Volatile compounds profile of fresh-cut peki fruit stored under different temperatures

Perfil dos compostos voláteis do pequi minimamente processado armazenado sob diferentes temperaturas

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Abstract

The objective of this work was to identify and verify the influence of time and temperature on the volatile compounds profile of fresh cut peki. Peki fruits were washed, sanitized, their kernels were extracted, and they were packaged and stored for 15 days at 0, 5, and 10 °C and 6 days at 22 °C. The volatiles compounds were analyzed by GC – MS. Ethyl hexanoate and ethyl octanoate were found in higher percentages, 63 and 16.3%, respectively. The determined volatiles were not influenced by the storage period. Hexanoic acid, ethyl 2-octenoate and ethyl decanoate were not influenced by the different temperatures. The temperatures 0, 5, and 10 °C did not influence ethyl hexanoate, ethyl 2-hexenoate and ethyl octanoate either. In addition, the temperatures 5, 10, and 22 °C did not influence ethyl hexanoate, cis-β-ocimene and ethyl octanoate. The temperature of 22 °C determined higher percentages of ethyl hexanoate and lower percentages of ethyl octanoate, in comparison to the temperature of 0 °C, and higher percentages of ethyl 2-hexenoate in comparison to the temperatures of 0, 5, and 10 °C. The temperature of 5 °C determined higher percentage of cis-β-ocimene when compared with the temperature of 0 °C. The storage temperatures of 0 and 5 °C were the most appropriate for the conservation.
Keywords: *exotic fruit; Caryocar brasiliense Camb.; storage.*

Resumo

O objetivo do trabalho foi identificar e verificar a influência do tempo e da temperatura sobre o perfil dos compostos voláteis de pequi minimamente processado. O pequi foi lavado, sanificado, seus pirênios extraídos, embalados e armazenados por 15 dias a 0, 5 e 10 °C e 6 dias a 22 °C. Os compostos voláteis foram analisados por CG-EM. Hexanoato de etila e octanoato de etila foram encontrados em porcentagens mais altas, 63 e 16.3% respectivamente. Os voláteis determinados não foram influenciados pelo tempo de armazenamento. Ácido hexanóico, 2-octenoato de etila e decanoato de etila não foram influenciados pelas diferentes temperaturas. As temperaturas 0, 5 e 10 °C também não influenciaram o hexanoato de etila, 2-hexenoato de etila e octanoato de etila. Por outro lado, as temperaturas 5, 10 e 22 °C não influenciaram o hexanoato de etila, cis-β-ocimeno e octanoato de etila. A temperatura de 22 °C determinou porcentagens mais altas de hexanoato de etila e porcentagens mais baixas de octanoato de etila em comparação à temperatura de 0 °C, e porcentagens mais altas de 2-hexenoato de etila em comparação a temperaturas de 0, 5 e 10 °C, enquanto a temperatura de 5 °C determinou porcentagens mais altas de cis-β-ocimeno, quando comparada com a temperatura de 0 °C. As temperaturas de armazenamento de 0 e 5 °C foram as mais apropriadas para a conservação.
Palavras-chave: *fruta exótica; Caryocar brasiliense Camb.; armazenamento.*

1 Introduction

The *"cerrado"* or Brazilian savanna region presents a wide range species of edible fruits which have long been used by human populations (BARBOSA, 1996). These native fruits are consumed in their natural state or in the form of fruit compote, porridges, cakes, bread, biscuits, jams, and liqueurs (ALMEIDA, 1998). Among the *'cerrado's* native, fruit trees, the peki tree deserves special attention on account of its high incidence in the *cerrado* landscape and fruit sensorial characteristics (PAULA, 2006) such as its totally exotic aroma.

The term 'pequi fruit' comes from the tupi language, in which "py" means 'skin' and "qui" refers to 'thorn'; such a combination results in a thorny skin possibly due to the fact that the kernels are covered with thin thorns (ALMEIDA; SILVA, 1994; RIBEIRO, 2000).

