presence of strange flavor in fruits and if they judged necessary they could make observations and comments regarding the characteristics of fruits being evaluated. Fruits with incidence of flesh browning were not used in sensorial evaluations.

The analysis of incidence and severity of flesh browning was evaluated by means of a cut in the cross section of fruits. In order to evaluate incidence

(%), the amount of pears that presented internal regions of the flesh with any kind of browning was counted. Fruits that presented formation of cavities inside the flesh were counted. Severity was evaluated according to the following scale: 1 = no incidence of flesh browning; 2 = low incidence of flesh browning, with up to 10% of flesh affected; 3 = moderate incidence of flesh browning, with 11 to 30% of flesh affected; and 4 = severe incidence of flesh browning, with more than 30% of flesh affected.

Flesh color was evaluated in terms of lightness values (*L*) using a Konica Minolta® CR 400 colorimeter. Thus, the lower the *L* value, the browner the flesh. Two readings per fruit were performed immediately after cut in the middle region of fruits.

Before application of treatments, four samples of 15 fruits were evaluated to determine the initial quality of pears, which presented average flesh firmness of 56.5 N, SSC of 12.9 ºBrix and TA of 0.28% of malic acid.

Data were submitted to analysis of variance (ANOVA), and percentage data were transformed by the formula: arcsin $[(x + 1) / 100]^{1/2}$ before being submitted to ANOVA. The Tukey test was used to compare means ($p < 0.05$). For these procedures, the SAS statistical software (SAS Institute, Cary, NC, USA) was used.

RESULTS AND DISCUSSION

CA conditions caused a significant difference ($p < 0.05$) in skin color, respiratory rate and ethylene production, texture attributes, flesh firmness, sensory analysis, flesh color and flesh browning incidence and severity. However, SS and TA attributes showed no significant difference among treatments ($p > 0.05$).

The ethylene production rate was higher in fruits kept under *LO* conditions with pCO_2 <0.03 kPa and pCO_2 = 1.0 kPa when compared to the other CA conditions (Table 1). After three days of shelf life, *LO* condition with pCO_2 <0.03 kPa presented the highest ethylene production rate. The increase in partial CO_2 pressures for *LO* conditions with pCO_2 of 1.0; 2.0 or even 3.0 kPa, may have limited the autocatalytic ethylene production, since CO_2 has an inhibitory effect on the action of this hormone (WANG AND SUGAR, 2013). Both at three and six days of shelf life, fruits stored under *ULO* presented lower ethylene production compared to those stored under *LO* with pCO_2 = 1.0 kPa. Possibly, the storage of fruits under *ULO* (pO_2 = 0.5 kPa) reduced the conversion of ACC (1-aminocyclopropane-1-carboxylic acid) into ethylene, because the ACC oxidase enzyme requires O_2 for reaction (BOTH et al., 2014). Some authors also claim that storage at low partial O_2 pressures may reduce the gene expression of the ACC oxidase enzyme (LARA et al., 2011; BOTH et al., 2014), which would also limit ethylene production.

At the end of storage, fruits stored under *LO* with pCO_2 <0.03 kPa and pCO_2 = 3.0 kPa presented higher respiratory rate than those stored under *ULO* (Table 1). After three days of shelf life, the respiratory rate was higher in fruits of *LO* treatment with pCO_2 = 1.0 kPa, compared to those stored under *ULO*. The reduction in the respiratory activity by *ULO* may be due to the decrease in the activity of several enzymes with oxidase activity, such as cytochrome c oxidase, polyphenol oxidase, ascorbic acid oxidase and glycolic acid oxidase (STEFFENS et al., 2007). In addition, O_2 is the ultimate acceptor in the electron carrier chain, and thus, low O_2 levels may restrict this respiration stage. After six days of shelf life, CA condition of *LO* with pCO_2 = 3.0 kPa presented the lowest respiratory rate, followed by *LO* conditions with pCO_2 = 2.0 kPa and *ULO*, which did not differ from each other. *LO* treatment with pCO_2 = 1.0 kPa provided higher respiratory rate in relation to *ULO* and *LO* conditions with CO_2 of 2.0 and 3.0 kPa, being, however, smaller compared to *LO* with pCO_2 <0.03 kPa. The lower respiratory rate of fruits stored under high partial CO_2 pressures is possibly related to the inhibitory effect that this gas exerts on the phosphofructokinase enzyme in the glycolytic route, as well as on the succinate dehydrogenase and isocitrate dehydrogenase enzymes in the tricarboxylic acid cycle (STEFFENS et al., 2007; WANG; SUGAR, 2013).

