JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Gas Chromatographic−Olfactometric Characterization of Aroma Compounds in Two Types of Cashew Apple Nectar

M. FILOMENA VALIM,[†,‡] RUSSELL L. ROUSEFF,*[,‡] AND JIANMING LIN[‡,§]

Food and Nutrition Department, Faculty of Pharmaceutical Sciences, Sao Paulo State University, UNESP, Araraquara, SP, Brazil, and Citrus Research and Education Center, University of Florida, IFAS, 700 Experiment Station Road, Lake Alfred, Florida 33850-2299

Cashew apple nectar is a secondary product from the production of cashew nuts and possesses an exotic tropical aroma. Aroma volatiles in pasteurized and reconstituted (from concentrate) Brazilian cashew apple nectars were determined using GC-MS and split, time-intensity GC−olfactometry (GC-O)/GC-FID. Methional, (*Z*)-1,5-octadien-3-one, (*Z*)-2-nonenal, (*E*,*Z*)-2,4-decadienal, (*E*,*E*)-2,4-decadienal, β-damascenone, and δ-decalactone were identified for the first time in cashew apple products. These compounds plus butyric acid, ethyl 3-methylbutyrate, 2-methylbutyric acid, acetic acid, benzaldehyde, homofuraneol, (*E*)-2-nonenal, γ-dodecalactone, and an unknown were the most intense aroma volatiles. Thirty-six aroma volatiles were detected in the reconstituted sample and 41 in the pasteurized sample. Thirty-four aroma active components were common to both samples. Ethyl 3-methylbutyrate and 2-methylbutyric acid were character impact compounds of cashew apple (warm, fruity, tropical, sweaty). Using GC-pFPD, 2-methyl-3-furanthiol and bis(2-methyl-3-furyl) disulfide were identified for the first time in cashew apple. Both were aroma active (meaty).

KEYWORDS: GC-O; character impact compounds; ethyl 3-methylbutyrate; 2-methylbutyric acid; pasteurization; sulfur volatiles

INTRODUCTION

The cashew tree (*Anacardium occidentale* L.) is an indigenous Brazilian tree, found primarily in the northeastern coastal region. Its main product, the cashew nut, is widely appreciated and is the primary reason for its cultivation. The bottom portion of the cashew is a pseudofruit called the cashew apple. It has a pulpy and juicy composition, rich in vitamin C (262 mg/100 g of juice), and also contains sugars, tannins, and minerals (primarily calcium, iron, and phosphorus). Currently ~1,500,000 metric tons of cashews are grown in Brazil alone. However, only ~6% of the total cashew apple production is utilized because the producer has a guaranteed market for only the cashew nuts, the remainder is a waste disposal problem (*1*). The most important cashew apple byproduct is the pasteurized and concentrated juice, which is widely accepted in the Brazilian market. The concentrate is consumed diluted with water and sweetened.

Several groups have attempted to characterize cashew apple flavor by quantifying cashew apple volatiles. MacLeod and de Troconis (*2*) reported that terpenes comprised 38% of total volatiles (the largest group) in Venezuelan cashew apple juice. They concluded that hexanal, limonene, 3-carene, *trans*-hex-2-enal, and benzaldehyde were important odor quality components on the basis of their relative abundance in the juice extract. Later, Maciel et al. (*3*) reported that esters comprise the largest single volatile group in Brazilian cashew apple juice. These authors also reported the presence of sulfur compounds such as dimethyl sulfide, dimethyl disulfide, and dimethyl trisulfide. They also reported that the characteristic pungent-sour odor of cashew apple juice was due to isobutyric and isovaleric acids. Bicalho et al. (*4*) quantified the volatiles in Brazilian cashew apple juice. Ninety compounds were reported; 69 were identified and 62 quantified.

The present study was carried out to identify the aroma active components present in cashew apple nectars. Two types of nectars were evaluated: one had been processed by thermal pasteurization and the other reconstituted from thermal concentration. Because not all volatiles have aroma activity, the identification of aroma active components of cashew apple nectar will allow the determination of both positive and negative aroma attributes. Because the nectars were prepared using different technologies, the comparison of aroma active components should allow for the evaluation of processing effects on individual aroma components. This knowledge will permit the addition of cashew apple as an ingredient in soft and fruit drinks and yogurt and dairy-based beverages and thus increase the use of this underutilized product.

* Corresponding author [fax (863) 956-4631; e-mail rlr@lal.ufl.edu].
† Sao Paulo State University.
‡ University of Florida.
§ Present address: Firmenich Inc., M-3, 250 Plainsboro Rd., Plainsboro, NJ 08536.

10.1021/jf025738+ CCC: $25.00
Published on Web 01/14/2003

MATERIALS AND METHODS

Nectar Samples. Two commercial samples (multiple containers) of cashew apple nectars produced in Brazil were flown to the United States for analysis. The first sample type consisted of a pasteurized cashew nectar containing 12% juice that also contained sodium disulfite and sodium benzoate. The other sample was a nectar reconstituted from concentrate. Both nectars contained added sugars, were aseptically packaged in 200 mL laminated cartons, and were stored at ambient temperature.

