Available online at www.sciencedirect.com
ScienceDirect
Food Chemistry 105 (2007) 1748–1754
Food Chemistry
www.elsevier.com/locate/foodchem

Analytical, Nutritional and Clinical Methods

# HS-SPME coupled to GC/MS for quality control of *Juniperus communis* L. berries used for gin aromatization

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Received 14 July 2006; received in revised form 9 March 2007; accepted 12 March 2007

**Abstract**

HS-SPME coupled to GC/MS was applied to the analysis of the volatile fraction of *Juniperus communis* L. berries, which are the principal ingredient used for gin aromatization. Seventy seven compounds were identified by comparison with reference compounds or tentatively identified by comparing their mass spectra and retention index with those reported in mass spectra libraries and literature, respectively. Seventy four were detected by SPME and sixty eight were detected by solvent distillation extraction (SDE). These were mainly mono- and sesquiterpenic compounds that represented more than the 80% of the gin's volatile composition. A high percent content was due to monoterpenoids, whose analysis could be important for the assessment of sensory quality control of juniper due to their impact on gin aroma. The main monoterpenoids detected in the headspace of the juniper berries from two periods of collection were terpinen-4-ol, *p*-cymene, β-myrcene, γ-terpinene, α-pinene and limonene. These represented more than the 70% of the sample's volatile fraction. The proposed SPME method required short times and the low cost of analysis and enabled to detect a number of compounds comparable with SDE or much higher than the number of compounds reported by other extraction techniques. The results suggested the suitability of this technique for the assessment of the volatile composition of juniper berries intended for gin flavouring.


*Keywords:* Solid phase microextraction; SPME; Mass spectrometry; *Juniperus communis* L. berries; Quality control

## 1. Introduction

Common juniper, *Juniperus communis* L. (Cupressaceae) is an aromatic and evergreen shrub, whose berries are known for their physiological properties (Barjaktarović, Sovilj, & Knez, 2005; Kallio & Jünger-Mannermaa, 1989). Juniper berries are widely used in flavours, perfumes and pharmaceuticals and to aromatise alcoholic beverages. In particular, they are used with other botanical ingredients in the production of commonly consumed juniper-based spirits, such as gin (Aylott, 2003). According to European regulations (EEC 1576/89), the main flavour in the most common and popular type of gin (London dry gin), which belongs to the "Distilled gin" class, should come from juniper berries. In fact, the "juniper" note was reported as the sensory characteristic distinguishing gins from other alcoholic beverages (McDonnell, Hulin-Bertaud, Sheehan, & Delahunty, 2001). Therefore, the main impact on the perception of dry gin flavour should be related to the presence of several aromatic volatile and semivolatile compounds derived from juniper berries. For this reason, the assessment of the volatile and semivolatile composition of this raw material is of great importance to assure the gin's final sensory quality. The composition of juniper essential oil may be influenced by several factors, such as the growth site, the plant age, the bushes form and the berries ripeness (Angioni, Barra, Russo, Coroneo, & Cabras, 2003; Kallio & Jünger-Mannermaa, 1989).

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0308-8146/$ - see front matter © 2007 Elsevier Ltd. All rights reserved.
doi:10.1016/j.foodchem.2007.03.026

Several analytical methods are available for analysing essential oil components from plant materials. Distillation methods such as steam distillation (SD), distillation-solvent extraction (SDE), microwave-assisted extraction (MAE) and supercritical fluid extraction (SFE) have traditionally been applied in this analysis. SDE appears to be the most favourable method for recovering mono- and sesquiterpenes and their oxygenated analogues. Heavier components (diterpenoids and phytosterols) have only been observed in MAE and SFE extracts (Marriott, Shellie, & Cornwell, 2001). One of the disadvantages of the distillation method is that the essential oils may undergo chemical alterations. In addition, heat-sensitive compounds can easily be destroyed. Solvent extraction may cause loss of volatiles during the vacuum evaporation of the solvent (Pourmortazevi, Baghaee, & Mirhosseini, 2004). Moreover, these techniques are time consuming. SFE avoids these problems, but it is expensive on a laboratory scale. Headspace techniques are readily applicable to qualitative analysis. They can be used for comparison and quality control purposes or for the investigation of possible adulteration. These techniques provide information on the compounds in the vapour phase, which are mainly responsible for the odour of the product (Coleman & Lawrence, 1997).

