HS-SPME GC-MS ANALYSIS OF FRESH AND RECONSTITUTED ORANGE JUICES

A. DE WINNE and P. Dirinck

Laboratory for Flavour Research, Catholic Technical University Sint-Lieven, K.U. Leuven Association, Gebr. Desmetstraat 1, BE-9000 Gent, Belgium

Abstract

In this study, the volatile flavour components of freshly squeezed orange juices and commercial processed orange juices were investigated. The freshly squeezed oranges were obtained from three different orange brands. Sampling of the processed orange juices consisted of one refrigerated fresh juice and four reconstituted juices packed in Tetrabrik or glass. Aroma components of all juices were extracted with headspace-solid phase micro extraction followed by gas chromatography-mass spectrometry. Semi-quantitative data, obtained from GC-MS profiling were statistically treated by multivariate analysis. Principal Component Analysis clearly demonstrated differences between freshly squeezed, refrigerated fresh and reconstituted orange juices. The volatile composition of the juices was influenced by the type of processing and also by the packaging. The analytical results were related with a ranking test for six descriptors by a trained laboratory panel (n= 12). Good correlations were obtained between analytical and sensory data.

Introduction

Orange juice is one of the most appreciated drinks and it is accepted all over the world. Its fresh and uniquely delicate flavour is due to complex combinations of several odour components (1). There is still an important flavour difference between freshly squeezed orange juices and commercially processed orange drinks (2-5). The sensory quality is of great importance to the consumer and several studies have shown that the aroma composition changes during storage (6). Nisperos-Carriedo and Shaw (2) found the most important changes in volatile components in juices that were reconstituted from concentrates, packaged by aseptic means and stored at room temperature. Further more, the cultivars of “Valencia” oranges (2-4), Brazilian “Pera” (5) and Turkish “Kozan” oranges (7) have been investigated. Static and dynamic headspace analyses were often used to determine the volatiles in different orange juice samples and orange cultivars (2, 4). Recently, the HS-SPME sampling method combined with gas chromatography mass spectrometry was used for the qualitative and quantitative analyses of volatile compounds in the headspace of the orange juice (8, 9) and lemon varieties (10).

The aim of this study was to analyse the volatile components in three freshly squeezed juices (no packaging) and five packed orange juices (Tetrabrik or glass) and to relate the volatile composition to sensory differences. Differences in volatile composition among the different packaging systems were also demonstrated.
**Experimental**

*Fruit juice samples.* Sampling consisted of five different commercial orange juices and three freshly squeezed orange juices as presented in (Table 1). The freshly squeezed juices were obtained from three different orange brands and bought in local food stores.

**Table 1. Evaluated fresh and processed orange juices.**

<table>
<thead>
<tr>
<th>Code</th>
<th>Type of juice</th>
<th>Type of packaging</th>
<th>Volume</th>
</tr>
</thead>
<tbody>
<tr>
<td>BG</td>
<td>fresh</td>
<td>no</td>
<td></td>
</tr>
<tr>
<td>CA</td>
<td>fresh</td>
<td>no</td>
<td></td>
</tr>
<tr>
<td>PA</td>
<td>fresh</td>
<td>no</td>
<td></td>
</tr>
<tr>
<td>TO</td>
<td>refrigerated fresh juice</td>
<td>Tetrabrik</td>
<td>1 l</td>
</tr>
<tr>
<td>MM</td>
<td>reconstituted with pulp</td>
<td>Tetrabrik</td>
<td>0,2 l</td>
</tr>
<tr>
<td>AS</td>
<td>reconstituted</td>
<td>Tetrabrik</td>
<td>1,5 l</td>
</tr>
<tr>
<td>GA</td>
<td>reconstituted with pulp</td>
<td>glass</td>
<td>1 l</td>
</tr>
<tr>
<td>LZ</td>
<td>reconstituted</td>
<td>glass</td>
<td>0,2 l</td>
</tr>
</tbody>
</table>

*Sensory analysis.* A ranking test with a panel of 12 panellists was performed comparing all processed orange juices with one of the freshly squeezed orange juice. The fresh juice (BG) was used as reference sample. Six descriptors were evaluated: aroma intensity, fresh flavour, sweetness, pulp, taste of water and bad aftertaste. Panel members were asked to organize the samples from low to high of the respective parameter.

*Sample preparation.* Isolation of the volatiles was performed by headspace-solid phase micro extraction (HS-SPME) with a 100 µm PDMS fibre (Supelco®, USA). Aliquots of 10 ml of reconstituted or fresh orange juice were poured into 20 ml vials and sealed with PTFE lined caps. Prior to HS-SPME, samples were allowed to equilibrate at 60 °C under agitation for 15 min. The volatiles were extracted for 30 minutes at 60 °C.