Chemicals. Standards compounds were obtained as follows: Ethyl butyrate, dimethyl disulfide, butyric acid, ethyl 2-methylbutyrate, methional, 1-octen-3-ol, 2-methylbutyric acid, 1-octen-3-one, 5-methylfurfural, 4-hydroxy-2,5-dimethyl-3(2*H*)-furanone, 1-octanol, sotolon, homofuraneol, vanillin, γ-decalactone, (*E*)-2-nonenal, (*E*)-2-undecenal, (*E*,*E*)-2,4-nonadienal, (*E*,*E*)-2,4-decadienal, eugenol, benzaldehyde, phenylacetic acid, acetic acid, and ethyl 3-methylbutyrate were purchased from Aldrich (Milwaukee, WI). Ethyl isobutyrate, δ-decalactone, and γ-dodecalactone were purchased from Lancaster Synthesis (Pelham, NH). 2-Methyl-3-furanthiol and bis(2-methyl-3-furyl) disulfide were purchased from Acros Organics (Springfield, NJ). Hexanal, (*Z*)-3-hexenal, and β-damascenone were obtained as gifts from SunPure (Lakeland, FL). (*Z*)-2-Decenal was found in purchased (*E*)-2-decenal at 5−10% level, and (*Z*)-2-nonenal was found in purchased (*E*)-2-nonenal at a similar level. (*E*,*Z*)-2,4-Nonadienal and (*E*,*Z*)-2,4-decadienal were, respectively, present in the purchased (*E*,*E*)-2,4-nonadienal and (*E*,*E*)-2,4-decadienal, whereas *trans*-4,5-epoxy-(*E*)-2-decenal was found in an oxidized sample of (*E*,*E*)-2,4-decadienal. Their identities were confirmed by retention indices, odor quality, and MS spectral database comparison.

Aroma Extraction. Nectars (30 mL) were centrifuged for 10 min (2000*g*) to separate serum from pellet. Volatiles in serum (10 mL) were extracted with a 1:1 mixture of pentane and diethyl ether (10 mL) using a Mixxor-like apparatus consisting of two joined 50 mL syringes as described in Bazemore et al. (*5*). The aroma extracts were concentrated to 100 μL under a stream of dry nitrogen and analyzed immediately.

Solid Phase Microextraction (SPME). Aliquots of 10 mL of nectars were transferred to 40 mL glass vials with plastic screw caps and Teflon-coated septa containing a micro stirring bar and placed on a water bath at 40 °C. After an equilibrium time of 45 min, an SPME (Supelco Co., Bellefonte, PA) fiber (50/30 μm DVB/Carboxen/PDMS) was manually inserted into the headspace of the sample bottle. Exposure time was 30 min; after that, the SPME was inserted into the GC injection port at 200 °C and kept there for 3 min for desorption.

Gas Chromatography (GC). Volatile constituents were separated using an HP-5890A GC (Palo Alto, CA) with a flame ionization detector (FID) and either a Zebron ZB-5 column (30 m × 0.32 mm i.d. × 0.50 μm) from Phenomenex (Torrance, CA) or a DB-Wax column (30 m × 0.32 mm i.d. × 0.50 μm) from J&W Scientific (Folsom, CA). The oven temperature for the ZB-5 column was programmed from 40 to 265 °C at 7 °C/min with a holding time of 5 min; 0.5 μL of the concentrated extract was injected in the splitless mode. The injector temperature was maintained at 220 °C and the detector temperature at 250 °C. The initial oven temperature for the DB-Wax column was 40 °C, which was increased to 240 °C at 7 °C/min with a holding time of 5 min.

GC—Olfactometry (GC-O). The second FID base was modified into an olfactometer by DATU (Geneva, NY). A time-intensity approach was used to evaluate odor quality and intensity at the sniffing port under GC conditions described previously. Assessors rated aroma intensity (0−15 scale) continuously throughout the chromatographic separation process using a linear potentiometer. Retention times and verbal descriptors were recorded to permit aroma descriptors to be coupled with computerized aroma time-intensity plots. Two trained assessors evaluated the sample in duplicate, thus producing four individual time-intensity aromagrams. Average intensity from the four runs was calculated for each odorant; if no peak was detected, a value of zero was assigned. An averaged time-intensity aromagram was constructed by plotting average intensity versus retention time.

Chromatograms and aromagrams were recorded and integrated using CromoPerfect version 5.0.0, Justice laboratory software (Justice Innovations, Inc., Palo Alto, CA). Peaks were identified using Kovats retention indices (*6*), calculated using retention time data from a series of alkane standards (C_5−C_{25}) run under the same chromatographic conditions.

GC-pFPD. Sulfur compounds were separated using a DB-5 column (30 m × 0.32 mm i.d. × 0.50 μm) on an HP-5890A GC using the same oven temperature program as for the GC-O. Injections were carried out with a Gerstel (Baltimore, MD) CIS-3 temperature-programmable injector. The initial injector temperature was 60 °C, and the final injector temperature was 200 °C. A sulfur-specific pFPD, OI Analytical model 5380 pulsed flame photometric detector (pFPD) (OI Analytical Co., College Station, TX), was used to detect low-concentration sulfur compounds. The detector temperature was set at 250 °C, and the sulfur gate time was 6−24.9 ms.