The qualitative and quantitative composition of juniper berries' essential oil has been subject to several investigations (Angioni et al., 2003; Barjaktarović et al., 2005; Chatzopoulou, de Haan, & Katsiotis, 2002; Chatzopoulou & Katsiotis, 1995; Gonny, Cavaleiro, Salgueiro, & Casanova, 2006; Kallio & Jünger-Mannermaa, 1989; Marongiu et al., 2006; Ochocka et al., 1997; Shahmir, Ahmadi, Mirza, & Korori, 2003). However, few studies have been carried out on the headspace volatiles of juniper berries. At the best of our knowledge, only the static headspace technique has been applied for the analysis of volatile constituents of *J. communis* cones. Twenty terpenic compounds were detected in this analysis (Chatzopoulou & Katsiotis, 2006). Among headspace techniques, solid phase microextraction (SPME) is a rapid, simple, inexpensive and solvent free technique for the extraction and preconcentration of volatile compounds. It is carried out by a fused silica fibre that is coated with different stationary phases and characterized by its high sensitivity to volatile organic compounds (Yang & Peppard, 1994). In recent years, this technique has been proposed for evaluating the aromatic quality control of several foods (Kataoka, Lord, & Pawliszyn, 2000; Plutowska & Wardencki, 2007). SPME's applications have been described in the analysis of the volatile compounds of several plant species (Bicchi, Drigo, & Rubiolo, 2000; Pawliszyn, 1999). However, to date no literature is available on its application to the analysis of juniper berries.

In the present study, the suitability of SPME coupled to gas chromatography/mass spectrometry (GC/MS) was evaluated as a simple and inexpensive method for undertaking the volatile composition analysis of *J. communis* berries used for dry gin aromatization.

## 2. Materials and methods

### 2.1. Reagents and plant material

Standard compounds β-myrcene, (*S*)-(−)-limonene, linalool, (−)-α-pinene, (−)-β-pinene, γ-terpinene, *p*-cymene, bornyl acetate, (−)-α-terpineol, (+)-terpinen-4-ol, (−)-β-citronellol, *t*-β-farnesene, nonanal benzaldehyde and manool (4a*R*-*trans*-5-(1,5,5,8a*S*-tetramethyl-2-methylenedecahydro-1-naphthalenyl)-3-*R*-methyl-1-penten-3-ol) were purchased from Fluka-Sigma-Aldrich (St. Louis, Missouri, USA) and Fluka. Caryophyllene oxide, β-elemol and β-eudesmol were from M.C.M. Klosterfrau (Köln, Germany). The SPME fibre used was a 2 cm long Divinylbenzene/Carboxen/Polydimethylsiloxane 50/30 μm (DVB/CAR/PDMS), from Supelco (Bellefonte, PA, USA). Before use, the fibre was conditioned as recommended by the manufacturer.

Dried ripe berries of *J. communis*, collected in November of 2002 and 2003 from Alt Urgell (Lleida, Spain), were purchased from Plantas Medicinales de Catalunya (L'Hospitalet de Llobregat, Spain).

### 2.2. HS-SPME and GC–MS analysis

Juniper berries were manually crushed in a mortar. Then, 0.2 g of crushed berries were placed in a 10 ml vial fitted with a silicone septum. This was then immersed in a silicon oil bath at 50 °C. After 5 min of sample conditioning and subsequent headspace equilibration, the fibre was exposed to the sample headspace for 30 min and immediately desorbed in the gas chromatograph injector.

GC analyses were performed on an Agilent Technologies 6890N Network gas chromatograph coupled to an Agilent Technologies 5973 Network quadrupole mass selective spectrometer and provided with a split–splitless injection port. Helium was the carrier gas, at a linear velocity of 38 cm/s. The separation of compounds was performed on Supelcowax-10 (Supelco Ltd., Bellefonte, PA, USA) and then on HP-5MS (Hewlett-Packard, Avondale, PA, USA) capillary columns (both 30 m × 0.25 mm ID, 0.25 μm film thickness). Column temperature was held at 40 °C for 5 min and increased to 250 °C at 3 °C/min, holding 10 min. The injector temperature was 260 °C. Desorption was carried out in the splitless mode during 2.5 min. Then, the fibre was maintained in the injector port during 10 min after opening the spit valve.