For evaluations carried out immediately at the end of storage and after three days of shelf life, fruits stored under LO condition with pCO_2 <0.03 kPa presented more yellow skin color (lower $h°$) than fruits stored under other CA conditions (Table 1). At the end of storage, *LO* condition with pCO_2 = 1.0 kPa gave fruits a less green skin color in comparison to fruits stored under *LO* with pCO_2 = 3.0 kPa, but did not differ from those stored under *ULO* and *LO* with pCO_2 = 2.0 kPa. According to Blaszczyk (2012), for pears stored under CA, higher partial CO_2 pressures provide greener fruits after storage due to the maintenance of higher chlorophyll a and b levels in the skin. Other authors have also attributed the higher retention of green color in pears stored under CA to lower O_2 partial pressures (GALVIS-SÁNCHEZ et al., 2003). After six days of shelf life, there was no difference in the skin color of fruits among treatments.

After storage, European pears tend to

exhibit a characteristic yellowish color during shelf life, and this behavior occurs due to the action of chlorophyllases, which are enzymes induced by the action of ethylene, accompanied by the synthesis of carotenoids in fruits (DHILLON AND MAHAJAN, 2011; GAGO et al., 2013). However, it is not advantageous for pears to exhibit a very yellow coloration at the end of the storage period, since this would be an indication that fruits are closer to the senescence stage. In addition, according to Cheng et al. (2012), excessive skin yellowing may cause commercial devaluation for some pear cultivars.

Storage under *LO* with pCO_2 <0.03 kPa provided higher flesh firmness at six days of shelf life compared to those stored under other CA conditions (Table 2). After storage, European pears showed high rate of flesh firmness loss during shelf life, which is due to the synthesis and action of several enzymes associated with modifications in the texture of fruits, such as polygalacturonases, β-galactosidases, α-arabinofuranosidase, β-xylosidases and pectinamethyl esterases (GALVIS-SÁNCHEZ AND MORAIS, 2001). Thus, the normal ripening of fruits is necessary for them to acquire a buttery and juicy texture, which is demanded by consumer markets of most of the world (MAKKUMRAI et al., 2014). However, after prolonged storage under inadequate CA conditions, some authors have reported abnormal ripening patterns in European pears, with fruits remaining firm and dry, being unable to develop desirable texture for consumption (MURAYAMA et al., 2002; MARTIN et al., 2015). In the present work, although flesh firmness of fruits stored under *LO* with pCO_2 <0.03 kPa is close to that indicated for the consumption of 'Rocha' pears (~20 N) (CAVACO et al., 2009), this condition provided lower rate of flesh firmness loss (60.3%) when compared to fruit stored under other conditions (firmness loss between 70.2 and 74.5%).

The strength for skin rupture was higher for fruits stored under *LO* with pCO_2 <0.03 kPa than those stored under *LO* with pCO_2 = 1.0 and 3.0 kPa (Table 2). In relation to the strength for flesh penetration, fruits stored under *LO* with pCO_2 <0.03 kPa presented the highest values in comparison to the other CA conditions (Table 2). On the other hand, *LO* conditions with pCO_2 = 2.0 and 3.0 kPa provided fruits with the lowest strength values for flesh penetration, but did not differ from those stored under *ULO*. *LO* condition with pCO_2 <0.03 kPa, for combining the highest O_2 content evaluated with pCO_2 <0.03 kPa in the storage atmosphere was, among all those evaluated, the one that caused less limitation on the metabolic activity of fruits during storage (Table 1). Possibly, fruits stored under this condition were closer to the senescence stage in relation to the other conditions evaluated, as a result of the higher action of enzymes that act on cell wall degradation, such as pectinametersterases (GALVIS SÁNCHEZ; MORAES, 2001). Some authors also suggest that the loss of ripening capacity in European pears after prolonged storage is associated with lower content of soluble polyuronides (MURAYAMA et al., 2002), as well as a low degree of depolymerization of pectins and hemicellulosic polysaccharides in fruit flesh (MURAYAMA et al., 2006).

There was no effect of CA conditions on TA and SSC of fruits (Table 2). Working with 'Royal Gala' apples, Both et al. (2014) did not observe differences in TA of fruits stored under CA conditions with ultra low (0.5 kPa) or low O_2 (1.2 kPa). However, Corrêa et al. (2010) found that the increase of pCO_2 from values below 0.5 kPa to 2.0 kPa caused an increase in TA in 'Fuji' apples. In addition to the partial gas pressures, other factors may influence the effect of CA on TA, such as species, crop, maturity stage at harvest and storage period (ELGAR et al., 1997). For SSC, Brackmann et al. (2005) also found no differences in the SSC levels of 'Pink Lady' apples in response to different CA conditions. According to these authors, large changes in SSC during CA storage are usually not perceived, since sugars are substrates of respiration, whose use only occurs after a marked consumption of organic acids.