GC-MS. Volatiles were separated and analyzed using a ThermoFinnigan GCQ (San Jose, CA) using a 60 m × 0.25 mm i.d. × 0.25 μm DB-5 column (J&W Scientific). The injector temperature and transfer line temperature were 200 and 250 °C, respectively. Headspace volatiles were concentrated and introduced into the chromatograph via SPME, and solvent-extracted volatiles were introduced through splitless injection. Helium was used as the carrier gas at 1 mL/min. The oven temperature program consisted of a single thermal gradient from 40 to 275 °C at 7 °C/min. The MS was set to scan from mass 40 to 300 at 2.0 scans/s in the positive electron impact mode. The ionization energy was set at 70 eV. NIST and Wiley databases were used for fragmentation spectrum identification.

RESULTS AND DISCUSSION

Identification of Aroma Active Compounds. In each sample, aroma activity was established by consensus of panel assessor GC-O time-intensity responses (minimum requirement, ≥50%). In this manner, 44 aroma active peaks were observed between both samples, 41 in the pasteurized cashew nectar, and 36 in the nectar reconstituted from concentrate. Aroma descriptors and linear retention index values on DB-5 and DB-Wax columns for these compounds are presented in **Table 1**. As shown in **Table 1**, 38 of the 44 aroma active compounds have been identified to different degrees. Twenty-seven of the 38 compounds were identified on the basis of aroma descriptors, standard retention indices, and MS spectral information. Four additional aroma compounds that contained sulfur (peaks 3, 9, 11, and 42; see **Table 1**) were identified on the basis of aroma descriptors, standard retention indices, and responses from the sulfur-specific pFPD detector. Information from sample compounds was further confirmed by comparing results with that from standard compounds. Seven compounds were tentatively identified on the basis of aroma descriptor and retention index data. GC-O of standard compounds supported these tentative identifications. Six aroma peaks remain unidentified.

Sensory Characteristics and the Origins of the Aroma Active Compounds. As shown in **Table 1**, many of the 44 aroma active compounds detected in the two samples have similar aroma quality. Therefore, they were grouped according to the similarity of their aroma descriptors.

Fruity, Floral. The 10 compounds in this aroma category are esters, lactones, or aldehydes. They include ethyl butyrate (**1**), ethyl isobutyrate (**2**), ethyl 3-methylbutyrate (**7**), ethyl 2-methylbutyrate (**8**), (*E*)-2-nonenal (**28**), γ-decalactone (**40**), δ-decalactone (**41**), γ-dodecalactone (**43**), and two compounds that remain to be identified. Ethyl butyrate (**1**), ethyl isobutyrate (**2**), and ethyl 2-methylbutyrate (**8**) are found in many fruits including apples (*7*) and citrus (*8*). γ-Dodecalactone (**43)** (fruity, floral sweet) and δ-decalactone (**41)** (coconut) were two of the more intense aroma active compounds in both samples. Together with γ-decalactone (**40)** (sweet fruit), they are probably responsible for the sweet, fruity tropical notes. They can be formed either

Table 1. Identification of Aroma Active Compounds in Pasteurized and Reconstituted from Concentrate Cashew Apple Nectars

peak[a]	compound	aroma descriptor	KI (DB-5)	KI (DB-Wax)	identification
1	ethyl butyrate	sweet, fruity	*b*	1057	LRI, AD, MS
2	ethyl isobutyrate	sweet, fruity	762	*c*	LRI, AD, MS
3	dimethyl disulfide	sulfur	785	1071	LRI, AD, pFPD
4	(*Z*)-3-hexenal[d]	green	795	nobsd	LRI, AD
5	hexanal	fishy, green	819	nobsd	LRI, AD, MS
6	butyric acid	cheesy, sweaty	834	1628	LRI, AD, MS
7	ethyl 3-methylbutyrate	cashew	839	1077	LRI, AD, MS
8	ethyl 2-methylbutyrate	sweet, fruity	846	1069	LRI, AD, MS
9	2-methyl-3-furanthiol	meaty	868	nobsd	LRI, AD, pFPD
10	acetic acid[d]	acid, sour	*c*	1449	LRI, AD
11	methional	potato	903	1463	LRI, AD, pFPD
12	1-octen-3-ol	mushroom	942	1438	LRI, AD, MS
13	2-methylbutyric acid	overripe fruit, sweaty, cashew	956	1667	LRI, AD, MS
14	1-octen-3-one	mushroom, metallic	976	1305	LRI, AD, MS
15	5-methylfurfural	caramel, burnt sugar	978	1560	LRI, AD, MS
16	(*Z*)-1,5-octadien-3-one	geranium, metallic, green	983	1312	LRI, AD, MS
17	benzaldehyde	burnt sugar, almond	996	1525	LRI, AD, MS
18	unknown	plastic	1032	nobsd	LRI, AD
19	4-hydroxy-2,5-dimethyl-3(*2H*)-furanone (Furaneol)	caramel, burnt sugar	1064	2039	LRI, AD, MS
20	unknown	sweet, fruity	1067	nobsd	LRI, AD
21	1-octanol	metallic, sulfur	1080	1557	LRI, AD, MS
22	unknown	rancid	1096	nobsd	LRI, AD
23	sotolon[d]	seasoning, mushroom	1113	2203	LRI, AD
24	unknown	mushroom, moldy	*f*	1996	LRI, AD
25	unknown	green pepper seed	1132	nobsd	LRI, AD
26	homofuraneol	caramel, burnt sugar	1139	2070	LRI, AD, MS
27	(*Z*)-2-nonenal	geranium, metallic	1149	1510	LRI, AD, MS
28	(*E*)-2-nonenal	soapy, floral, sweet	1162	1538	LRI, AD, MS
29	(*E*,*Z*)-2,4-nonadienal	geranium, pungent	1196	nobsd	LRI, AD, MS
30	(*E*,*E*)-2,4-nonadienal	fatty, fried	1217	1709	LRI, AD, MS
31	(*Z*)-2-decenal	geranium, floral	1250	1627	LRI, AD, MS
32	phenylacetic acid[d]	sweet, honey, floral	1274	2574	LRI, AD
33	(*E*,*Z*)-2,4-decadienal	geranium, metallic	1297	1758	LRI, AD, MS
34	(*E*,*E*)-2,4-decadienal	fatty, fried, pungent	1319	1832	LRI, AD, MS
35	eugenol[d]	clove, spicy	*g*	2141	LRI, AD
36	(*E*)-2-undecenal	geranium, metallic, pungent	1365	1758	LRI, AD, MS
37	*trans*-4,5-epoxy-(*E*)-2-decenal[d]	green, metallic	1380	2012	LRI, AD
38	β-damascenone	honey, sweet, fruity	1391	1832	LRI, AD, MS
39	vanillin	vanilla, sweet	1410	2589	LRI, AD, MS
40	γ-decalactone	dried fruity, sweet	1472	nobsd	LRI, AD, MS
41	δ-decalactone	sweet, coconut	1503	2216	LRI, AD, MS
42	bis(2-methyl-3-furyl) disulfide	meaty	1542	*h*	LRI, AD, pFPD
43	γ-dodecalactone[d]	sweet, floral, fruity	1685	2399	LRI, AD
44	unknown	dried fruity	1697	nobsd	LRI, AD