The temperatures of the ion source and the transfer line were 175 and 280 °C, respectively. Electron impact mass spectra were recorded at 70 eV ionization energy, 2 scan/s.

The GC–MS analysis was carried out in the complete scanning mode (SCAN) in the 40–300 u mass range.

Compounds were identified by comparing their mass spectra and retention times with those of standard compounds, or else by comparing their mass spectra with those of the mass spectra libraries Wiley 6 and NIST 2.0. Moreover, Kovat's indices (calculated with reference to a homologous series of *n*-alkanes) were determined on two

chromatographic capillary columns with distinct polarity and retention indices determined with reference to a homologous series of fatty acids methyl esters, were determined on the Supelcowax-10 capillary column. These indices were then compared with retention indices available in the literature.

Compounds were quantified as area percentages of total volatiles.

### 2.3. SDE extraction

For SDE extraction, 14 g of crushed berries and 400 ml of bidistilled water were placed in the flask of a Likens-Nickerson apparatus. A second flask with a 5 ml mixture of pentane and dichloromethane (3:1) (SDS, Peypin, France) was used as the organic phase, and the mixture was then boiled for 4 h. The mixture of pentane and dichloromethane was chosen as organic solvent with the aim of obtaining different solvent polarities without exceeding the water density. In this way, the original arrangement of the extraction system could be maintained. A cooling closed loop of ethylenglicol was used to avoid the loss of any volatile compound. After cooling, the extract fraction was collected and dried with anhydrous $Na_2SO_4$. The extract (0.3 μl) was then injected in the gas chromatograph in the splitless mode.

## 3. Results and discussion

The headspace of *J. communis* berries intended for dry gin aromatization was analysed by applying the analytical method previously developed for the analysis of gin headspace (Vichi, Riu-Aumatell, Mora-Pons, Buxaderas, & Lopez-Tamames, 2005). The SPME extraction conditions were chosen in order to favour the determination of the less volatile terpenic compounds in addition to the more volatile terpenoids. Besides the more volatile monoterpenoids, which appeared as major compounds (Table 1), the SPME extraction at 50 °C for 30 min enabled minor compounds with a poor volatility to be detected, such as oxygenated sesquiterpenes. This is due to the fact that high temperatures enhance the mass transfer of analytes from the sample to the headspace and increase their concentration in the gas phase, thus improving the sensitivity of less volatile compounds. However, as the adsorption of analytes by the fibre is an exothermic process, the increase in temperature affects negatively the adsorption of the more volatile analytes (Zhang & Pawliszyn, 1995). Extraction temperatures above 50 °C were not taken into consideration, to avoid possible alterations of the sample.

The application of the SPME method to the analysis of the juniper berries headspace led to the identification or tentative identification of seventy four compounds. These mainly consisted of mono- and sesquiterpenes and their oxygenated derivatives (Table 1). This method enabled fifty-eight of the seventy compounds detected in samples of six widely consumed commercial gin brands (Vichi et al., 2005) to be identified. This represents more than the 80% of gin total compounds. The remaining gin compounds may be derived from other botanical species used in the aromatization process. Moreover, minor compounds were detected in juniper berries which were not detected in the gin's volatile fraction. They were probably masked by other chromatographic peaks. Juniper monoterpenoids, which are supposed to influence the sensory characteristics of gins by contributing to the "juniper" sensory note (Riu-Aumatell, Vichi, & Mora-Pons, submitted for publication), ranged from the 82.5% to the 89% of total compounds determined by SPME in the juniper berries. The analysis of these compounds could be extremely important in the sensory quality control of juniper, due to their impact on the gin's aroma. The identification results and the relative content (%) of the compounds detected in juniper berries collected in two distinct years are reported in Table 1, together with the identification methods employed. In order to guess how the different compounds would contribute to the gin global aroma, the same table reports the odour notes associated with each compound, when available in the literature. With the aim to compare the results obtained by SPME with those given by a distillation method, a solvent/distillation extraction (SDE) was carried out on the same juniper berries samples (Table 1). Fig. 1 shows the chromatographic profile of a juniper berries sample extracted by SPME (a) and by SDE (b). The identification of the chromatographic peaks, according to Table 1 is also reported.

