

Short communication

Headspace gas chromatography–mass spectrometry of volatile compounds in murici (*Byrsonima crassifolia* L. Rich)

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Abstract

Northern and Northeastern Brazil have a natural diversity of fruits, many of which are considered exotic, presenting different flavors and aromas. The enormous diversity of fruits represents a promising area for research on aromas. There is also a great potential for the manufacture of juices, desserts or other processed products. Murici is a typical fruit from these regions presenting a different flavor, reminiscent of that of cheese. This fruit is consumed mainly as juice, ice cream or as liquor, greatly appreciated by the local population. Headspace volatile compounds of three lots of the fruit from Ceará (Fortaleza) were collected by suction on Porapak Q for 2 h and desorbed with 300 μ l of acetone. The isolated volatile compounds were separated by high resolution GC. Forty-six volatile compounds were detected, of which 41 were identified by gas chromatography–mass spectrometry and Kovats indices. The most abundant compounds were ethanol (28.3%) and ethyl hexanoate (25.1%). Butanoic acid (5.1%), hexanoic acid (5.1%) and methyl butyrate (2.8%) were also detected in the headspace of the fruit and confirm its unusual cheese aroma.

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Keywords: Headspace analysis; *Byrsonima crassifolia*; Fruits; Food analysis; Volatile organic compounds; Aroma compounds

1. Introduction

Brazil has a great variability of fruits which present exotic flavor, thus having a great economic potential. Murici (*Byrsonima crassifolia* L. Rich) is a small round fruit of 1–2 cm in diameter. When ripe it exhibits a yellow coloration, softer pulp and develops a characteristic and unusual cheese aroma. The fruit is found in the Northern and Northeastern regions of the country and is mainly consumed as juice, jellies, confectionery and liquor.

Volatile compounds are responsible for the aroma and flavour of foods. The same fruit, coming from different regions or of different varieties can present a different volatile composition. Volatile compounds of canned murici from the Amazonian region were previously studied by Alves and Jennings [1]. These authors used a modified Nickerson–Likens distillation-extraction unit, and in spite of 71 volatile compounds being detected, only 23 were identified. The present work aims to characterize the headspace volatile compounds of murici samples from the Northeastern region of Brazil. The headspace volatiles can represent more closely the true aroma of the samples and in this case, fresh fruits were used.

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2. Materials and methods

2.1. Selection of the raw material and sample preparation

Three lots of fresh fruits of ripe murici of 1 kg each were acquired from three different points in the local market of Fortaleza, Ceará. They were previously frozen at -18°C and transported by air to Campinas, SP. Fruits with similar ripening were sampled in 2 consecutive years, 2000 and 2001.

The three lots were analyzed in duplicate. Fruits were immersed in distilled water for 30 min, the fruit pulp was manually separated from the seed and diluted 1:4 (w/w) with water in a warring blender. After the addition of a (30%, w/w) solution of NaCl to 300 g of the obtained juice, the final solution was put in a 1-l round bottomed flask for collection of the volatile compounds.

2.2. Isolation of the volatile compounds

The flask containing the sample solution was connected to a Porapak Q trap which, in turn was connected to a water aspirator. All tubings and connections were of glass or PTFE. The volatiles were swept to the trap by suction [2]. It was found necessary to monitor the vacuum with a gauge to ensure constant pressure throughout the trapping period. An absolute pressure of 0.64 p.s.i. was used for all samples (1 p.s.i.=6894.76 Pa). Volatile compounds were collected for 2 h and then eluted with 300 μl of acetone (chromatographic grade, EM Science, Merck). The polymer was previously conditioned under a stream of nitrogen at 170°C for 8 h.

2.3. Gas chromatography

A Varian gas chromatograph, 3800 model (Walnut Creek, CA, USA) equipped with an FID detector was used. Volatile compounds were separated in a DBWax (J&W Scientific, Folsom, CA, USA) fused-silica capillary column, 30 m \times 0.25 mm I.D. and 0.50 μm film thickness. Chromatographic conditions were: split/splitless injector used in the splitless mode; hydrogen as the carrier gas with a flow of 1.3 ml/min; injector and detector temperatures of 200 and 250°C , respectively. The column tempera-

ture was maintained at 50°C for 10 min then increased at $3^{\circ}\text{C}/\text{min}$ to 200°C and held for 10 min at this temperature. Two μl of the samples were injected.

2.4. Kovats indices

A solution of alkanes ($\text{C}_7\text{--C}_{19}$, Polyscience) was prepared and co-injected with the sample under the same conditions as described for the chromatographic analysis. Kovats indices were obtained in two columns with different polarity stationary phases, DBWax and VA1. The apolar column was a fused-silica capillary column 30 m \times 0.25 mm I.D. and 0.25 μm film thickness (Varian, USA).