[a] Elution order on DB-5 column. Compounds were identified by comparison of aroma descriptor (AD) and linear retention index (LRI) on DB-5 and DB-Wax columns with standard compounds and mass spectrometry (MS). [b] Coelutes with hexanal. [c] Elutes with solvent peak. [d] Tentative identification. [e] nobsd, not observed. [f] Coelutes with sotolon. [g] Coelutes with (*E*)-2-undecenal. [h] Coelutes with 1-octen-3-one.

from δ- and γ-hydroxy fatty acids or triglycerides by heating (*9*) or from an in vivo biosynthetic pathway.

Meaty, Sweaty, Sulfur. The seven compounds in this aroma category include dimethyl disulfide (**3**), butyric acid (**6**), 2-methyl-3-furanthiol (**9**), methional (**11**), 2-methylbutyric acid (**13**), and bis(2-methyl-3-furyl) disulfide (**42**). The primary sources for the sulfur-containing compounds in this group are from the degradation of the sulfur-containing amino acids, methionine and cysteine. Methional (**11**) (potato aroma) is reported here for the first time as one of the major aroma active compounds in thermally processed cashew apple nectar. Cashew apple has a high amount of vitamin C, 150−310 mg/100 g (*1*). Therefore, the dehydroascorbic acid, which could be expected to be present, could provide the dicarbonyl source to react with methionine to form methional via Strecker degradation (*10*).

Green, Metallic, Mushroom, Fatty. This category contained the largest number of aroma compounds, 17, and included (*Z*)-3-hexenal (**4**), hexanal (**5**), 1-octen-3-ol (**12**), 1-octen-3-one (**14**), (*Z*)-1,5-octadien-3-one (**16**), 1-octanol (**21**), (*Z*)-2-nonenal (**27**), (*E*,*Z*)-2,4-nonadienal (**29**), (*E*,*E*)-2,4-nonadienal (**30**), (*Z*)-2-decenal (**31**), (*E*,*Z*)-2,4-decadienal (**33**), (*E*,*E*)-2,4-decadienal (**34**), eugenol (**35**), (*E*)-2-undecenal (**36**), *trans*-4,5-epoxy-(*E*)-2-decenal (**37**), and two unknowns. These mono- and double-unsaturated fatty aldehydes, vinyl ketones, an unsaturated aldehyde, and an epoxy unsaturated aldehyde are typically found in lipid oxidation flavors. Oleic, palmitic, and linolenic fatty acids are the predominant fatty acids in Brazilian cashew apple pulp (*11*). The presence of (*Z*)-2-decenal (**31**) is indicative of the oxidation of oleic acid, which is the major fatty acid present in Brazilian cashew apple pulp. Both hexanal (**5**) and 2,4-decadienal (**33**, **34**) are the expected oxidation products of linoleic acid (*11*, *12*). (*E*,*E*)-2,4-Decadienal (**34**) can be further oxidized to *trans*-4,5-epoxy-(*E*)-2-decenal (**37**). 1-Octen-3-one (**14**) (mushroom, metallic-like) has been shown to be enzymatically derived from linoleic acid (*13*).