The peki fruits, which contains from 1 to 4 seeds under a pasty, oleaginous, yellow pulp (internal mesocarp), are rich in oil, carbohydrates, proteins and vitamins (MARTINS; TEIXEIRA; OLIVEIRA, 1983). Studies on the centesimal composition of the peki fruit's edible pulp (internal mesocarp), found in the southern region of the state of *Minas Gerais,* show that it has 49.2% of moisture, 20.5% of lipids, 4.2% of protein, 6.8% of fibers, 0.4% of ashes, 18.9% of glycides, 20000 UI of vitamin A, and 105 mg.100 g^{-1} of vitamin C (VILAS BOAS, 2004).

Nevertheless, the consumption of peki fruits may cause some inconveniences such as a great microbiological contamination since the fruits are picked up from the ground and many split open exposing the internal mesocarp to direct contact with the soil; post-harvest losses may also occur; and commercialization

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takes place only in the regions which produce the fruit, apart from the fact that many housewives complain about the difficulty of peeling the fruit whereas others dislike the strong residual odor which remains on their hands after the contact with the fruit.

In order to solve those problems, minimal processing is an ally since it can guarantee the maximum convenience and quality are expected by modern consumers by offering a product with complete hygienic-sanitary safety, as well as by contributing to an increase in consumption not only in the peki producing regions, but also throughout the entire country as well (RODRIGUES, 2005).

Fresh-cut fruit and vegetables are defined as products which undergo operations of selection, washing, and peeling and cutting, all of which eliminate their non-edible parts, such as skin, stalk, and seeds. Such operations are followed by slicing and then they are ready for immediate consumption (100% of use), which enables the plant to maintain freshness, quality, and sanitation.

In order to achieve this, temperature control is very important since it minimizes the effects of damage on tissues in fresh-cut products (BRECHT, 1995). The use of refrigerated storage for this kind of product is based on the notion that low temperatures delay the growth of a majority of microorganisms by reducing their respiratory rate and are effective in reducing enzymatic activities (LUENGO; CALBO, 2001) increasing the useful life of products and lowering losses in aroma, taste, color, texture, and other quality attributes of the stored product (ANTUNES; DUARTE FILHO; SOUZA, 2003).

The typical aroma of a fruit is usually developed during a short period of time, which coincides with its full ripening, time during which the metabolism is characterized by catabolism of small quantities of lipids (fatty acids), proteins (amino acids), and carbohydrates. These elements are, enzimatically, converted into volatile compounds (SEYMOUR;TAYLOR; TUCKER, 1993) such as esters, lactones, alcohols, acids, aldehydes, ketones, acetals, hydrocarbons and some phenols, and ethers and heterocyclical oxygenated compounds. Volatile compounds have a small molecular weight (PM < 250), are found in complex mixtures, and the existing relation between their concentration and proportion is of considerable importance (CHITARRA; CHITARRA, 2005).

A real concern in submitting peki fruits (*Caryocar brasiliense* Camb.) to minimal processing is the possibility of depriving them of their aroma, but their use to aromatize various culinary dishes is the main reason for consumption. At higher storage temperatures, volatiles' synthesis routes are activated; part of those volatile compounds emanates from the environment and the other part may become liquid again when the fruit is submitted to refrigeration (which affects the internal cells). On the other hand, when held in intercellular spaces, the external cells are affected, which modifies the product's sensorial characteristics. The main volatile compounds that behave in such a way are the esters derived from the reaction between alcohols of C_1-C_6 and carboxylic acids of C_1-C_6 (WEICHMANN, 1987).

Due to these factors, this study aims to verify the influence of time and temperature on the volatile compounds profile of fresh cut peki (*Caryocar brasiliense* Camb.) when submitted to temperatures of 0 ± 1 °C, 5 ± 1 °C, 10 ± 1 °C, and 22 ± 1 °C during 15 days of storage.

2 Material and methods

Ripe peki fruits were picked up from the ground on three different farms in the town of Cordisburgo, which is located at an altitude of 720 m, in the metropolitan region of Belo Horizonte, Minas Gerais, forming an only batch. The peki fruits were collected randomly and had intact epicarp and did not present any sign of damage or rottenness. After previous selection, the fruits were transported to the fruit and vegetables Post-harvest Laboratory of the Food Science Department at the Federal University of Lavras (UFLA), located in the city of Lavras, Minas Gerais.