The main monoterpenoids detected by SPME in the headspace of the juniper berries were: terpinen-4-ol, *p*-cymene, β-myrcene, γ-terpinene, α-pinene, limonene and α-terpinene (Table 1). They represented around the 70% of the sample's volatile fraction. These results are in agreement with those obtained by SDE (Table 1) and those previously reported by other authors (Angioni et al., 2003; Barjaktarović et al., 2005; Kallio & Jünger-Mannermaa, 1989; Shahmir et al., 2003). However, by SPME extraction, α-pinene did not give the highest response, as observed by SDE (Table 1) and as previously described in juniper berry essential oil. The SPME extraction conditions could have led to a lower uptake of the most volatile compounds, as mentioned above. Moreover, the concentration of terpenes in juniper berries seems to be influenced by growth factors. Terpinen-4-ol and α-terpinolene levels are higher in growth sites that are far from the sea, α-pinene amounts in berries from pyramidal bushes are lower than quantities in prostrate bushes. In addition, the age of the plant seems to be related to the content of some sesquiterpenes (Kallio & Jünger-Mannermaa, 1989).

Among sesquiterpenic hydrocarbons extracted by both SPME and SDE, the highest uptakes were given by the sum of γ- and δ-cadinene, followed by α- and γ-muurolene, selinene and *t*-β-caryophyllene. The main oxygenated sesquiterpenes were the tentatively-identified torreyol, α-cadinol, spathulenol and T-muurulol (Table 1). This is in

Table 1
Characterization and percent amounts of volatile compounds in the *Juniperus communis* berries' headspace, extracted by SPME and SDE