The different CA conditions evaluated had no effect on sensory attributes (Table 3). As TA and SSC content of fruits did not differ among treatments (Table 2), it is possible that this result was also perceived by consumers during the acceptance test, providing similar scores for the sweetness / acidity attribute. Variations in flesh firmness and strength for flesh penetration, among storage conditions (Table 2), were not perceived or did not affect the tasters' acceptance of texture attribute. Working with 'Rocha' pears submitted to different CA conditions for nine months, Galvis-Sánches et al. (2004) found that fruits with different flesh firmness values presented similar scores in sensory evaluations for attribute "firmness", similar to results obtained in the present study. For none of the storage conditions evaluated, strange taste in fruits was observed.

The incidence of flesh browning was higher in fruits stored under *LO* with pCO_2 = 3.0 kPa than in fruits stored under lower CO_2 content (*ULO* and *LO* with pCO_2 <0.03 kPa), but did not differ from those stored under *LO* combined with pCO_2 of 1.0 or 2.0 kPa (Table 4). Flesh color attribute *L* of fruits stored under *LO* with higher CO_2 content

(2.0 and 3.0 kPa) was also lower (browner flesh) than in fruits stored under *ULO* (Table 4). Thus, it is possible to observe that the increase in partial CO_2 pressures (from values lower than 0.03 to 3.0 kPa) is more related to the incidence of flesh browning, than the decrease of partial O_2 pressures (from 1.0 to 0.5 kPa), since pears stored under *ULO* (0.5 kPa) had low incidence of flesh browning (6.2%). These results indicate that *LO* condition with pCO_2 = 3.0 kPa is not indicated for the storage of 'Rocha' pears since it provides high incidence of flesh browning (48%), possibly associated with higher partial CO_2 pressures inside the flesh, which favors the occurrence of the disorder (FRANCK et al., 2007). The increase in CO_2 content reduces fruit respiration by inhibiting the tricarboxylic acid cycle and the glycolytic route (WANG AND SUGAR, 2013). Thus, higher partial CO_2 pressures favor the shift from aerobic respiration to anaerobic respiration in fruits, causing the synthesis of toxic products to cells and damaging the cell membranes (PEDRESCHI et al., 2009). With the loss of membrane selectivity and the cell decompartmentalization process, phenolic compounds that are located in vacuole can come into contact with the polyphenol oxidase enzyme (PPO), being oxidized in very reactive compounds that form brown colored polymers, causing the symptoms of flesh browning (YAN et al., 2013).

The CA conditions that provided the lowest flesh browning severity values in the present work, such as *ULO* and *LO* with pCO_2 = 1.0 kPa, are not recommended for the storage of 'Rocha' pears produced in Portugal (CAVACO et al. , 2009; GAGO et al., 2013), mainly because they induce the occurrence of flesh browning in fruits. According to Franck et al. (2007), the susceptibility to flesh browning is related to several pre-harvest factors, especially to climatic conditions inherent to each production site. Fruits that grow under milder temperatures in the orchard are less susceptible to some physiological disorders, such as flesh browning in apples (CORRÊA et al., 2010) and pears (FRANCK et al., 2007), when compared to fruits that grow under lower temperatures. Possibly, these factors would provide pears produced in Brazil (with milder temperatures) lower susceptible to flesh browning than those produced in Portugal (with lower temperatures) and, therefore, greater tolerance to more limiting CA conditions, with lower partial pressures of O_2 and higher of CO_2.

TABLE 1 - Skin color, ethylene production and respiratory rate of 'Rocha' pears submitted to different storage conditions under controlled atmosphere for 270 days (-0.5 ± 0.1 ° C and RH of 96 ± 2%) evaluated after zero, three and six days of shelf life (20 ± 5 ºC and RH of 63 ± 2%).