As soon as the fruits arrived at the laboratory, they were washed with neutral detergent, rinsed in running water, and stored at 15 ± 1 °C under a relative humidity between 90 and 95%, for approximately 13 hours in the processing room.

After this stage, the peki fruits were sanitized with sodium hypochlorite (NaClO) at 200 ppm (pH 7.0) for 15 minutes, than they were hand cut and pitted (removal of the internal mesocarp). The peki fruits were sanitized with sodium hypochlorite (NaClO) at 100 ppm (pH 7.0) for more than 15 minutes and, after that, they were sieved using a plastic strainer for the removal of water excess. Afterwards, the seeds were weighed (approximately 110 g), disposed in plastic trays of polypropylene (15 × 11.5 × 4.5 cm) with lids, transported to the chambers, and stored at different temperatures (0 ± 1 °C, 5 ± 1 °C, 10 ± 1 °C and 22 ± 1 °C) under a relative humidity of 95% in triplicates for 15 days.

Every three days, fresh-cut peki fruits were taken out of the polypropylene packages, frozen with liquid nitrogen, disposed in plastic bags, and stored at –84 °C. After 15 days, the frozen peki fruits were transported in polystyrene containers to the Laboratory of Molecular Bioactivity of the Institute of Chemistry at the Federal University of Goiás, state of Goiânia, where the extraction, identification, and quantification of most of the volatile compounds were carried out.

The essential oil of fresh-cut peki fruits (approximately 110 g) was extracted by hydrodistillation in a modified Clevenger machine with the use of a triple condenser for one hour. The oil was collected with 20 drops of CH_2Cl_2 Ultra Residue 95% (J T Baker), dried over Na_2SO_4 anhydrous P.A (*Dinâmica*), and disposed in amber glass flasks. After that, the CH_2Cl_2 was evaporated with liquid nitrogen and the glass flask was weighed in order to determine the effectiveness of the extracted oil. In the subsequent stage, the oil was stored in a freezer at –18 °C until its analysis.

Oil sample analyses were performed in a GC-MS Shimadzu QP5050A instrument using the following conditions: a CBP-5 (Shimadzu) fused silica capillary column (30 m long × 0.25 mm i.d. × 0.25 µm film thickness composed of 5% phenylmethylpolysiloxane) connected to a quadruple detector operating in the EI mode at 70 eV with a scan mass range of 40-400 *m/z* at a sampling rate of 1.0 scan s^{-1}; carrier gas He (1 ml min^{-1}); and the injector and interface temperatures were 220 and 240 °C, respectively, with a split ratio of 1: 20. The injection volume was 0.4 µL (20% in CH_2Cl_2) in the split mode and the oven temperature was raised from 60 to 246 °C, with an increase of 3 °C min^{-1}, then 10 °C min^{-1} to 270 °C, holding the final temperature for 5 minutes. Individual components were identified by comparing their retention indexes, made through co-injection with a C_8-C_{32} n-alkanes series (VAN

DEN DOOL; KRATZ, 1963), mass spectra with those of the literature (ADAMS, 2001) and a computerized MS-data base using NIST libraries (1988).

Due to the fact that the seeds stored at 22 ± 1 °C lasted only six days of storage, two experiments in Completely Randomized Design (CRD) were carried out: experiment 1, with a factorial 4 × 3, that is, four different temperatures (0 ± 1 °C, 5 ± 1 °C, 10 ± 1 °C9, and 22 ± 1 °C) and three time periods (0, 3, and 6 days), in triplicates; and experiment 2, with a factorial 3 × 6, that is, three different temperatures (0 ± 1 °C, 5 ± 1 °C, and 10 ± 1 °C) and 6 time periods (0, 3, 6, 9, 12, and 15 days), in triplicates. Each experimental device was made up of a polypropylene plastic package which contained approximately 110 grams of peki fruit.

The statistical analyses were carried out with the aid of the SISVAR program (FERREIRA, 2000). After a variance analysis, the most significant averages were compared using the Tukey test, at 1 and 5% of probability. The polynomial regression models were selected based on the significance of the F test of each tested model and also on the coefficient of determination.