Sweet, Burnt Sugar, Caramel. The eight compounds in this aroma category include 5-methyl-furfural (**15**), benzaldehyde (**17**), 4-hydroxy-2,5-dimethyl-3(2*H*)-furanone (Furaneol) (**19**), sotolon (**23**), homofuraneol (**26**), phenylacetic acid (**32**), β-damascenone (**38**), and vanillin (**39**). The presence of 4-hydroxy-

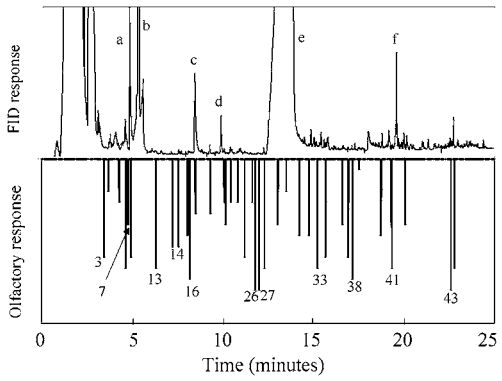

Figure 1. Chromatogram (top) and aromagram (bottom, inverted) of a pasteurized nectar extract containing sodium benzoate as a preservative on a 5% phenyl column. Peak: a, furfural; b, ethyl 2-methylbutyrate; c, benzaldehyde; d, limonene; e, benzoic acid; f, ethyl benzoate. Numbers in aromagram correspond to components in **Table 1**. See text for chromatographic conditions.

2,5-dimethyl-3(2*H*)-furanone (Furaneol) (**19**) has been reported in thermally treated fruit and juices, such as orange juice (*14*), grape juice (*15*), tomato (*16*), mango (*17*), and lychee (*18*).

Other furanones such as sotolon (**23**), homofuraneol (**26**), and 5-methylfurfural (**15**) are most likely Maillard reaction products. β-Damascenone (**38**), one of the most intense aroma active compounds in either nectar, is a degradation product of β-carotene and other carotenoids such as neoxanthin. β-Carotene is one of the major carotenoids in cashew apple juice (*19*).

The observed phenylacetic acid (**32**) is probably a metabolic product derived from the Strecker reaction with phenylalanine. The Strecker aldehyde, phenylacetaldehyde, has been reported to be oxidized to the corresponding acid during thermal treatments (*20*). In this study, the aroma intensity of phenylacetic acid was higher in the pasteurized nectar compared to the reconstituted nectar.

Instrumental versus Olfactory Detection. As shown in **Figure 1** the chromatogram on the top and the aromagram (inverted) on the bottom contain very different information. The sample shown is a pasteurized nectar containing added sodium benzoate (as a preservative) separated on a 5% phenyl column. The largest FID peak corresponds to benzoic acid (the predominant form of sodium benzoate in the acidic juice). The MS total ion current chromatogram and the FID chromatogram were similar as the FID detector is essentially a mass detector. It is worth noting that the components that are present in highest concentration, for example, benzoic acid, produce no olfactory response. The small olfactory peaks beneath the large benzoic acid correspond to other coeluting components. (See **Table 1** for their identification.) Many of the major FID peaks such as peaks d, e, and f (limonene, benzaldehyde, and ethyl benzoate, respectively) exhibit little to no olfactory activity. Conversely, some of the most intense olfactory responses were found in regions with little FID activity, for example, peaks 11, 17, 27, 28, 39, and 42. The aromagram contains only those components that have aroma activity, which requires only that compounds be present at concentrations that exceed their olfactory threshold. The olfactory thresholds for compounds corresponding to peaks 11, 17, 27, 28, 39, and 42 are obviously extremely low as their analytical concentrations are so low they produce little FID signal. As an example, peak 11, methional, has an olfactory threshold of 0.2 ppb ng/L in water (*21*). Therefore, studies that utilized only mass detectors (such as MS and FID) would not have detected many of the most intense aroma volatiles, because so many of them are found at such low concentrations.

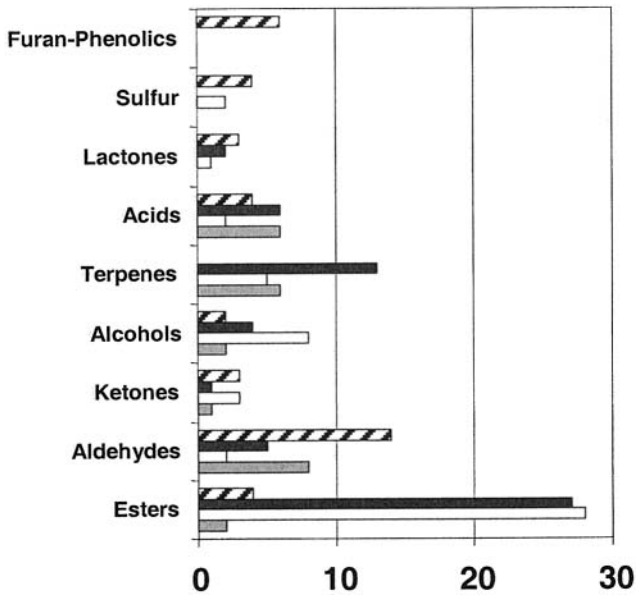

Figure 2. Comparison of reported volatiles in cashew apple according to chemical functional group and cited reference: current study (slashed line bar), Bicalho, 2000 (■), Maciel, 1986 (□) and MacLeod, 1982 (gray shaded bar).

Comparison with Previous Studies. As shown in **Figure 2** there is a wide variation in the number and kinds of aroma volatiles observed in cashew apple samples. These differences can be attributed to a number of factors including differences in chromatographic resolution, cultivar differences, fresh versus processed samples, and, perhaps most importantly, detection and sample preparation differences.