The conditions used in the analysis with the apolar VA1 column were: splitless injection with a volume of 2 μl ; hydrogen as the carrier gas at a flow-rate of 1.2 ml/min. Injector and detector temperatures were 200 and 250°C , respectively. The temperature was maintained for 10 min at 40°C , then raised to 110°C at a rate of $2^{\circ}\text{C}/\text{min}$, to 200°C at $5^{\circ}\text{C}/\text{min}$ and held at this temperature for 10 min.

2.5. Gas chromatography–mass spectrometry

A gas chromatograph GC17A coupled to a mass spectrometer, Shimadzu GC–MS model QP 5000, was used. The interface temperature was 240°C , the ionization voltage was 70 eV and scanning between 35 and 350 amu. GC–MS analysis was performed in two columns of different polarities, DBWax and VA1. The conditions of analysis were the same as those of the GC analysis. Helium was used as the carrier gas, with a flow-rate of 1.3 ml/min.

2.6. Assignment of structures

Identification of the volatile compounds was carried out by comparison among the mass spectra of standard compounds found in the instrument library (US National Institute of Standards and Technology, NIST) and the mass spectra of unknown compounds. The retention indices obtained from two columns of different polarities were also compared with values found in the literature. The unknown compound was considered “tentatively identified” when only the mass spectra data were obtained. Compounds were

considered positively identified when data were confirmed by the mass spectra of standard pure compounds.

3. Results and discussion

Forty-six volatile compounds from the headspace of murici were detected by high-resolution gas chromatography, of which 41 were identified. Three volatile compounds were considered tentatively identified and 17 were identified for the first time in murici.

Table 1 presents the volatile composition of the headspace of this fruit in the 2 consecutive years. The variability demonstrated by the standard deviation was expected and inherent to the analysis of fresh fruits. Kovats indices demonstrated the same elution sequence as demonstrated by the Kovats indices found in the literature for both columns, DBWax and DB1.

The predominant compounds in area percentages were esters (52–56%) followed by alcohols (29–32%). The major compounds were ethanol (28.1%), ethyl hexanoate (25.1%), methyl hexanoate (5.2%), butanoic acid (5.1%) and hexanoic acid (5.1%).

Esters were also the most abundant compounds in the canned Amazonian murici analyzed by Alves and Jennings [1]; ethyl esters such as ethyl butyrate, hexanoate and octanoate were predominant. These authors commented that specific interactions among the volatile compounds could be responsible for the characteristic aroma of this fruit, since no single compound showed the characteristic aroma of the fruit.

The great variety of ethyl esters and the presence of carbonyl compounds such as 2-pentanone, 2-heptanone and hexanal in the present study, agreed with the previous work. However, in our study, a great number of alcohols, two sulphur compounds, aromatic compounds and fatty acids were also identified. Sulphur compounds, even in very small quantities can have a great influence on the sensorial properties of a food. The ethyl 3-(methylthio)propanoate detected in murici was previously identified in passion fruit [3] and pineapple [4,5]. Teai et al. [5] claimed that this compound plays a great role in Polynesian pineapple aroma. Ethyl lactate was

also reported in pineapple [4,5] and in feijoa [6] in low concentrations.

The other sulphur compound (3-methylthiopropanol), was found in wine [7–10] and it was described as raw potato [11], sweet soup and meat-like [12].

Among the carbonyl compounds, 3-hydroxybutan-2-one presented the greatest area percentage. This compound was found in several tropical fruits such as cupuaçu [13,14], sapota [15], cashew-apple [16,17], annona fruits [18], guava [19–22] and feijoa [6]. In guava from Japan, it was found as the major compound (81%) [19]. Its aroma was described as slightly musty in sapota [15]. In cashew apples, it was described as slightly sweet [17].

The presence of ethanol can indicate the occurrence of fermentation. Ethanol was determined in kiwi [23], in guava [24] as the principal fruit component and in mangoes [25]. According to these authors, a high concentration of ethanol in mangoes is an indication that the fruits are going bad. In murici, ethanol was detected in high amounts (28.3%) and probably was an indication that fermentation had occurred. The local population usually consumes these fruits in a very ripe stage.

Fatty acids have been detected in several tropical fruits [13,16,17,19,21,26]. Isobutanoic and isopentanoic acids contribute to the characteristic pungency of cashew apples [16]. Acetic, propanoic, butanoic and 2-methylbutanoic acids were important compounds contributing to the flavor of cupuaçu [13]. Butanoic and hexanoic acids have been described in the literature as cheese aroma [10,27].

The characteristic aroma of murici is possibly related to the fatty acids. According to Bauer [28], this fruit possesses a goatly cheesy aroma, with pineapple and cherry notes. The presence of ethyl 3-(methylthio)propanoate in murici can explain the pineapple note. This author stated that this type of fruit, presenting a very unusual aroma similar to that of cheese, would not be accepted on the USA market, although, it could be accepted in yogurts.

