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Volatile and non-volatile chemical composition of the white guava fruit (*Psidium guajava*) at different stages of maturity

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Abstract

The effect of the maturation stages on the chemical composition and volatile compounds of the white Guava (*Psidium guajava*) Cv. Cortibel was investigated during three different stages. The stages were characterized by evolution of color, pH, titratable acidity, sugars, soluble solids, vitamin C and volatile components. The fruits were stored at 24 °C and air humidity of 74% for 13 days. The volatile extracts were obtained using headspace technique and analyzed using gas chromatograph/mass spectrometry (GC/MS) system. The titratable acidity and sugars decreased. The pH level and amount of vitamin C increased throughout progress of maturation. The behavior of volatile compounds of fruits in the three stages of maturation was: in immature fruits and those in their intermediate stage of maturation, were predominantly the aldehydes such as (*E*)-2-hexenal and (*Z*)-3-hexenal. In mature fruits, esters like *Z*-3-hexenyl acetate and *E*-3-hexenyl acetate and sesquiterpenes caryophyllene, α-humulene and β-bisabollene are present.

Keywords: Guava; Chemical and non-chemical composition; Fruit ripening

1. Introduction

In general, fruits are harvested after having reached a physiological maturity stage, when development is completed and growing has ceased. From this point on, post-harvest ripening begins, and fruits acquire the organoleptic characteristics to be consumed (Manrique & Lajolo, 2004; Watada, 1986). The guava (*Psidium guajava*) is a native fruit of the American tropics. It is commercially important because of its flavor and aroma. It is nutritionally important due to its excellent source of vitamin C, niacin, riboflavin and vitamin A. The types and amounts of sugars determine the flavor of guavas. Generally, total sugars increases initially and then decreases during ripening. However, the relative proportions of its chemical composition change according to the cultivar and environmental conditions such as the climate and soil. Depending on the cultivar, the flavor compound may accumulate at different proportions during ripening, and thus may result in guava fruits having distinctive aroma and tastes. (Ali & Lazan, 1997; MacLeod & Troconis, 1982). In this study, the influence of different stages of maturation in the volatile and non-volatile chemical composition of the white guava was investigated.

2. Material and methods

2.1. Fruits

Cultivar Cortibel Guava fruits (*Psidium guajava*), were harvested from different trees in the southeast region of Brazil, in Santa Tereza City, Espírito Santo state. The fruits were harvested in February, in the morning and at

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random. The experiments were carried out at the Food Process Laboratory of Universidade Estadual do Norte Fluminense (State University of Northern Rio de Janeiro state). The fruits were cleaned and sanitized in sodium hypochlorite solution at 120 ppm for 15 min and wiped dry at room temperature. The fruits were stored in cardboard boxes with six fruits each, under atmosphere conditions: room temperature at 24 °C (±2 °C) and air humidity of 74% (±2). Air temperature and humidity were registered by a termohygrometer (Dwyer instruments INC. Model 485).

2.2. Non-volatile analyses

The analyses of the non-volatile chemical composition of the guava were carried out in the following way: three lots of fruits were used, each one containing three fruits visually presenting the same physical characteristics. Each lot was a replicate, performance the triplicate. The analyses were carried out on the first day, on the sixth day and on the thirteenth day post harvest.

2.2.1. Fruit color

The color of the fruit at different stages of maturity was instrumentally determined by using a colorimeter (Hunterlab, Miniscan model). The readings were made at three equidistant points in the equatorial axis of fruits. The results are presented through parameter Hunter "L", "a", "b", C and $h°$. The Hunter parameters, L range from 0 (dark color) to 100 (white color), a −80 to 0 (green color) and 0 to 100 (red color). b from −100 to 0 (blue color) and 0 to 70 yellow (positive value). C is a chroma obtained from $[(a^2 + b^2)]^{1/2}$ and $h°$ is hue angle obtained from arctangent of b/a, as McGuire (1992).

2.2.2. Titratable acidity

Titratable acidity was determined by standard procedures (AOAC, 1984) and the results expressed as percentage of citric acid.

2.2.3. pH

The pH of the pulp was determined by using digital pHmeter (330-SET).