| No. | Compound | ID[a] | $RI_{fame}$[b] | $KI_{wax}$[c] | $KI_{HP-5}$[d] | %[e] SPME | %[e] SDE | Odour note |
|---|---|---|---|---|---|---|---|---|
| *Monoterpenes* | | | | | | | | |
| 1 | Tricyclene[f] | RI[g], MS[h] | 105 | 1001 | 920 | 0.08–0.16 | – | |
| 2 | α-Pinene | S[i], RI, MS | 113 | 1017 | 929 | 8.53–13.42 | 24.9–26.5 | Pine-like, resinous[1] |
| 3 | α-Thujene | RI,MS | 115 | 1022 | 923 | 1.15–1.45 | 3.04–3.14 | Wood, green, herb[2] |
| 4 | α-Fenchene | RI,MS | 128 | 1050 | 939 | 0.03–0.04 | 0.03–0.33 | |
| 5 | Camphene | RI,MS | 131 | 1056 | 942 | 0.16–0.26 | 0.03–0.34 | Camphor[2] |
| 6 | β-Pinene | S,RI,MS | 148 | 1100 | 969 | 0.74–0.97 | 1.51–1.92 | Resinous, woody, dry[1] |
| 7 | Sabinene | RI,MS | 155 | 1117 | 968 | 1.64–3.09 | 4.23–6.43 | Pepper, turpentine, wood[2] |
| 8 | Verbenene | RI,MS | 157 | 1119 | 948 | 0.14–0.80 | 0.04–0.66 | |
| 9 | Thuja-2,4(10)-diene[f] | RI,MS | 162 | 1131 | 979 | 0.07–0.17 | 0.07–0.08 | |
| 10 | δ-3-Carene | RI,MS | 167 | 1141 | 1011 | 0.10–0.12 | 0.03–0.15 | Sweet[1], lemon, resin[2] |
| 11 | 1(7),4,8-*o*-Menthatriene[f] | MS | 171 | 1151 | 992 | 0.02–0.02 | 0.01–0.06 | |
| 12 | α-Phellandrene | RI,MS | 175 | 1159 | 999 | 0.31–0.67 | 0.17–0.18 | |
| 13 | β-Myrcene | S,RI,MS | 177 | 1172 | 987 | 10.6–11.3 | 3.56–5.41 | Wet soil, musty[1], balsamic, spice[2] |
| 14 | α-Terpinene | RI,MS | 181 | 1177 | 1011 | 1.38–4.22 | 2.02–2.21 | Lemon, citrus[1] |
| 15 | Limonene | S,RI,MS | 190 | 1200 | 1025 | 4.41–5.99 | 3.59–5.16 | Citrus-like, fresh[1], lemon, orange[2] |
| 16 | β-Phellandrene | RI,MS | 194 | 1204 | 1026 | 0.84–2.18 | 0.38–0.57 | Mint, terpentine[2] |
| 17 | 1,3,8-*p*-Menthatriene[f] | RI,MS | 200 | 1219 | 1049 | 0.06–0.29 | 0.09–0.24 | Turpentine[2] |
| 18 | γ-Terpinene | S,RI,MS | 213 | 1244 | 1055 | 3.09–10.15 | 3.36–3.49 | Lemon, lima-like[1], turpentine[2] |
| 19 | *t*-Ocimene | RI,MS | 213 | 1254 | 1046 | 0.03–0.06 | – | Ssweet, herb[2] |
| 20 | *p*-Cymene | S,RI,MS | 229 | 1275 | 1020 | 7.58–13.50 | 2.55–5.17 | Fresh, solvent, citrus[1] |
| 21 | α-Terpinolene | RI,MS | 233 | 1283 | 1083 | 3.39–1.36 | 1.02–1.14 | Citrus, pine[1] |
| 22 | *o*-Cymene[f] | RI,MS | 238 | 1291 | 1095 | 0.05–0.03 | 0.02–0.03 | |
| 23 | *p*-Cymenene[f] | RI,MS | 311 | 1430 | 1318 | 0.06–0.20 | 0.36–0.57 | |
| *Oxygenated monoterpenes* | | | | | | | | |
| 24 | (*z*)-Rose oxide | RI,MS | 250 | 1369 | 1107 | 0.11–0.18 | – | Sweet, rose, green, flower[2] |
| 25 | (*t*)-Rose oxide[f] | RI,MS | 259 | 1417 | 1115 | 0.04–0.09 | 0.01–0.02 | Flower[2] |
| 26 | *cis*-Linalool oxide[f] | RI,MS | 306 | 1460 | 1064 | 0.16–0.29 | 0.06–0.08 | Flower[2] |
| 27 | *trans*-Linalooloxide[f] | MS | 321 | 1475 | – | 0.06–0.20 | – | Flower[2] |
| 28 | Camphor | RI,MS | 354 | 1495 | 1137 | 0.12–0.12 | 0.26–0.55 | Camphor[2] |
| 29 | Verbenol | RI,MS | 359 | 1504 | 1144 | – | 0.16–0.05 | |
| 30 | Linalool | S,RI,MS | 377 | 1552 | 1097 | 0.25–0.35 | – | Floral, citrus, green[1] |