Extraction and concentration procedure differences used in the various studies undoubtedly contributed to the lack of qualitative agreement. The 4 h Likens−Nickerson extraction employed by MacLeod and de Troconis (*2*) may have created thermally induced artifacts. This extraction technique is rarely used today because the long exposure to elevated temperatures has been shown to produce artifacts and degradation of aroma active compounds (*22*). The 4 h simultaneous distillation extraction employed by Bicalho et al. (*4*) also may have induced artifacts for the same reason. In contrast, Maciel et al. (*3*) employed a dynamic headspace purge and trap using Tenax with thermal desorption. In the current study, room temperature liquid−liquid extraction was used to remove volatiles from the sample matrix. A portion of volatiles with boiling points near that of the solvents employed may be lost during the concentration process. As employed in this study, these losses can be minimized by gentle evaporation with a stream of nitrogen and by employing low boiling solvents.

Chromatographic resolution also varied among the cited studies. MacLeod and de Troconis (*2*) employed wide-bore (4 mm) packed columns, whereas all of the other studies employed high-resolution capillary columns. Therefore, it is not surprising to see that they reported the fewest number of volatiles (see **Figure 2**).

GC-MS has been the primary measurement tool in all prior cashew apple studies, whereas in the current study GC-O is the primary measurement tool, which utilizes the sensitivity and selectivity of the human nose. In the current study, GC-MS is used primarily to confirm human sensory data when the component is present within detection limits of the MS. MacLeod and de Troconis (*2*) reported that terpenes were quantitatively the predominant group of volatiles in cashew

apple, followed by aldehydes. They listed the important odor quality components as hexanal, car-3-ene, limonene, (*E*)-2-hexenal, and benzaldehyde and also suggested that benzaldehyde might make a relevant contribution to cashew apple flavor.

The number and kinds of esters present in cashew apple are also disputed. Maciel et al. (*3*) and Bicalho et al. (*4*) both reported over 20 esters, whereas MacLeod and de Troconis (*2*) report only 2 esters. The latter finding can be explained by the limited chromatographic resolution available when this study was conducted. In the current study, 5 esters were found to have aroma activity, but 7 others were identified via mass spectrometry that lacked aroma activity.

Maciel et al. (*3*) reported that esters were the predominant compounds, which contributed to the sweet, fruity flavor character of cashew apple. Olfactory impressions emanating from the GC were noted but not associated with individual aroma compounds. They concluded that methyl and ethyl butyrate, ethyl isovalerate, and isovaleric and isobutyric acids were responsible for the characteristic flavor of cashew apple. Our GC-O studies support some of their findings and disagree with others. 2-Methylbutyric acid (**13**) and ethyl isovalerate (ethyl 3-methylbutyrate, **7**) were definite cashew apple character impact compounds. As seen from the intensity values in **Table 1**, these three character impact compounds are not only character impact compounds but also among the most potent. Of the other two compounds mentioned in the earlier study, ethyl butyrate had limited aroma activity but isobutyric acid had no aroma activity in our samples.

Due to the unique selectivity and sensitivity of the human nose over 20 aroma compounds are reported in cashew apple products for the first time. Newly reported compounds include (*Z*)-2-nonenal (**27**), (*E*,*Z*)-2,4-nonadienal (**29**), (*E*,*E*)-2,4-nonadienal (**30**), (*Z*)-2-decenal (**31**), (*E*,*Z*)-2,4-decadienal (**33**), (*E*,*E*)-2,4-decadienal (**34**), (*E*)-2-undecenal (**36**), sotolon [4,5-dimethyl-3-hydroxy-2(5*H*)-furanone, **23**], β-damascenone (**38**), 2-methyl-3-furanthiol (**9**), methional (**11**), (*Z*)-1,5-octadien-3-one (**16**), homofuraneol [2(or 5)-ethyl-4-hydroxy-5(or 2)-methyl-3(2*H*)-furanone, **26**], 4-hydroxy-2,5-dimethyl-3(2*H*)-furanone (Furaneol) (**19**), vanillin (**39**), 1-octen-3-one (**14**), 1-octen-3-ol (**12**), *trans*-4,5-epoxy-(*E*)-2-decenal (**37**), bis(2-methyl-3-furyl) disulfide (**42**), and δ- and γ-decalactone (**40**, **41**). Most of these compounds are at low concentrations but possess extremely low aroma thresholds. These potent, low-level compounds are difficult to identify using traditional FID or MS TIC detectors. Several of these newly reported compounds such as 2-methyl-3-furanthiol (**9**), methional (**11**), 4-hydroxy-2,5-dimethyl-3(2*H*)-furanone (**19**), and vanillin (**39**) may be products of the thermal processes used to produce these commercial products.

Prior to this study, the only sulfur volatiles previously reported were dimethyl disulfide and dimethyl trisulfide (*3*). Using the highly sensitive, sulfur-selective pFPD detector, it was possible to identify additional sulfur volatiles that produced no corresponding FID peaks because their levels were so low. Neither 2-methyl-3-furanthiol (**9**) nor bis(2-methyl-3-furyl) disulfide (**42**) had been previously reported in cashew apple. These compounds displayed aroma activity of low and moderate intensity, respectively, and were described as meaty-smelling.