2.2.4. Total soluble solids

Total soluble solids were determined by using Atago refractometer (model PR 201) and the results expressed as °Brix.

2.2.5. Sugar

About 1 g of sample was separated from each one of the three groups of fruits, making up three samples, and mixed with a 75% acetonitrile aqueous solution until reaching 50 mL, in accordance with Macrae (1988). After that, the samples were mechanically shaken in the ultra sonic bath (Model USC 1400) for 10 min, and then filtered through a Minisart filter (RC4, Sarotrius). The amount of sugar (glucose, fructose and sucrose) was determined by High Pressure Liquid Chromatography (HPLC) in a Shimadzu chromatograph system, equipped with a refraction index detector (RID – Model 10A). The Chromatographic conditions were: Lichrospher Column100 NH_2 5 μm (250 × 4 mm), loop of 100 μL. In the mobile phase, acetonitrile aqueous solution (75%) was used and at flux of 1 mL/min.

An extract was obtained out of each group and out of each extract three values of the content of the different sugars were obtained, of which an average was then made. The average of each extract was used as replicate, thus obtaining the triplicates. It is important to note that the triplicates are from different extracts.

2.2.6. Vitamin C

Three different samples were ground with 10 mL of water and homogeneous. A 1-g portion of guava fruit was put into tube and added oxalic acid solution (5%) until to complete 10 mL. The samples were centrifugate (Model HERMLEZ 382 K) at 5 °C for 10 min and 450 rpm. The upper phase was filtered through a Minisart filter (RC4, Sartorius). Amount of vitamin C was obtained by HPLC using a chromatography Shimadzu system. An ultraviolet visible detector (UV) was utilized. The column was an ODS-II C18 (ID 4.6 × 250 mm), a loop of 20 μL. The chromatographic conditions were: wavelengths of 254 nm, flow 1 mL/min. The mobile phase was prepared with 5 mmol/L of amonie brometocetyltrimetyll and 50 mmol/L of monobasic potassium phosphate and pH adjusted to 4.0 with H_3PO_4, as described by Benlloch, Fane, and Frigola (1993). The results were expressed as ascorbic acid.

The procedures utilized to determine the sugar content in triplicate was applied for vitamin C.

2.2.7. Statistic analyses

Three lots containing three fruits each were used to obtain the samples and each sample was considered a repetition, performing a triplicate. The data were assessed as average and analyzed by variance and regression analysis and average were compared using test Tukey, with a probability $P \leqslant 0.05$. Statistic Analysis System (SAS) was used.

2.3. Volatile composition

Volatile compounds were obtained using headspace technique as described by Franco and Rodriguez-Amaya (1983). Three replicate extractions were made in each stage and for each extraction six fruits were used. Fig. 1 shows the apparatus utilized to obtain the extracts. Six fruits were mashed (Wallita Master) and a homogeneous solution was obtained. Three hundred grams of sample were mixed with NaCl 30% w/w. It is necessary to inhibit enzymatic reactions. After that, the mixture was put into a balloon of 1000 mL. The glass column (capture column), containing porapak Q (polymer of 80–100 mesh) was adapted at the