pO_2 and pCO_2 (kPa)	Day 0	Day 3	Day 6
		Skin color (h^o)	
0.5 and <0.03	96.02 ab	93.67 a	86.00 ^{ns}
1.0 and <0.03	88.18 c	87.97 b	84.86
1.0 and 1.0	93.92 b	93.26 a	85.14
1.0 and 2.0	97.59 ab	93.75 a	85.86
1.0 and 3.0	98.54 a	92.51 a	84.71
VC (%)	2.2	1.4	1.9
		Ethylene production rate (nmol C_2H_4 kg^{-1} s^{-1})	
0.5 and <0.03	0.074 b	0.063 c	0.074 b
1.0 and <0.03	0.156 a	0.202 a	0.117 ab
1.0 and 1.0	0.132 a	0.116 b	0.134 a
1.0 and 2.0	0.075 b	0.113 bc	0.099 ab
1.0 and 3.0	0.076 b	0.101 bc	0.096 ab
VC (%)	20.4	17.6	21.6
		Respiratory rate (nmol CO_2 kg^{-1} s^{-1})	
0.5 and <0.03	103.63 b	491.12 b	756.96 c
1.0 and <0.03	143.95 a	623.58 ab	1134.11 a
1.0 and 1.0	122.71 ab	716.16 a	904.51 b
1.0 and 2.0	127.77 ab	651.22 ab	742.64 c
1.0 and 3.0	137.45 a	580.01 ab	568.49 d
VC (%)	10.2	13.5	4.6

*Means followed by the same vertical letter did not differ by the Tukey test ($p<0.05$). ns: not significant ($p>0.05$).

TABLE 2 - Flesh firmness, strength for skin rupture and flesh penetration, titratable acidity and soluble solids content of 'Rocha' pears submitted to different storage conditions under controlled atmosphere for 270 days (-0.5 ± 0.1 and 96 ± 2% RH), and evaluated after six days of shelf life (20 ± 5 ºC and UR of 63 ± 2%).

pO_2 and pCO_2 (kPa)	Flesh firmness (N)	Strength for skin rupture (N)	Strength for flesh penetration (N)	Titratable acidity (%)	Soluble solids content (ºBrix)
0.5 and <0.03	16.00 b	4.62 ab	1.29 bc	0.149 ns	12.77 ns
1.0 and <0.03	22.41 a	5.33 a	1.70 a	0.124	13.12
1.0 and 1.0	16.83 b	4.52 b	1.40 b	0.139	12.70
1.0 and 2.0	15.12 b	4.87 ab	1.15 c	0.131	13.12
1.0 and 3.0	14.41 b	4.20 b	1.12 c	0.098	13.40
VC (%)	7.8	7.9	7.4	18.3	7.4

*Means followed by the same vertical letter did not differ by the Tukey test (p<0.05). ns: not significant (p>0.05).

TABLE 3 - Mean scores of the sensory attributes of 'Rocha' pears submitted to different storage conditions in controlled atmosphere for 270 days (-0.5 ± 0.1 ºC and RH of 96 ± 1%), and evaluated after six days of shelf life (20 ± 5 ºC and RH of 63 ± 2%).

pO_2 and pCO_2 (kPa)	Attribute	
	Sweetness/acidity	Texture
0.5 and <0.03	4.36ns	4.00ns
1.0 and <0.03	4.64	4.68
1.0 and 1.0	4.73	4.68
1.0 and 2.0	5.27	4.86
1.0 and 3.0	5.05	4.95
VC (%)	24.8	29.5

ns: not significant (p>0.05).

TABLE 4 - Incidence and severity of flesh browning and flesh color of 'Rocha' pears submitted to different storage conditions under controlled atmosphere for 270 days (-0.5 ± 0.1ºC and RH of 96 ± 2%) maintained for six days of shelf life (20 ± 5 ºC and RH of 63 ± 2%).

pO_2 and pCO_2 (kPa)	Flesh browning incidence (%)	Flesh browning severity (1-4)*	Flesh color (*L*)
0.5 and <0.03	6.20 b	1.09 b	80.48 a
1.0 and <0.03	8.88 b	1.12 b	79.27 ab
1.0 and 1.0	18.21 ab	1.32 b	78.64 ab
1.0 and 2.0	26.52 ab	1.39 b	77.52 b
1.0 and 3.0	48.02 a	1.79 a	77.45 b
VC (%)	35.3	10.8	1.2

*Severity scale: 1 = no incidence; 2 = up to 10% of flesh affected; 3 = 11 to 30% of flesh affected; and 4 = 30% of flesh affected. Means followed by the same vertical letter did not differ by the Tukey test (p<0.05). ns: not significant (p> 0.05).

CONCLUSIONS

Storage of 'Rocha' pears in CA for 270 days under *LO* condition with pCO_2 <0.03 kPa provides less flesh firmness loss in fruits after the storage period and yellower skin at the end of storage.

LO condition with pCO_2 = 3.0 kPa is not indicated for the storage of 'Rocha' pears, as it provides greater occurrence of flesh browning.

ULO (pO_2 = 0.5 kPa and pCO_2 <0.03 kPa) and *LO* conditions with pCO_2 = 1.0 kPa are the most indicated for the storage of 'Rocha' pears, delaying the ripening of fruits without, however, inhibiting the development of texture or increasing the occurrence of flesh browning.

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