3 Results and discussion

The yield of the essential oil extracted from 110 g of fresh-cut peki fruits was 0.0020% for the seeds stored at 0 ± 1 °C; 0.0022% for the ones stored at 5 ± 1 °C; 0.0037% for the ones stored at 10 ± 1 °C, and 0.022% for those stored at 22 ± 1 °C. The kernels or seeds which were stored at 0 ± 1 °C, 5 ± 1 °C, and 10 ± 1 °C differed, statistically, from those stored at 22 ± 1 °C ($p < 0.01$), all of which produced higher yields in the extraction. These amounts are greater than the ones obtained from the extraction of essential oil of the lemon tree's fruit ('Siciliano' and 'Eureka') using the same method, Clevenger, which produced 5.13 kg of oil by ton of fruit or 0.000513% (GRASSI FILHO; PENTEADO; SANTOS, 2005). However, such amounts did not produce the yield found in the peki fruit pulp (internal mesocarp) (0.09%) by Marques (2001), probably due to the use of another extraction technique (steam distillation). It is important to mention that according to Laencina, Melendreras and Flores (1988), the yield is not associated only with the method of extraction, but also with the genetic material and their different reactions to environmental conditions.

The differences in the yield of the essential oil extraction are probably due to the fact that the plant's physiological metabolism is faster in higher temperatures as far as the respiratory rate is concerned (CHITARRA; ALVES, 2001). Damiani et al. (2006), in a research on the respiratory behavior of peki fruits under various temperatures (0 ± 1 °C, 5 ± 1 °C, 10 ± 1 °C) during 15 days of storage, verified that the higher the temperature, the more intense the respiration is, which therefore reduces the product's useful life. Since kernels breathe more intensely, there is an increase of acyl-CoA which, united with a molecule of alcohol – both derived from the β-oxidation or the performance of the enzyme lipoxigenase (LOX) in the lipidic fractions, as well as the metabolism of amino acids, through the alcohol acyltransferase enzyme (AT) –, will form a greater quantity of volatiles (KNEE, 2002).

Studies carried out by Rodrigues (2005) on fresh-cut peki fruits stored at 6 °C during 15 days show that the peroxide index (a product of the oxidation of lipids in the presence of oxygen) increased during that span of time (90.626 to 138.205 μg of $O_2.g^{-1}$). This proves, therefore, the performance of lipoxygenase enzymes on unsaturated acids.

The kernels stored at 22 ± 1 °C, presented, after the sixth day of storage, undesirable characteristics for consumption, such as an unpleasant odor, darkening and softening of the pulp, and fungi on the product's surface. For this reason, the analyses of volatile compounds were carried out up to the sixth day for the kernels stored at this temperature.

The main volatile compounds identified in fresh-cut peki fruits stored under different temperatures (0 ± 1 °C, 5 ± 1 °C, 10 ± 1 °C , and 22 ± 1 °C) are presented in Table 1.

In the total ion chromatogram, 16 peaks were identified (Figure 1). Their larger areas were made up of 7 components, mainly esters. They are: hexanoic acid, ethyl hexanoate, ethyl 2-hexenoate, cis-β-ocimene, ethyl octanoate, ethyl 2-octenoate, and ethyl decanoate. Hexanoic acid, ethyl hexanoate, and ethyl decanoate were attemptly identified by comparing mass espectra and elution order. Among them, the ethyl hexanoate and the ethyl octanoate may be considered impact compounds due to their association with the peki fruit's characteristic aroma, and because they may be found in higher percentages (approximately 63 and 16.3%, respectively).

These volatile compounds were found by Passos et al. (2002) when they researched the influence of pluviometric precipitation on the composition of volatiles in the kernels of peki fruit (*Caryocar brasiliense*).

According to the retention time of each peak in the analysis, the index of retention of each compound (Table 1) was calculated using the Van Den Dool and Kratz's equation (1963). This method, together with the generated mass specters, made it possible to identify each compound with the aid of software in the gaseous chromatographer and connected to the mass spectrometer (CG-MS). Finally, in comparison with the literature, the seven main compounds obtained from the essential oil were identified.

None of the seven identified compounds underwent significant quantitative changes during the storage period. However, the isolated factor temperature significantly influenced some compounds ($p < 0.05$).