Aroma Differences between Nectar Products. As expected, fewer aroma volatiles (**36**) were found in the samples reconstituted from concentrate than in the pasteurized sample (**41**) because most of the aroma volatiles are typically removed along with the water during thermal concentration. Products reconstituted from concentrate typically have a flavor package added to restore many of the aroma components lost during the concentration process. These flavor packages are typically produced from a portion of the condensate from the first stage of the evaporator. Thus, two of the three aroma impact compounds [ethyl 3-methylbutyrate (**7**) and 2-methylbutyric acid (**13**)] are found at similar or higher levels in the reconstituted product. However, as seen in **Figure 3**, many of the low-boiling (early-eluting) compounds such as dimethyl disulfide (**3**), hexanal (**5**), and butyric acid (**6**) are missing in the product from concentrate because they would be the most difficult to condense and trap.

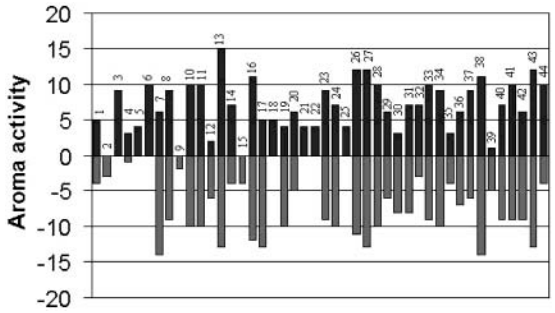

Figure 3. Aroma active components of pasteurized cashew nectar (top) and from concentrate cashew nectar (bottom). Peak numbers refer to the same numbered compounds in **Table 1**. Aroma intensity from 0 to 5 represents background components, 5–10 represents secondary components, and 10–15 represents primary aroma components.

Of the 44 aroma active compounds found between the two sample types, 33 were common to both. **Figure 3** compares the average aroma intensity from consensus aromagrams for pasteurized cashew nectar (top) and reconstituted from concentrate cashew nectar (inverted bottom). It is interesting to note that many aroma components were found in strikingly similar proportions in both types of products. However, several compounds are found only, or at greatly enhanced levels, in the product from concentrate. Because this product has been heated twice, once during concentration and once after being reconstituted, it is likely these are thermally induced compounds.

There are differences in the most intense aroma peaks from the consensus aromagrams between the two types of products because of the heating and volatile restoration differences previously discussed. The most potent aroma active compounds (>10) in pasteurized cashew nectar included butyric acid (**6**), acetic acid (**10**), methional (**11**), 2-methylbutyric acid (**13**), (*Z*)-1,5-octadien-3-one (**16**), homofuraneol (**26**), (*Z*)-2-nonenal (**27**), (*E*)-2-nonenal (**28**), (*E*,*Z*)-2,4-decadienal (**33**), β-damascenone (**38**), δ-decalactone (**41**), γ-dodecalactone (**43**), and an unknown compound (dried fruit-like aroma). The most potent aroma active compounds found in reconstituted from concentrate cashew nectar included ethyl 3-methylbutyrate (**7**), acetic acid (**10**), methional (**11**), 2-methylbutyric acid (**13**), (*Z*)-1,5-octadien-3-one (**16**), benzaldehyde (**17**), homofuraneol (**26**), (*Z*)-2-nonenal (**27**), (*E*)-2-nonenal (**28**), (*E*,*E*)-2,4-decadienal (**34**), β-damascenone (**38**), and γ-dodecalactone (**43**). These compounds accounted for 46 and 52.6%, respectively, of the total aroma of pasteurized cashew nectar and reconstituted from concentrate cashew nectar. Compounds with slightly less intensity (aroma activity of 9) include dimethyl disulfide (**3**), ethyl 2-methylbutyrate (**8**), sotolon (**23**), and (*E*,*E*)-2,4-decadienal (**34**) in pasteurized cashew nectar. In the reconstituted from concentrate sample, ethyl 2-methylbutyrate (**8**), sotolon (**23**), (*E*,*Z*)-2,4-decadienal (**33**), γ-decalactone (**40**), and δ-decalactone (**41**) produced similar aroma intensities.

Because the nectar reconstituted from concentrate has undergone two thermal processes compared to the single heat

treatment in pasteurization, it should not be surprising to find thermally generated aroma compounds such as 4-hydroxy-2,5-dimethyl-3(2*H*)-furanone (**19**), β-damascenone (**38**), and vanillin (**39**) in higher concentration in this product. Other sulfur-based volatiles such as 2-methyl-3-furanthiol (**9**) and its dimer bis(2-methyl-3-furyl) disulfide (**42**) can also be considered to be thermal degradation products (*23*).

Unfortunately, three of the four aroma volatiles found only in the pasteurized nectar remain to be identified.

ACKNOWLEDGMENT

We thank O-I Analytical Co. for the use of the pFPD detector and Dr. Laura Chambers for helpful comments.

NOTE ADDED AFTER ASAP

This article was released ASAP on 1/14/03 before final corrections were made. The incorrect version of Figure 2 appeared instead of the correct one. The correct version was posted 1/16/03.

LITERATURE CITED

(1) Maia, G. A. Production and processing of tropical fruit juices from Brazil. *Proceedings of the 23rd IFU Symposium*, Havana, Cuba, Feb 7−13, 2000; International Federation of Fruit Juice Producers: Paris, 2000; pp 128−139.

(2) MacLeod, A. J.; de Troconis, N. G. Volatile Flavor Components of Cashew Apple (*Anacardium occidentale*). *Phytochemistry* **1982**, *21*, 2527−2530.

