Aromatic Profile of Aqueous Banana Essence and Banana Fruit by Gas Chromatography–Mass Spectrometry (GC-MS) and Gas Chromatography–Olfactometry (GC-O)

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Gas chromatography—mass spectrometry (GC-MS) and gas chromatography—olfactometry (GC-O) were used to determine the aromatic composition and aroma active components of commercial banana essence and fresh banana fruit paste. Totals of 43 and 26 compounds were quantified in commercial banana essence and fresh banana fruit paste, respectively. Five new components in commercial banana essence were identified as methyl butyrate, 2,3-butanediol diacetate, 2-hydroxy-3-methyl-ethylbutyrate, 1-methylbutyl isobutyrate, and ethyl 3-hydroxyhexanoate. A total of 42 components appear to contribute to the aromatic profile in banana. Isoamyl acetate, 2-pentanol acetate, 2-methyl-1-propanol, 3-methyl-1-butanol, 3-methylbutanal, acetal, isobutyl acetate, hexanal, ethyl butyrate, 2-heptanol, and butyl butyrate had high concentrations and were most detected by GC-O panelists in the commercial banana essence. Volatile components found only in fresh banana fruit paste that were detected by aroma panelists include E-2-hexenal, limonene, and eugenol.

Keywords: Flavor; aroma; GC-MS; GC-O; volatile; banana essence; banana fruit

INTRODUCTION

Banana fruit has a pleasant flavor and is widely consumed throughout the world. Analytical research on the aroma compounds in this fruit has been carried out for the past 40 years. Many authors have studied the aromatic profile of this fruit using different analytical methods (1-5). As a result of these studies, 152 components have been identified, and the principal aroma compounds found in banana were pentan-2-one and isoamyl and isobutyl esters and other uncommon esters of pentan-2-ol, hexan-2-ol, hept-4(Z)-en-2-ol, oct-4(Z)en-1-ol, and oct-5(Z)-en-1-ol (5). Shiota (5) asserted that these compounds were the probable cause for the fruity ester aroma found in bananas. A study carried out by Jordán et al. (6) identified a total of 45 components in banana essence. These authors determined the relative abundance of these compounds and concluded that 3-methyl-1-butanol, 2-methyl-1-butanol, 1-butanol-3methyl acetate, 2-pentanone, and 1-butanol were the components found in highest relative abundance.

However, the real role of each component in the aromatic quality of the fruit can only be known by the study of their aroma activity. One of the biggest problems in the study of aroma is the selection of those components that have a real contribution to the flavor of food (7). The term odor activity value (OAV) was defined by Rothe and Thomas (8) a few years after the introduction of gas chromatography by James and Martin in 1952 (9).

Gas chromatography-olfactometry (GC-O) analysis has proved to be a powerful way of determining key

aroma compounds in wine research (10). In general, it is quite difficult to judge the sensorial relevance of one compound directly from GC-O. For this reason during the past 40 years, different techniques have been developed to determine the sensorial contribution of each component to the aroma of foods. Aroma extraction dilution analysis (AEDA) (11), CharmAnalysis (12), and Osme (13) are the methods most used in GC-O analysis. These techniques have been widely applied to determine the relevance of the volatile components in food by various researchers (10, 14–16). So far, this technique has not been applied to the aromatic profile of banana.

An integrated approach involving the joint determination of the volatile compounds in banana fruit and banana essence by gas chromatography-mass spectrometry (GC-MS) and the GC-O profile was therefore undertaken to provide useful information about the most important active contributors to the aroma of banana.

MATERIALS AND METHODS

Fresh Banana Fruit Paste. Mature banana fruits (*Musa sapientum* L. var. Cavendish) from Honduras at the seventh stage of ripeness were purchased from a local supermarket in Winter Haven, FL.

Commercial Banana Essence. Essence samples were obtained from a local processor in Florida. The aqueous essences were obtained from a whole fruit puree from mature banana fruits (*M. sapientum* L. var. Cavendish) from Honduras at the seventh stage of ripeness using a low-temperature short-time steam distillation process. The source and condition of the banana for processing were the same as for the fruit analyzed.