According to the data on Table 1, the volatiles hexanoic acid, ethyl 2-octenoate, and ethyl decanoate were not significantly influenced by the various temperatures. Temperatures of 0 ± 1 °C, 5 ± 1 °C, and 10 ± 1 °C did not influence, differently, the volatiles ethyl hexanoate, ethyl 2-hexenoate, and ethyl octanoate either. Apart from that, the temperatures of 5 ± 1 °C, 10 ± 1 °C, and 22 ± 1 °C did not influence, differently, the volatiles ethyl hexanoate, cis-β-ocimene, and ethyl octanoate. The temperature of 22 ± 1 °C led to higher percentages of ethyl hexanoate (82.34%) and lower percentages of ethyl octanoate (7.22%), in comparison with the temperature of 0 °C (42.20 and 27.95%, respectively), as well as higher percentages of ethyl 2-hexenoate (2.59%) in comparison with temperatures of 0 ± 1 °C (0.93%), 5 ± 1 °C (1.05%), and 10 ± 1 °C (1.24%). The temperature of 5 ± 1 °C led to a higher percentage rate of cis-β-ocimene (1.22%), when compared with the temperature of 0 ± 1 °C (0.51%).

Table 1. Scores of retention calculated using the Dool and Kratz (1963) equation, retention rates proposed by the literature (ADAMS, 2001), and percentage averages of volatile compounds of the essential oil in fresh-cut peki fruits stored at 0 ± 1 °C, 5 ± 1 °C, and 10 ± 1 °C for 15 days and 22 ± 1 °C for 6 days.

Volatiles compounds/peak	IR Calc.	IR Lit.	Temperature of storage (% área)				
			0 °C	5 °C	10 °C	22 °C	CV*
hexanoic acid/6	971.94	–	6.64[a]	4.45[a]	10.70[a]	0.12[a]	189.10
ethyl hexanoate/7	1000.11	–	42.20[b]	61.03[ab]	65.78[ab]	82.34[a]	47.14
ethyl 2-hexenoate/9	1037.73	1044	0.93[b]	1.05[b]	1.24[b]	2.59[a]	62.84
cis-β-ocimene/10	1043.04	1050	0.51[b]	1.22[a]	0.70[ab]	1.09[ab]	68.89
ethyl octanoate/13	1193.93	1197	27.95[a]	16.40[ab]	13.62[ab]	7.22[b]	72.39
ethyl 2-octenoate/14	1242.22	1249	3.85[a]	3.70[a]	2.13[a]	0.96[a]	84.76
ethyl decanoate/16	1392.55	–	3.72[a]	4.83[a]	5.08[a]	0.04[a]	118.49

Averages followed by the same letter, in lines, do not differ in the Tukey test to 5%; and *Average values of the coefficient of variation.

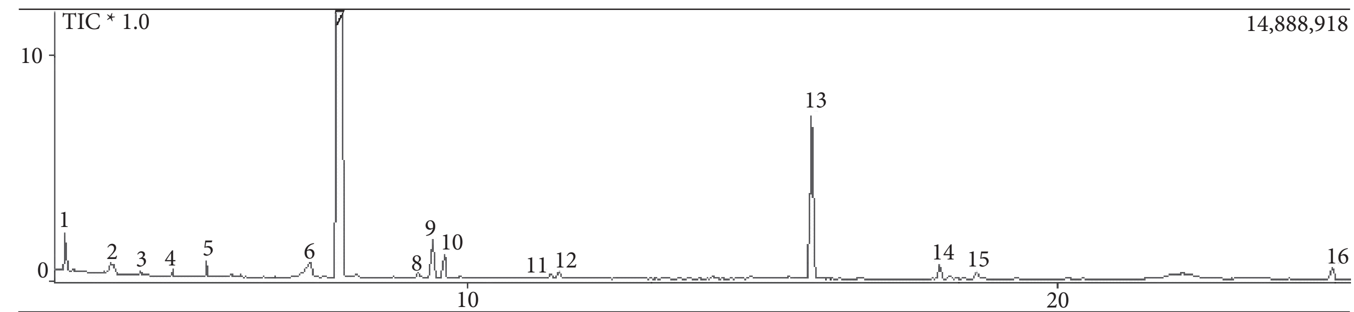

Figure 1. Total Ions chromatogram obtained in the analysis of gas chromatography in fresh-cut peki fruits stored under different temperatures (0 ± 1 °C, 5 ± 1 °C, 10 ± 1 °C, and 22 ± 1 °C) for 15 days.