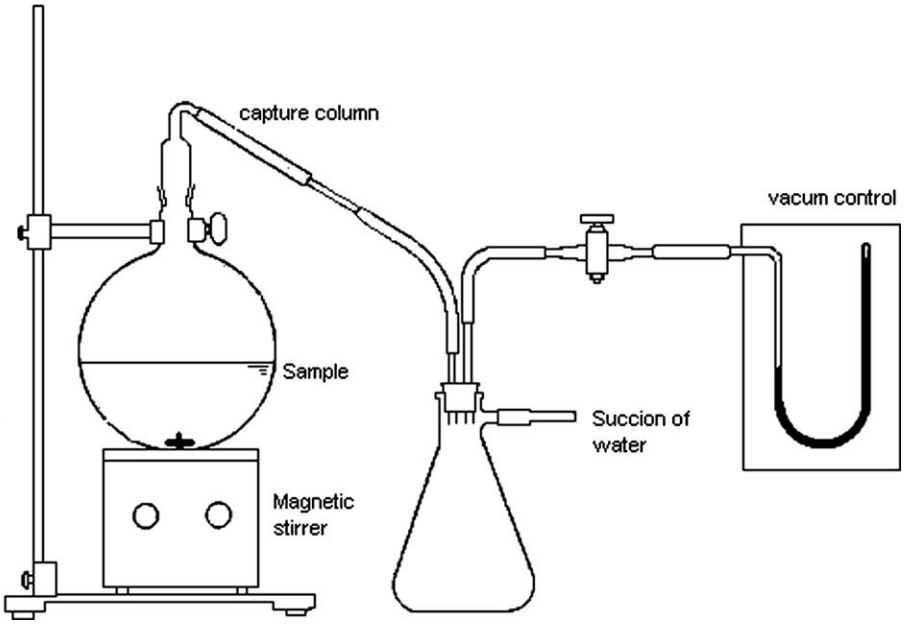

Fig. 1. Apparatus to capture volatile compounds (Franco & Rodriguez-Amaya, 1983).

balloon exit. The conditions were: vacuum suction, room temperature and 2 h of capture. The volatile extract captured in the porapak was separated using 300 μL methane dichloride (chromatography degree) and put into a flask. The flask was stored at 0 °C.

2.4. GC-mass spectral analyses

Each concentrated extract sample was analyzed through GC–MS, using a Shimazu, QP 500 system, with a DB –5 (30 m × 0.25 mm i.d. × 0.5 μm) column. Operating conditions of GC were as follows: helium was used as carrier gas at a constant flow rate of 1.0 mL/min; temperature of injector 240 °C and detector 230 °C and split 1:10. The volume injected was of 2 μL. Temperature programming was: 35–50 °C/6 min, 5 °C/min; 50–70 °C, 2 °C/min; 75–150 °C, 3 °C/min; 150–200 °C, 5 °C/min. The mass spectrum analyses were performed at 70 eV.

The volatile substances of guava fruits were identified, by means of comparison only, through mass spectrum, GC–MS database and Kovats index retention.

For the Kovats index, a standard mixture C_6 to C_{24} was used in the following conditions: initial temperature at 60 °C and programmed to rise 3 °C/min up to 240 °C.

3. Results and discussion

3.1. Non-volatiles analyses

Table 1 shows the results of color, pH, titratable acidity, total soluble solids, sugars and vitamin C in white guava fruits at different stages of maturity. The results are the average of three replications.

Table 1
Physical and chemical composition of guava at different stages of maturity

Parameters	Maturity stages		
	Immature	Intermediate	Mature
Color[a]			
L	53.83	64.05	71.87
a	−9.78	−2.99	4.23
b	22.25	28.90	32.83
C	24.30	29.05	33.10
h°	113.73	95.91	82.66
pH[a]	4.07	4.48	4.54
Titable acidity (% citric acid)[a]	0.39	0.46	0.44
Total soluble solid (°Brix)[a]	7.40	8.6	8.4
Sucrose (mg/100 g sample)[a]	10.57	11.43	11.37
Glucose (mg/100 g sample)[a]	22.50	15.16	14.13
Fructose (mg/100 g amostra)[a]	21.46	17.96	18.39
Vitamin C (mg ascorbic acid/100 g sample)[a]	76.80	145.35	168.36

[a] SAS® program statistic: *F* value and Tukey test, variance analysis, significance level 5% .The effect was significant ($P \leqslant 0.05$) for all the parameters shown in this table.

3.1.1. Fruit color

The color of white guava fruits was expressed by Hunter parameters (*L*, *a*, *b*, *C* and *h*°) as shown in Table 1. When data *a* and *b* were converted to chroma (*C*) the values became positive. The loss of green color and increase of yellow color is evidenced by the increase of values chroma from 24.3 to 33.10. *L* parameters increased from 53.83 to 71.87. The hue angle (*h*°) values obtained were: 113.73, 95.91 and 82.66 for immature, intermediate and mature fruits,

respectively. Statistic analysis of the results showed significant effect ($P \leqslant 0.05$) in the three different stages. These results are according to Mattiuz and Durigan (2001a) who studied guava fruits of the varieties "Pedro Sato" and "Paluma" stored at room temperature. Pereira (2003) and Brecht (1980) related that the color of fruits is associated with synthesis and degradation of pigments. Pereira (2003) showed that for white guava fruits the green color decreased along with the decrease of chlorophyll level and yellow color increased with the increase of carotenoid level.