(3) Maciel, M. I.; Hansen, T. J.; Aldinger, S. B.; Labows, J. N. Flavor Chemistry of Cashew Apple Juice. *J. Agric. Food Chem.* **1986**, *34*, 923−927.

(4) Bicalho, B.; Pereira, A. S.; Neto, F. R. A.; Pinto, A. C.; Rezende, C. M. Application of High-Temperature Gas Chromatography−Mass Spectrometry to the Investigation of Glycosidically Bound Components Related to Cashew Apple (*Anacardium occidentale* L. Var. Nanum) Volatiles. *J. Agric. Food Chem.* **2000**, *48*, 1167−1174.

(5) Bazemore, R.; Goodner, K.; Rouseff, R. Volatiles from Unpasteurized and Excessively Heated Orange Juice Analyzed with Solid Phase Microextraction and GC-Olfactometry. *J. Food Sci.* **1999**, *64*, 800−803.

(6) Kovats, E. *Advances in Chromatography*; Dekker: New York, 1965; pp 229−247.

(7) Lopez, M.; Lavilla, T.; Graell, J.; Recasens, I.; Vendrell, M. Effect of different CA conditions on aroma and quality of golden delicious apples. *J. Food Qual.* **1999**, *22*, 583−597.

(8) Buettner, A.; Schieberle, P. Evaluation of Aroma Differences between Hand-Squeezed Juices from Valencia Late and Navel Oranges by Quantitation of Key Odorants and Flavor Reconstitution Experiments. *J. Agric. Food Chem.* **2001**, *49*, 2387−2394.

(9) Forss, D. A. Mechanisms of formation of aroma compounds in milk and milk products. *J. Dairy Res.* **1979**, *46*, 691−706.

(10) Tressl, R. H.; Helak, B.; Martin, N.; Kersten, E. Formation of amino acid specific Maillard products and their contribution to thermally generated aromas. In *Thermal Generation of Aromas*; Parliament, R. J., Ho, C.-T., Eds.; American Chemical Society: Washington, DC, 1989.

(11) Maia, G. A.; Brown, W. H.; Whiting, F. M.; Stull, J. W. Cashew fatty acids. *HortScience* **1975**, *10*, 233−234.

(12) Tressl, R. B.; Bahri, D.; Kossa, M. Formation of off-flavor components in beer. In *The Analysis and Control of Less Desirable Flavors in Foods and Beverages*; Charalambous, G., Ed.; Academic Press: New York, 1980; pp 293−318.

(13) Josephson, D. B.; Lindsay, R. C. Enzymatic generation of volatile aroma compounds from fresh fish. In *Biogeneration of Aroma*; Parliament, R., Ed.; American Chemical Society: Washington, DC, 1986; pp 201−219.

(14) Tatum, J. H.; Nagy, S.; Berry, R. E. Degradation Products Formed in Canned Single-Strength Orange Juice During Storage. *J. Food Sci.* **1975**, *40*, 707−709.

(15) Baek, H. H.; Cadwallader, K. R.; Marroquin, E.; Silva, J. L. Identification of Predominant Aroma Compounds in Muscadine Grape Juice. *J. Food Sci.* **1997**, *62*, 249−252.

(16) Buttery, R. G.; Takeoka, G. R.; Ling, L. C. Furaneol—Odor Threshold and Importance to Tomato Aroma. *J. Agric. Food Chem.* **1995**, *43*, 1638−1640.

(17) Sakho, M.; Chassagne, D.; Crouzet, J. African Mango Glycosidically Bound Volatile Compounds. *J. Agric. Food Chem.* **1997**, *45*, 883−888.

(18) Ong, P. K. C.; Acree, T. E. Gas Chromatography Olfactory Analysis of Lychee (*Litchi chinesis* Sonn.). *J. Agric. Food Chem.* **1998**, *46*, 2282−2286.

(19) Cecchi, H. M.; Rodriguezamaya, D. B. Carotenoid Composition and Vitamin-A Value of Fresh and Pasteurized Cashew-Apple (*Anacardium occidentale* L) Juice. *J. Food Sci.* **1981**, *46*, 147−149.

(20) Hofmann, T.; Münch, P.; Schieberle, P. Quantitative Model Studies on the Formation of Aroma-Active Aldehydes and Acids by Strecker-Type Reactions. *J. Agric. Food Chem.* **2000**, *48*, 434−440.

(21) Guadagni, D. G.; Buttery, R. G.; Turnbaugh, J. G. Odor thresholds and similarity ratings of some potato chip components. *J. Sci. Food Agric.* **1972**, *23*, 1435−44.

(22) Maignial, L.; Pibarot, P.; Bonetti, G.; Chaintreau, A.; Marion, J. P. Simultaneous distillation-extraction under static vacuum: isolation of volatile compounds at room temperature. *J. Chromatogr.* **1992**, *606*, 87−94.

(23) Lin, J.; Rouseff, R.; Barros, S.; Naim, M. Aroma Composition Changes in Early Season Grapefruit Juice Produced from Thermal Concentration. *J. Agric. Food Chem.* **2002**, *50*, 813−819.

Received for review June 16, 2002. Revised manuscript received November 5, 2002. Accepted November 10, 2002. This research was supported by the Florida Agricultural Experiment Station and approved for publication as Journal Series R09215.

JF025738+