Extraction of Volatile Compounds. *Fresh Banana Fruit Paste.* The complete fresh fruit was finely sliced, and 120 g was pureed at room temperature for ~ 1 min in a blender to obtain a homogeneous paste. Volatile compounds present in the puree were extracted with 80 mL of methylene chloride

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(Sigma Chemical Co., St. Louis, MO) by stirring for 1 h with a magnetic stirrer at ~2 °C. The resulting mixture was centrifuged for 10 min (5000 rpm, 3000*g*), and the organic layer was dried with sodium sulfate and concentrated to 0.1 mL using a distillation-rectification system. For the concentration of the sample a Vigreux column (Vidra-Foc E-19874) was inserted between the heater and the condenser. The temperature of the sample was held between 35 and 36 °C by the application of vacuum, and the condenser temperature was held at -13 °C. Cyclohexanone at 10 ppm was used as internal standard and was added to the sample before extraction with the solvent.

Commercial Banana Essence. Isolation of volatile compounds was made using a liquid extraction technique. Volatile components present in 3 mL of essence with cyclohexanone at 10 ppm were extracted twice using 2 mL of methylene chloride for each extraction, mixed between two connected syringes, and combined. The final emulsion was treated under the same conditions as the banana paste and subsequently concentrated to 0.1 mL using a flow of nitrogen. The extraction efficiency for the commercial banana essence and fresh banana fruit paste was 98%, as determined from the recovery of the volatile components in three successive extractions.

GC-MS Analysis. Qualitative and quantitative analyses of the volatile compounds were made using an Agilent model 5973N MSD mass spectrometer with a 7683 autosampler and a model 6890 gas chromatograph equipped with a 30 m \times 0.25 mm i.d. HP-5 (cross-linked phenyl-methyl siloxane) column with 0.25 mm film thickness (Agilent, Palo Alto, CA). The initial oven temperature was held at 40 °C for 6 min. It was then increased at 2.5 °C/min to 150 °C and finally at 90 °C/ min to 250 °C. The injection port and ionizing source were kept at 250 and 280 °C, respectively. The split ratio was 10:1 with $2 \,\mu$ L of sample injected. There was a solvent delay of 2 min, after which time the mass spectrum was collected from m/z35 to 300, generating 5.27 scans/s. Compound identifications were made by comparison of the mass spectra and retention times with those of corresponding standards (Aldrich Chemical Co., St. Louis, MO; Bedoukian Research, Inc., Danbury, CT) for all compounds except 2-pentanol acetate, E-4-hepten-2-ol, ethyl 2-hydroxy-3-methylbutyrate, 1-methylbutyl isobutyrate, isoamyl 3-methyl butyrate, and hexyl isovalerate, which were identified by the NIST98 library (NIST, Gaithersburg, MD).

Quantification. For the purpose of quantifying components identified, linear regression models were determined using standard dilution techniques and cyclohexanone as internal standard. Target ions were used in the identification and quantification of each component by the mass spectrometry system. Standard compounds were used in all cases if commercially available. For the quantification of the six compounds that were not available, linear regression of similar components was used. The substitutions were as follows: 2-pentanol acetate was quantified using isoamyl acetate, E-4-hepten-2-ol by E-4-hexenol, ethyl 2-hydroxy-3-methylbutyrate by isoamyl isobutyrate, isoamyl 3-methylbutyrate by isoamyl isobutyrate, and hexyl isovalerate by hexyl butyrate.