3.1.2. pH and Titratable acidity

The pH increased slowly during the different maturity stages whereas titratable acidity increased in the immature and intermediary stage of maturation and decreased in the maturity stage. Increase in the pH and titratable acidity show the formation of organic acids during maturation. These increases on both parameters are associated with high concentration of undissociated organic acids, stored in the vacuole and the fruits use these acids as respiratory substrate (Medlicott & Jeger, 1987). Bashir, Abu-Goukh, and Abu-Bakr (2003) reported similar results to pink and white guava pulp. Titratable acidity increased from 0.15% to 0.19% of citric acid up to the climacteric peak and declined thereafter from 0.19% to 0.154% citric acid for white guava.

3.1.3. Total soluble solids

Total soluble solids increased during ripening (7.40–8.6 °Brix), but on the maturity stage decrease to 8.4 °Brix. This probably happened because of high consumption of sugars due to respiration rate (Sharaf & El-Saadany, 1996; Singh, Singh, & Chauan, 1981). Rodrigues, Agarwal, and Saha (1971) reported similar behavior for guava cultivar Sefeda. The authors showed a gradual increase of the content of soluble solids of guava fruit with maturation, except during the end of the growth period. According to El Bulk, Babiker, and El Tinay (1997) for different cultivars of guava total soluble solids content gradually increased with fruit development in all cultivars. The results obtained were: between 7.70, 6.20, 6.6 and 9.70 °B for Shambati, Pakistani, Shendi and Ganib cultivars, respectively, after first 15 days post harvest and 13.2, 11.1, 12.2 and 12.5 °B when the fruits were 126 days. The authors observed an increase in total soluble solids after 106 days.

3.1.4. Reducing sugar

Sucrose value increased slowly (10.57–11.37 mg/100 g sample), while glucose and fructose decreased during ripening, 22.50–14.13 mg/100 g sample and 21.46–18.39 mg/100 g sample, respectively. The expected results were decrease in the sucrose and increase of glucose and fructose, because each sucrose molecule would probably constitute a glucose and a fructose molecule. But a different effect was observed in this study. This probably happened either due to reduced percentage of sucrose in the fruit or due to the quick consumption of sugars, as explained earlier. Sharaf and El-Saadany (1996) showed decrease in the amount of glucose and fructose of guava during ripening. Of the total sugar, El Bulk et al. (1997) showed that in different

Table 2
Probable volatile compounds of guava at their three stages of maturity

	KI	Immature	Intermediate	Mature	MW	ID
Carbonyl compounds						
(*E*)-2-hexenal	854*	16.81	15.23	1.13	98	a, b, c
(*Z*)-3-hexenal	857*	7.96	1.11	0.86	98	a, b, c
Esters						
Ethyl hexanoate	996	2.8	0.34	1.75	144	a, b, c
trans-3-Hexenyl acetate	1003	nd	0.66	17.8	142	a, b, c
cis-3-Hexenyl acetate	1005	nd	nd	21.78	142	a, b, c
Methyl benzoate	1092	nd		2.08	136	a, c
Methyl octanoate	1123	2.6	0.54	1.35	158	a, c
3-Phenylpropyl-acetate	1368	nd	nd	2.74	178	a, b, c
Monoterpenes						
Limonene	1027	tr	nd	3.99	136	a, b, c
cis-Ocimene	1036	nd	nd	3.81	136	a, b, c
m-Cymenene	1083	nd	nd	1.04	132	a, b, c
Sesquiterpenes						
Caryophylene	1419	nd	tr	12.96	204	a, b, c
α-Humulene	1453	tr	tr	7.85	204	a, b, c
β-Acoradiene	1464	nd	nd	0.87	204	a, b, c
β-Bisabollene	1509	nd	0.30	5.46	204	a, b, c
β-Urcumene	1480	nd	nd	0.69	202	a, b, c
cis-p-Bisabollene	1495	nd	nd	0.78	204	a, b, c
trans-p-Bisabollene	1517	nd		0.87	204	a, b, c