The averages, which are seemingly very different but statistically equal, are due to the great number of the variation coefficients, which are found in all of the volatile compounds of fresh-cut peki fruits stored at 0 ± 1 °C, 5 ± 1 °C, and 10 ± 1 °C for 15 days and at 22 ± 1 °C for 6 days. Such a fact is explained by the great genetic variation of this fruit, which has been proved by studies carried out by researchers such as Oliveira (1998) who states the existence of genetic variability between and within three populations of peki as a consequence of the high gene flow – a result of the dispersion of pollen and seeds over long distances – and of the high rate of cross fecundation (84%). Naves et al. (2004) and Nascimento et al. (2004) concluded that the contribution to the variation of physical and chemical characteristics of the fruit, which is attributed to regions, plants, and to the fruit themselves, is indeed great. In other words, fruit are different in different areas, plants, and even inside the same plant. Rodrigues et al. (2004) also stated that the kernels significantly change their chemical composition according to the place of origin, and such a difference may be attributed to the climate, soil, and etc.

Studying volatile compounds in the peki fruit (kernels) at room temperature and using the same method of extraction and identification used in this study, De Paula et al. (2000) obtained average numbers of 94.41% of the compound ethyl hexanoate, which surpassed the value found in this study (82.34% at a temperature of 22 ± 1 °C). However, 2.22% of the compound ethyl octanoate was below the average found here (7.22%, at a temperature of 22 ± 1 °C).

Passos et al. (2002) found the following percentages in volatile compounds of the peki fruit's kernels (*Caryocar brasiliense*) at room temperature, low pluviometric precipitation, and high pluviometric precipitation, respectively: 0.8 and 4.7% of hexanoic acid, 91 and 70.9% of ethyl hexanoate, 3.5 and 7.0% of ethyl 2-hexenoate, 0.9% of cis-β-ocimene during the period of high pluviometric precipitation, 3.5 and 7.2% of ethyl octanoate, 1.2 and 4.4% of ethyl 2-octenoate, and 0.6% of ethyl decanoate during the period of high pluviometric precipitation. It is worthy to notice that the averages of the compounds hexanoic acid (0.12%), ethyl 2-hexenoate (2.59%), ethyl 2-octenoate (0.96%), and ethyl decanoate (0.04%), found in the present study, were lower than the ones found by Passos et al. (2002). However, the averages of the compounds ethyl hexanoate (82.34%) and ethyl octanoate (7.22%) match the ones presented by the researchers in question. The compound cis-β-ocimene was the only one which showed a higher percentage in relation to Passos et al. (2002), which is 1.09%.

According to Matteis and Fellmann (1999), the production of volatile compounds, like the esters, is reduced in fruits packed and stocked under refrigeration. Lamikanra and Richard (2002) confirmed it studying the volatile compounds of fresh-cut melons, and they found a decrease in the concentration of esters in a period of 24 hours due to the storage at 4 °C.

From the results presented at Table 1, it is possible to observe that the same behavior described by those authors occurred with the compounds ethyl hexanoate – which presented a percentage of 82.34% in the kernels stored at a temperature of 22 °C and only 42.20% in the kernels stored at a temperature of 0 °C – and also occurred with ethyl 2-hexenoate, which reached 2.59% in the kernels stored at room temperature and only 0.93% in those stored at 0 °C. However, the compound ethyl octanoate showed an antagonistic behavior, as it presented a percentage of 7.22% in fresh-cut peki fruits stored at room temperature, and 27.95% stored at 0 °C.

4 Conclusions

The exotic volatiles compounds of the peki fruit's internal mesocarp (kernels) derive basically from esters, in which the substances ethyl hexanoate and ethyl octanoate are found in greater proportions, both in the fruits stored at 0 ± 1 °C, 5 ± 1 °C, and 10 ± 1 °C and at 22 ± 1 °C.

Considering the obtained results and the useful life of the product, the storage temperatures of 0 ± 1 °C and 5 ± 1 °C are the most appropriate for the conservation of fresh-cut peki fruits for up to 15 days without causing considerable damage to their characteristic aroma based on their volatile profile.

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