*Gas Chromatography-Mass Spectrometry (GC-MS) - Principal Component Analysis (PCA).* A HP-6890N/5973 GC-MS system (Agilent Technologies®), with a PONA column (HP 50 m x 0,2 mm i.d., film thickness 0,50 µm, Agilent Technologies®) was used under the following conditions: initial oven temperature was hold at 40 °C for 5 min., then programmed from 40 to 110 °C at 10 °C/min, from 110 to 180 °C at 2 °C/min, from 180 to 250 °C at 10 °C/min and finally maintained 6 min at 250 °C. Injection was performed in the splitless mode. The mass spectra were obtained by electron impact at 70eV. Identification of the volatiles was based on comparison of the spectra with the spectra of the Wiley library. Semi-quantitative determinations of the volatile components were calculated by relating the peak intensities to the intensity of nonane as internal standard and were expressed as µg/l of juice. For interpretation of the semi-quantitative data of respectively the reconstituted and fresh orange juices and for visualization of the relationships between the different samples and their volatile composition, principal component analyses were performed using Unscrambler® 6.1 (Camo, Norway) statistical software.
Results and Discussion

The fresh orange sample BG was significantly evaluated as the sweetest sample with the freshest flavour, the highest level of pulp and with the lowest bad aftertaste. The reconstituted sample in glass GA had significantly lower fresh flavour, probably due to a bad aftertaste. Also the other sample packed in glass LZ scored low in freshness and in aroma intensity. Apart from the reference sample BG, the samples TO and AS scored high for fresh flavour and sweetness. The sample MM had the lowest amount of pulp.

Aroma profiles were obtained by HS-SPME-GC-MS. Especially aldehydes, esters and terpenes were identified. (Figure 1) illustrates a histogram of the sum of the concentrations (µg/l) of the identified monoterpene hydrocarbons quantified in all analysed fruit juices. The highest concentrations of monoterpenes were found in the juices GA and LZ, the reconstituted orange juices, packed in glass. These monoterpenes were more volatile and were probably better kept due to the glass packaging. In the TetraBrik packaging, an important part of them was lost. The polyethylene liner of the package could absorb some of the flavour components. In relation with the sensory test, the samples GA and LZ were evaluated as samples with a less fresh taste. The higher level of monoterpene hydrocarbons in these samples probably added an undesirable character to the orange juice. According to Shaw (1), the essential oil of orange peel contains high levels of limonene and a high peel oil level could contribute to a bitter flavour. The juice GA had also a high α-pinene content, which could reflect addition of amounts of peel oil by processors.

![Figure 1. Histogram of the sum of concentrations of monoterpene hydrocarbons and aldehydes (µg/l juice), in all orange juices](image)

Variation in aldehyde content among the processed and fresh products is also shown in (Figure 1). Generally, the fresh orange juices showed higher aldehyde levels. Octanal and decanal were considered as important contributors to orange flavour (2, 7). Shaw (1) and Buettner and Schieberle (6) proved that the fruity ethyl butanoate and the citrus-like decanal were the most potent odorants in Valencia Late and Navel Oranges. In relation with the sensory test, the reference freshly squeezed sample BG was evaluated as the sample with the freshest taste.

Principal component analysis (PCA) of the HS-SPME-GC-MS profiles allowed a good classification between fresh orange juice and reconstituted orange juice (Figure 2). All freshly squeezed samples had a positive PC1-score (61%) and all processed samples had a negative PC1-score. Shaw et al. (3) showed with headspace GC-
technique that aseptically packaged reconstituted juice had a different profile. The refrigerated fresh orange juice TO had a strong positive PC2-score, the reconstituted orange juices had a negative PC2-score. In the reconstituted orange samples a good classification according to the packaging composition (Tetrabrik or glass) was obtained (not presented). The two orange juices packed in glass were located at the negative PC1-score. The reconstituted orange juice AS, was situated in the middle of the figure. From (Figure 2), one may conclude that the presence of the aldehydes (octanal, neral and geranial) and the esters (ethyl octanoate and ethyl decanoate) are very important for the flavour of a fresh orange juice. The HS-SPME-GC-MS-PCA approach explained the important sensory differences between processed orange juices and freshly squeezed orange juices. The sensory difference could be related to the absence of important aroma components such as aldehydes, and ethyl esters in the reconstituted orange juices.

Figure 2. PCA bi-plot of the HS-SPME-GC-MS results of all orange juices.

References