KI: Kovats index, *Chyaui et al. (1992); MW: molecular weight; nd: not identified; tr: trace: <0.05; ID: identification [a] by GC/MS, [b] Kovats index and [c] literature McLfferty and Stauffer (1989).

cultivars of guava fruits the fructose represented around 20%; 38%; 37% and 41% for Shambati, Pakistani, Shendi and Ganib cultivars, respectively, while sucrose represented around 21%; 39%, 37% and 45% and glucose 59%, 23%, 25% and 14%, respectively. In this work, the data obtained were: fructose sugar presented proportion 40.3%; glucose 36.2% and sucrose 23.5%. These result show that the chemical composition of fruits change according to soil, climate, season and source.

3.1.5. Ascorbic acid

Ascorbic acid percentages (vitamin C) increased during ripening. In the immature fruits the amount was 76.8 mg ascorbic acid/100 g samples, during intermediate and

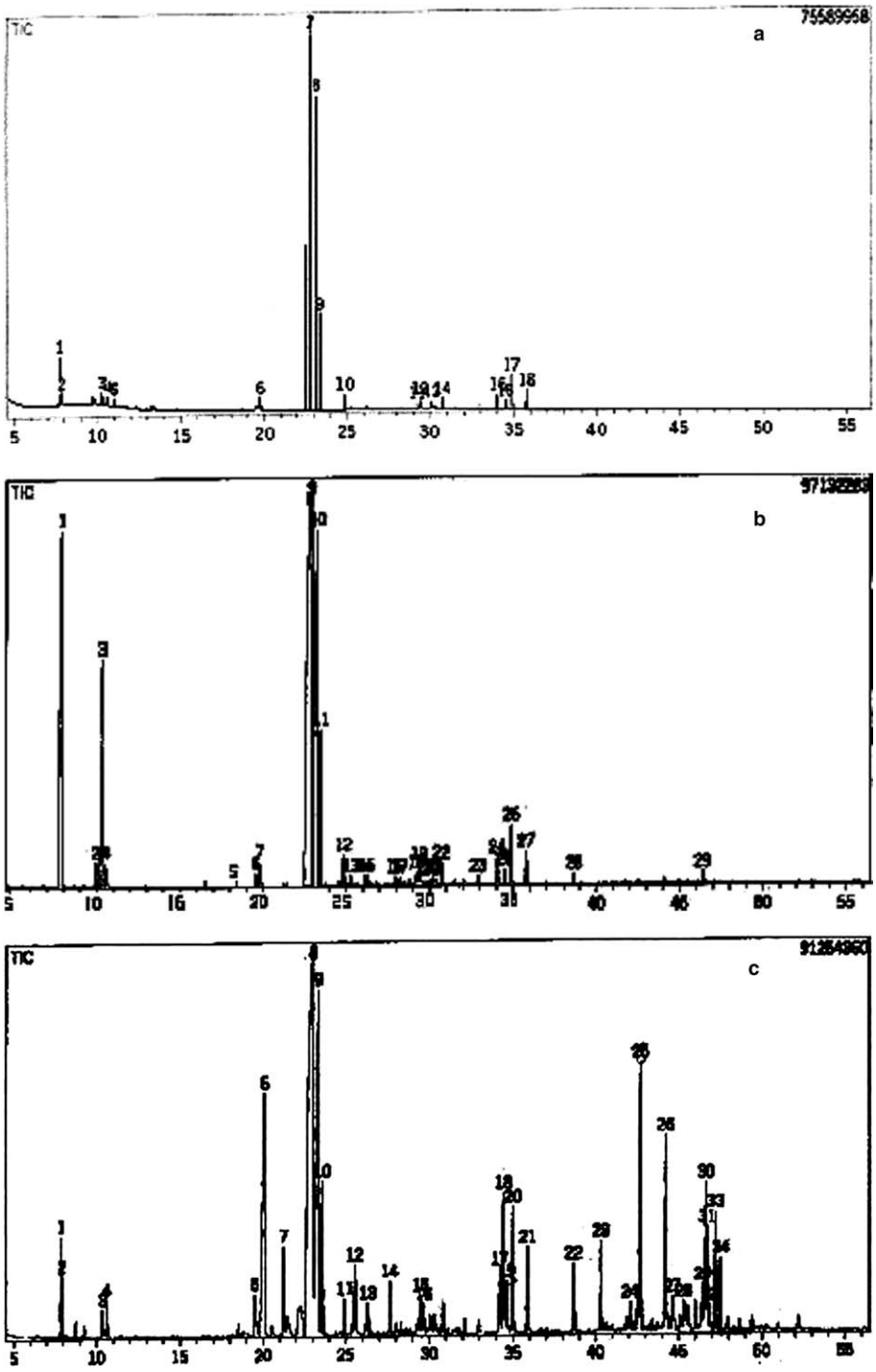

Fig. 2. (a) Volatiles compounds from guava in the immature stage; (b) volatile compounds from guava in the intermediary stage of maturity; (c) volatile compounds from guava in the maturity stage.

mature stages the amount of vitamin C was 126.21 and 168.36 mg ascorbic acid/100 g samples, respectively. High amount of ascorbic acid presented in the white guava in this study, showed that sugars were consumed during ripening for ascorbic acid synthesis. Studies show different behavior in the ascorbic acid content in the guava fruits. El-Zorkani, 1968 (cited by Bashir et al., 2003) reported that a maximum level of ascorbic acid was reached at the mature green stage and in the mature stage starts to decline rapidly. Bashir et al. (2003) reported that content ascorbic acid in pink and white guava of Sudan decreased along maturation process, from 86.3 to 75 mg/100 g fresh weight for white guava and 70 to 55 mg/100 g fresh weight for pink guava. Rathore (1976) showed that the amount of vitamin C in guavas stored at room temperature increased from 55.7 to 1014.4 mg ascorbic acid/100 g samples and Rodrigues et al. (1971) reported increase in the vitamin C content (ascorbic acid) during ripeninig period. These results are in agreement with the ones obtained in this study.