GC-O. The analysis was carried out using a Hewlett-Packard 5890 series II plus chromatograph equipped with a 30 m \times 0.25 mm i.d. HP-5 (cross-linked phenyl-methyl siloxane) column with 0.25 mm film thickness (Palo Alto, CÅ) directly connected to a flame ionization detector (FID) and a sniffing port (Gerstel, Inc., Baltimore, MD). The injector and detector temperatures were maintained at 250 and 280 °C, respectively. The volume of extract analyzed and oven program temperatures were the same as those described above for the GC-MS. Humidified air was added in the sniffing port at 100 mL/min. Three panelists were used for the detection and verbal description of the odor active components identified in both extracts. The panelists recorded their response by pressing a spring-loaded electronic switch when they perceived the aroma components. The FID mode was used to determine the retention times of the volatile components in both extracts and compared with the retention times measured in the olfac-

Table 1.	Volatile	Components	Quantified	in Banana
Essence	and Bana	ana Fruit by	GC-MS	

		concn ^a (ppm)		
		commercial	fresh	
compound	RT	banana essence	banana fruit	
2-methyl-1-propanol	2.16	100.53 ± 10.23	nd	
3-methylbutanal	2.4	32.89 ± 5.97	nd	
butanol	2.51	304.34 ± 10.2	1.06 ± 0.34	
2-pentanone	2.79	474.64 ± 18.72	2.71 ± 0.29	
2-pentanol	2.97	319.99 ± 11.81	14.26 ± 2.63	
3-hydroxy-2-butanone	3.12	66.43 ± 5.79	20.33 ± 1.24	
<i>n</i> -propyl acetate	3.2	10.13 ± 0.9	0.07 ± 0	
methyl butyrate	3.35	1.48 ± 0.06	0.46 ± 0	
acetal	3.5	19.48 ± 0.65	nd	
3-methyl-1-butanol	3.75	877.55 ± 45.62	7.9 ± 1.56	
pentanol	4.45	3.65 ± 0.3	nd	
isobutyl acetate	4.62	266.51 ± 9.42	2.45 ± 0.36	
2-hexanone	5.09	2.59 ± 0.3	nd	
hexanal	5.39	31.19 ± 2.11	21.47 ± 5.73	
ethyl butyrate	5.51	11.71 ± 0.18	0.15 ± 0	
butyl acetate	6.08	106.54 ± 5.97	1.32 ± 0.37	
ethyl crotonate	7.52	0.23 ± 0.02	nd	
2-pentanol acetate	7.78	159.59 ± 15.58	4.79 ± 1.41	
E-2-hexenal	7.88	nd	32.2 ± 1.01	
E-2-hexen-1-ol	8.68	4.24 ± 0.99	nd	
hexanol	8.87	38.29 ± 3.68	1.17 ± 0.1	
E-4-hexen-1-ol	9.31	3.15 ± 0.53	nd	
isoamyl acetate	9.2	229.73 ± 17.33	4.85 ± 1.38	
E-4-hepten-2-ol	9.4	34.41 ± 1.91	nd	
2-heptanol	10.49	19.05 ± 1.41	nd	
isobutyl isobutyrate	11.24	0.68 ± 0.02	nd	
3-hepten-2-one	12.48	3.82 ± 0.37	nd	
isobutyl butyrate	13.7 14.28	$\begin{array}{c} 14.92 \pm 0.63 \\ 3.63 \pm 0.16 \end{array}$	1.26 ± 0.36 nd	
ethyl 2-hydroxy-3-methyl- butyrate	14.20	3.03 ± 0.10	nu	
butyl butyrate	16.17	12.6 ± 0.33	0.83 ± 0.15	
Z-3-hexenyl acetate	16.8	0.32 ± 0.02	nd	
isoamyl isobutyrate	17.18	1.27 ± 0.09	0.31 ± 0	
hexyl acetate	17.28	2.41 ± 0.08	0.57 ± 0.1	
4-hexenyl acetate	17.82	1.94 ± 0.18	nd	
limonene	17.94	nd	0.94 ± 0	
1-methylbutyl isobutyrate	18.02	2.05 ± 0.18	0.43 ± 0.1	
Z-3-octen-1-ol	19.83	9.23 ± 0.62	nd	
Z-5-octen-1-ol	20.9	6.93 ± 0.89	nd	
2,3-butanediol diacetate	21.17	1.63 ± 0.33	nd	
isoamyl butyrate	22.14	nd	7.13 ± 1.23	
isoamyl 3-methylbutyrate	22.81	1.14 ± 0.16	1.48 ± 0.17	
fenchol	23.05	10.39 ± 1.79	nd	
ethyl 3-hydroxyhexanoate	24.16	2.65 ± 0.2	nd	
borneol	26.14	9 ± 0.79	nd nd	
terpinen-4-ol	26.77	1.47 ± 0.18 1.07 \pm 0.00		
α-terpineol	27.66 27.89	1.07 ± 0.09 nd	nd 0.06 ± 0	
hexyl butyrate hexyl isovalerate	30.74	nd	$egin{array}{c} 0.06\pm0\ 0.06\pm0 \end{array}$	
eugenol	37.19	nd	0.00 ± 0 2.65 ± 0.29	
eugenoi	57.10		2.00 1 0.20	