The percentages of ethanol, methyl butyrate and ethyl hexanoate were lower in the year 2002, while the area percentage of butanoic and hexanoic acids, and of ethyl hexanoate increased. The fruits collected in 2002 could therefore present a more intense characteristic cheese aroma.

Table 1
Volatile compounds (%) of sample murici (*Byrsonima crassifolia* L. Rich)

| Compound | RI ^b | RI ^c | Means or amplitude (n=12) | SD |
|--|-----------------|-----------------|---------------------------|------|
| Ethyl acetate ^{TI} | <900 | <700 | tr | – |
| Ethanol | 946 | <700 | 28.3 | 7.07 |
| 2-Pentanone ^a | 992 | <700 | tr–0.44 | – |
| Butanoic acid, methyl ester | 1000 | 725 | 2.81 | 2.41 |
| 2-Butanol | 1046 | <700 | tr–0.72 | – |
| Butanoic acid, ethyl ester ^a | 1057 | 806 | 17.0 | 1.13 |
| Hexanal ^a | 1100 | – | tr–0.99 | – |
| 1-Propanol, 2-methyl | 1122 | <700 | tr–1.08 | – |
| Isoamyl acetate | 1133 | 882 | tr | – |
| 2-Pentanol | 1138 | 700 | 1.22 | 0.66 |
| 1-Butanol ^a | 1180 | 718 | tr–0.81 | – |
| Hexanoic acid, methyl ester | 1184 | 911 | 5.28 | 3.43 |
| 2-Heptanone | 1191 | 895 | tr | – |
| Butanoic acid, butyl ester ^a | 1205 | 999 | 2.24 | 0.71 |
| 1-Butanol, 3-methyl ^a | 1224 | 732 | tr–0.27 | – |
| Hexanoic acid, ethyl ester ^a | 1229 | 1000 | 25.1 | 4.47 |
| Propanoic acid, 2-methyl, 3-methyl-butyl ester ^{TI} | 1234 | 1059 | tr | – |
| 1-Pentanol ^a | 1272 | – | tr–0.38 | – |
| 2-Butanone, 3-hydroxy ^a | 1292 | 736 | 1.15 | 0.02 |
| Hexanoic acid, propyl ester | 1320 | – | tr | – |
| 2-Heptanol ^a | 1334 | 906 | tr–0.26 | – |
| Ethyl lactate | 1349 | – | tr–0.138 | – |
| 1-Hexanol ^a | 1362 | 827 | tr | – |
| Octanoic acid, methyl ester ^a | 1385 | 1106 | tr | – |
| 3-Hexen-1-ol, (Z) | 1391 | – | tr | – |
| Hexanoic acid, butyl ester | 1399 | 1194 | tr–0.16 | – |
| Butanoic acid, hexyl ester | 1425 | – | tr–0.42 | – |
| Octanoic acid, ethyl ester ^a | 1449 | 1194 | tr–3.43 | – |
| Acetic acid | 1474 | <700 | tr–0.41 | – |
| Benzaldehyde ^a | 1533 | 969 | tr | – |
| Ethyl-3-(methylthio)propanoate | 1577 | – | tr | – |
| Propanoic acid, 2-methyl | 1583 | 820 | tr | – |
| Propylene glycol | 1597 | – | tr | – |
| Hexanoic acid, hexyl ester | 1614 | 1367 | tr | – |
| Butanoic acid | 1640 | 786 | 5.12 | 2.4 |
| Acetophenone | 1660 | 1077 | tr | – |
| 3-Methyl butyrate | 1690 | – | tr | – |
| 2,4-Dimethylbenzaldehyde | 1710 | – | nd–0.56 | – |
| 1-Propanol, 3-(methylthio) | 1730 | – | tr–0.29 | – |
| 4-Ethyl benzaldehyde | 1732 | 1174 | tr | – |
| NI | – | – | nd–0.47 | – |
| Hexanoic acid ^a | 1861 | 1013 | 5.1 | 1.49 |
| NI | – | – | tr–0.2 | – |
| Octanoic acid ^a | >1900 | 1180 | tr–0.125 | – |
| Phenol, 3-ethyl | >1900 | 1176 | tr–0.21 | – |
| NI | – | – | tr | – |
| Decanoic acid | >1900 | – | tr | – |

TI, compounds tentatively identified; SD, standard deviation, mean % area murici compounds years 2001 and 2002; tr, traces (mean value below 0.1%); nd, not detected; NI, compounds not identified.

^a Compounds positively identified.

^b RI, retention indices on a Carbowax column.

^c RI, retention indices on a DB1 column.

Acknowledgements

Financial support for this work was provided by PRONEX (Programa de Apoio a Núcleos de Excelência) process no. 4196091-500.

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