3.2. Volatile composition of guava during its three different stages of maturity

The identified compounds and quantitative distribution are represented in Table 2. The results were shown as average from triplicate of three different extractions from six fruits each. The chromatographic profile of fresh ripe guavas reveals that it contains 18 volatile compounds. The compounds were not confirmed through internal standards.

Fig. 2(a) shows the compounds of immature fruits. In this stage, the aldehyde compounds class is predominant. The major components are (*E*)-2-hexenal and (*Z*)-3-hexenal, 16.81% and 7.96%, respectively. The composition at this stage is typical of immature fruits or fruits that are in the physiologic maturity period.

Fig. 2(b) shows the chromatogram of volatile compounds of fruits in the intermediary stage of maturity, between physiological maturity and mature stage. The relative proportions of aldehydes (*E*)-2-hexenal and (*Z*)-3-hexenal decreased. Earlier studies have shown that aldehydes are compounds which are present in immature fruits and also work as compounds of impact in aroma of guava maturity (Chyaui, Chen, & Wu, 1992; Hatanaga, Kajiwara, & Sekiya, 1986). During this stage, the esters are present in low proportions.

Fig. 2(c) shows chromatogram of fruits in their maturity stage. Relative proportions of esters increased. *cis*-3-Hexenyl acetate and *trans*-3-hexenyl acetate are major compounds with 21.78% and 17.80%, respectively. These compounds are strongly related to the flavor of mature fruits (Hatanaga et al., 1986).

Another important class of compounds was the hydrocarbons sesquiterpenes. Within this group caryophyllene (12.96%), α-humulene (7.85%) and β-bisabollene (5.46%) are present. Caryophyillene is the major sesquiterpene hidrocarbon in the volatile compounds of mature fruits. These groups of compounds were identified in previous papers as constituents of guava fruits in the maturity (Chyaui et al., 1992; MacLeod & Troconis, 1982).