^{*a*} Concentration \pm standard deviation. nd, not detected.

tometry runs. Data were collected using the Turbochrom data system (PE Nelson, San Jose, CA).

RESULTS AND DISCUSSION

Volatile Components in Commercial Banana Essence and Fresh Banana Fruit Paste. In the analysis of volatile components present in commercial banana essence, a total of 43 compounds were identified and quantified including 16 alcohols, 20 esters, 2 aldehydes, 4 ketones, and 1 acetal (Table 1). 3-Methyl-1-butanol followed by 2-pentanone, 2-pentanol, butanol, isobutyl acetate, isoamyl acetate, 2-pentanol acetate, 2-methyl-1-propanol, and butyl acetate were the components found in greatest concentration.

In fresh banana fruit paste, a total of 26 compounds were identified and quantified, including 16 esters, 5 alcohols, 2 aldehydes, 2 ketones, and 1 terpenic hydro-

 Table 2. Descriptors of the Volatile Components Detected by GC-0

	RT			no. of panelists detecting component	
	commercial	fresh		commercial	fresh
component	banana essence	banana paste	descriptors	banana essence	banana paste
NPI ^a	2.07	2.08	pungent, acid	3	2
2-methyl-1-propanol	2.36		acid, fruity, floral	3	
3-methylbutanal	2.62	2.62	pungent	3	2
3-hydroxy-2-butanone		3.37	butyric acid, pungent		3
<i>n</i> -propyl acetate	3.45	3.49	acid/propionic acid	2	2
acetal	3.86		fruity, green	3	
3-methyl-1-butanol	3.95	3.97	rancid, pungent	3	3
NPI	4.16		cheese	1	
isobutyl acetate	5.11	5.11	plastic, rancid, pungent	3	3
hexanal	5.95	5.95	herbal, grassy, green	3	3
ethyl butyrate	6.08	6.11	fruity, green, floral	3	3
NPĬ	6.48*		green, acid, pungent	2	
butyl acetate	6.67		sweet, candy	1	
<i>E</i> -2-hexenal		8.45	floral, herbal		3
2-pentanol acetate	8.6	8.7	herbal, sweet, floral	3	2
isoamyl acetate	9.95	9.95	over-ripe banana, sweet	3	3
NPI	10.09		acid, bad cheese	2	
NPI	10.45		floral	1	
2-heptanol	11.3	11.5	fruity, humid, pungent, acid	3	2
NPI	11.6*		spicy, mint, sweet	1	
isobutyl isobutyrate	12.2		pungent, rancid	2	
NPI		14.1	fruity		1
NPI		16.15	plastic, pungent		3
NPI	16.25*	16.5	herbal, metallic/pungent, plastic	3	3
NPI		16.9^{b}	floral, grassy		2
butyl butyrate	17.38		spicy, grassy	3	
Z-3-hexenyl acetate		18.12	fruity, floral	-	2
limonene		19.1	lemon		1
Z-3-octen-1-ol	21.6		pungent, rancid	2	_
NPI		23.18^{b}	smoky, herbal, burnt		3
isoamyl butyrate	23.65		fruity, floral, acid	2	-
NPI	Relee	23.4^{b}	smoky, woody	~	1
fenchol		24.7	woody, coffee		2
NPI		26.66	fruity, sweet		$\tilde{2}$
NPI	27.16	27.2^{b}	green, floral, herbal	2	$\tilde{3}$
NPI		28.3^{b}	pungent		2
terpinen-4-ol	28.45	2010	dusty	1	
NPI	20110	29.2^{b}	mushroom, humid, plastic	÷	2
NPI	32.95*	20.2	popcorn	2	~
NPI	02.00	34.2^{b}	smoky, woody	~	3
NPI		38.53 ^b	fruity, floral, fresh		3
eugenol		38.95	cinnamon, spicy, floral		3
		00.00	spiej, norui		0