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References

Ali, Z. M., & Lazan, H. (1997). Guava. In S. K. Mitra (Ed.), *Post harvest physiology and storage of tropical and subtropical fruits* (pp. 145–165). New York: CABI Publishing.

Bashir, H. A., Abu-Goukh, A., & Abu-Bakr, A. (2003). Compositional changes during guava fruit ripening. *Food Chemistry, 80*, 557–563.

AOAC (1984). Official methods of analysis (14th ed., pp. 420–423). Washington, DC.

Benlloch, R., Fane, R., & Frigola, A. (1993). A quantitative estimate of ascorbic and isoascorbic acid by hight performance liquid Chromatography application to citric juices. *Journal of Liquid Chromatography, 16*(14), 3113–3122.

Brecht, P. E. (1980). Use of controlled atmospheres to retard deterioration of produce. *Food technology, 34*(3), 45–50.

Chyaui, C. C., Chen, S., & Wu, M. (1992). Differences of volatile and nonvolatile constituents between mature and ripe guava (*Psidium guajava* Linn) fruits. *Journal of Agricultural and Food Chemistry, 40*, 846–849.

El Bulk, R. E., Babiker, E. F. E., & El Tinay, A. H. (1997). Changes in chemical composition of guava fruits during development and ripening. *Food Chemistry, 59*(3), 395–399.

Franco, M. R. B., & Rodriguez-Amaya, D. B. (1983). Trapping of soursop (*Annona muricata*) juice volatiles on porapaq Q by suction. *Journal Science Agriculture, 34*, 293–299.

Hatanaga, A., Kajiwara, T., & Sekiya, J. (1986). Fatty acid hidroperoxide lyase in plant tissues: volatile aldehyde formation from linoleic and linolenic acid. In T. H. Parliment & R. Croteau (Eds.), *Biogeneration of aromas. ACS symposium series 317* (pp. 167–175). Washington, DC: American Chemical Society.

Macrae, R. (1988). *HPLC in food analyses*. Academic Press, 77p.

MacLeod, A. J., & Troconis, N. G. (1982). Volatiles flavor components of guava. *Phytochemistry, 40*(6), 1339–1342.

Manrique, G. D., & Lajolo, F. M. (2004). Cell-wall polysaccharide modifications during postharvest ripening of papaya fruit (*Carica papaya*). *Postharvest Biology and Technology, 33*, 11–24.

Mattiuz, B., & Durigan (2001a). Efeito de injúrias mecânicas na firmeza e coloração de goiabas das cultivares Paluma e Pedro Sato. *Revista Brasileira de Fruticultura, 23*(2), 277–282.

McGuire, R. G. (1992). Reporting of objective color measurements. *HortScience, 27*(12), 1254–1255.

McLfferty, F. W., & Stauffer, D. (1989). *The Wiley/NBS Registry of mass spectral data*. New York: Wiley.

Medlicott, A. P., & Jeger, M. J. (1987). The development and application of postharvest handing treatments to manipulate ripening mangoes. In R. T. Pinsley (Ed.), *Mangoes: a review* (pp. 56–77). London: Commonwealth Science Council.

Pereira, T. (2003). Conservação pós-colheita de goiaba (*Psidium guajava* L.) cv. Cortibel de polpa branca. Dissertação de tese de mestrado, Campos dos Goytacazes,UENF, RJ.

Rathore, D. S. (1976). Effect of season on the growth and chemical composition of guava (*Psidium guajava* L.) fruits. *The Journal of Horticultural Science, 51*(1), 41–47.

Rodrigues, R., Agarwal, P. C., & Saha, N. K. (1971). Physicochemical changes during development of safeda guava fruit. *Indian Food Packer, 25*, 5–20.

Sharaf, A., & El-Saadany, S. S. (1996). Biochemical studies on guava fruits during different maturity stages. *Annals of Agriculture Science, 24*(2), 975–984.

Singh, B. P., Singh, H. K., & Chauan, K. S. (1981). Effect of post-harvest calcium treatments on the storage life of guava fruits. *Indian Journal of Agricultural Science, New Delhi, 51*(1), 44–47.

Watada, A. E. (1986). Effects of ethylene on the quality of fruits and vegetables. *Food Technology, 40*(5), 82–85.