^{*a*} NPI, not positively identified. ^{*b*} Not detected by FID.

carbon. In the analysis of the paste it is important to note that the two aldehydes correspond with the components found in greatest concentration: E-2-hexenal (compound not identified in the essence) and hexanal. E-2-Hexanal has been previously identified in banana fruit (1, 3, 17, 18). However, this component was not considered by Shiota (5) as one of the most abundant compounds in banana fruit. Shiota described the major components in banana fruit as 2-pentanone and isoamyl and isobutyl esters followed by the alcohols, which constitute the alcohol moiety of such esters.

It is interesting to note the large difference between the numbers of alcohols found in the commercial essence and in the fruit paste. There were 11 more alcohols found in the essence than in the fruit. There are several possible explanations for this. The alcohols in the essence, other than 2-methyl-1-propanol, butanol, and 3-methyl-1-butanol, are in low concentrations, with an average of <9 ppm. It could be that these compounds are simply present in too small a concentration to be detected by the GC-MS in the fresh banana fruit paste. The difference could also be due to solubilities. In general, alcohols are soluble in water. Because this was steam distillation, this could have played a role in the presence of the alcohols in the essence. Another possibility is that the alcohols are reaction products generated during the processing or storage of the essence.

Other compounds present in fresh banana fruit paste in high concentration are 3-hydroxy-2-butanone, 3-methyl-1-butanol, 2-pentanol, isoamyl acetate, isoamyl isobutyrate, and eugenol. The last compound was not identified in the commercial banana essence. The compounds identified in banana fruit have been reported by several researchers in the past, except for methyl butyrate. However, in banana essence two new compounds have been identified using retention times and mass spectra of the standards (methyl butyrate and 2,3butanediol diacetate), and three have been tentatively identified using the NIST library (2-hydroxy-3-methylethylbutyrate, 1-methylbutyl isobutyrate, and 3-hydroxyethyl hexanoate).

The concentration of compounds is higher in the commercial essence than in the fresh banana fruit paste, but the relative concentration factor is not constant for all of the compounds. The difference in the concentration factors for differing compounds can be attributed to many possible factors. The main contributor to the concentration factor would be boiling point/vapor pressure. The compounds with higher vapor pressure, and thus lower boiling points, have higher concentration

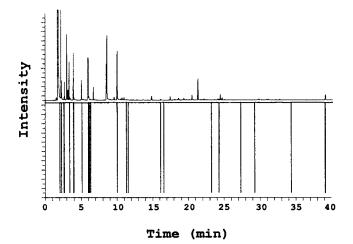


Figure 1. Plot of an FID chromatogram (top) and an aromagram (bottom) of fresh banana fruit paste using a GC-O system.

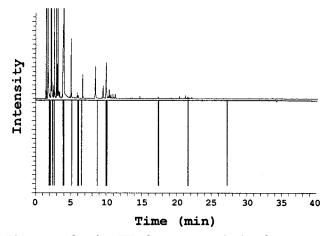


Figure 2. Plot of an FID chromatogram (top) and an aromagram (bottom) of commercial banana essence using a GC-O system.

factors in general, whereas the compounds with lower vapor pressure are less volatile during the extraction process and thus remain in the fruit. Another factor could be the degree of affinity that compounds have for the pulp versus the serum of the banana purée. Some compounds could be formed or degraded during the processing or storage, which would lead to an apparently different concentration factor. A combination of these possibilities is most likely. It would be useful if the true nature of these differences was known, but determining the causes for the variations in concentration factors was beyond the scope of this investigation. As the flavor thresholds are not known, a way to infer the importance of each compound in the aromatic profile is by GC-O.

GC-O. The descriptors obtained for all volatile components perceived in commercial banana essence and banana paste are shown in Table 2. A total of 42 components appear to contribute to the aromatic profile in banana fruit and banana essence (Figures 1 and 2). Among them, 19 have not been identified by GC-MS, and 12 of these do not present any peak in the FID chromatogram (Table 2). Most of the FID undetected volatile aroma components were perceived only in the banana paste and correspond with higher molecular weight compounds.

Figure 1 is an example of a plot of an FID chromatogram and an aromagram of fresh banana fruit paste from a panelist. There are 20 peaks in the aromagram that were detected for this replication. The number of peaks detected varied from run to run and from panelist to panelist. One can see the moderate number of peaks in the chromatogram that have no corresponding peak in the FID chromatogram. This means that there are compounds detected that have an aroma but are at concentrations below the detection limit of the FID and, therefore, not shown in the FID chromatogram. This is important because these compounds could contribute significantly to the overall aroma of the product but are not included in the current quantization.

Figure 2 is similar to Figure 1, except that it is of commercial banana essence. The most striking feature of this figure is the difference from Figure 1 in the later part of the chromatogram. After ~ 10 min, the aromagrams differ dramatically, with 9 of the 10 peaks seen in the aromagram from fresh banana fruit paste missing from the aromagram of commercial banana essence. Six of these were those with no discernible peak in the FID chromatogram. Additionally, three new peaks are observed in the aromagram from the commercial essence that were not detected in the fresh banana fruit paste. Due to the trace levels of the compounds it is difficult to determine their contribution to the overall aroma of banana.

According to these results, not all of the components found in greatest concentration in the commercial banana essence by GC-MS contribute to the aroma in banana, such as 2-pentanone, 2-pentanol, butanol, and isobutyl acetate. However, isoamyl acetate, 2-pentanol acetate, 2-methyl-1-propanol, 3-methyl-1-butanol, 3methylbutanal, acetal, isobutyl acetate, hexanal, ethyl butyrate, 2-heptanol, and butyl butyrate contribute and define the aroma in this commercial fruit essence because their odor was detected by all panelists in all replications.

With respect to the volatile components identified in fresh banana paste, *E*-2-hexenal and hexanal were the compounds quantified in highest concentration. They also seem to be important contributors to the aroma in fruit as they have been recognized by the three panelists. Volatile components detected by panelists only in fresh banana paste include *E*-2-hexenal, 3-hydroxy-2butanone, *Z*-3-hexenyl acetate, limonene, fenchol, and eugenol, which contribute floral and herbal notes. The other volatile components that have been detected by panelists but not identified in general were said to impart a pungent, floral, fruity, smoky, woody, and spicy aroma that may contribute to the different aromas of essence and fresh fruit.

Conclusion. In this study of the aromatic composition of commercial banana essence and fresh banana fruit paste a total of 49 components were identified and quantified. Among them, new compounds reported for the first time include methyl butyrate, 2-hydroxy-3-methylethyl butyrate, 1-methylbutyl isobutyrate, 2,3-butanediol diacetate, and 3-hydroxyethylhexanoate.

Twenty-three aroma active peaks plus 19 unknown peaks were characterized by GC-O to contribute to the pleasant aroma of banana. Most of these unknown volatile components were detected only in fresh banana paste and correspond with high molecular weight compounds. These compounds in general are characterized as pungent, floral, fruity, smoky, woody, and spicy aromas that may cause the difference between the aromas of banana essence and fresh fruit.

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