| 31 | *cis*-Sabinene hydrate | RI,MS | 381 | 1556 | 1060 | 0.13–0.23 | 0.40–0.46 | Balsamic[2] |
| 32 | Bornyl acetate | S,RI,MS | 386 | 1565 | 1282 | 0.21–0.47 | 0.32–0.53 | Sweet, herbaceous, piney[1] |
| 33 | Terpinen-4-ol | S,RI,MS | 400 | 1593 | 1175 | 22.5–30.6 | 9.1–11.7 | turpentine, nutmeg, must[2] |
| 34 | Myrtenal | RI,MS | 409 | 1602 | 1190 | 0.20–0.30 | 0.22–0.71 | Spice[2] |
| 35 | Pinocarveol[f] | RI,MS | 425 | 1690 | 1139 | 0.42–0.78 | 0.68–1.43 | Flower[2] |
| 36 | *p*-mentha-1,5-dien- 8-ol | RI,MS | 431 | 1710 | 1170 | 3.07–4.04 | 0.39–1.11 | |
| 37 | *t*-Carenol[f] | MS | 438 | 1727 | – | 0.10–0.37 | 0.66–0.95 | |
| 38 | Verbenone[f] | RI,MS | 440 | 1729 | – | 1.41–1.70 | 0.83–3.17 | |
| 39 | Terpenyl acetate | RI,MS | 444 | 1731 | 1342 | 0.23–0.84 | 0.24–0.33 | Herbaceous, sweet, mild[1], wax[2] |
| 40 | α-Terpineol | S,RI,MS | 447 | 1736 | 1186 | 2.26–3.99 | 5.20–5.54 | Floral, lilac-like[1], oil, anise, mint[2] |
| 41 | Carvone[f] | RI,MS | 462 | 1759 | 1279 | 0.09–0.17 | 0.93–2.73 | Mint, basil, fennel[2] |
| 42 | Cuminal | RI,MS | 478 | 1776 | 1234 | 0.74–1.50 | 0.42–0.49 | Acid, sharp[2] |
| 43 | β-Citronellol | S,RI,MS | 481 | 1780 | 1227 | 0.28–0.48 | 0.21–0.34 | Rose[2] |
| 44 | Myrtenol | RI,MS | 492 | 1788 | 1192 | 0.32–0.43 | 0.35–0.64 | |
| 45 | *t*-Carveol | RI,MS | 515 | 1825 | 1240 | 0.23–0.50 | 0.22–0.58 | Fresh, spearmint, caraway[2] |
| 46 | *p*-Cymen-8-ol[f] | RI,MS | 521 | 1871 | 1229 | 1.90–2.82 | 0.79–1.01 | Citrus, must[2] |
| 47 | *cis*-Carveol[f] | RI,MS | 526 | 1879 | 1216 | 0.09–0.10 | 0.07–0.09 | Caraway[2] |
| 48 | Perillyl alcohol[f] | RI,MS | 586 | 1970 | 1290 | 0.04–0.05 | – | Green, pungent, fatty[1] |
| *Sesquiterpenes* | | | | | | | | |
| 49 | α-Cubebene | RI,MS | 321 | 1446 | 1340 | 0.33–0.68 | 1.20–1.26 | Herb, wax[2] |
| 50 | α-Copaene | RI,MS | 330 | 1470 | 1362 | 0.63–0.88 | 0.77–1.51 | Wood, spice[2] |
| 51 | β-Cubebene | RI,MS | 362 | 1518 | 1377 | 0.11–0.11 | 0.12–0.19 | Citrus, fruit[2] |
| 52 | *t*-β-Caryophyllene | RI,MS | 389 | 1571 | 1403 | 0.59–1.27 | 5.09–5.63 | Wood, spice[2] |
| 53 | γ-Elemene | RI,MS | 413 | 1618 | 1421 | 0.47–1.00 | 1.63–2.30 | Green, wood, oil[2] |
| 54 | α-Humulene | RI,MS | 422 | 1709 | 1435 | 0.62–0.95 | 2.69–3.03 | Wood[2] |
| 55 | *t*-β-Farnesene | S,RI,MS | 431 | 1719 | 1446 | 0.34–0.86 | 1.00–1.46 | Wood, citrus, sweet[2] |
| 56 | γ-Muurolene | RI,MS | 446 | 1723 | 1454 | 0.86–1.59 | 0.97–1.04 | |
| 57 | Germacrene D | RI,MS | 449 | 1733 | 1462 | 0.56–1.17 | 1.07–1.30 | Wood, spice[2] |
| 58 | α-Selinene | RI,MS | 453 | 1740 | 1470 | 0.86–1.63 | 2.04–5.56 | Wood[2] |
| 59 | α-Muurolene | RI,MS | 455 | 1748 | 1478 | 0.86–1.24 | 0.73–0.78 | Wood[2] |

(*continued on next page*)

Table 1 (*continued*)

| No. | Compound | ID[a] | $RI_{fame}$[b] | $KI_{wax}$[c] | $KI_{HP-5}$[d] | %[e] SPME | %[e] SDE | Odour note |
|---|---|---|---|---|---|---|---|---|
| 60 | δ-Cadinene | RI,MS | 471 | 1767 | 1504 | 3.97–5.90 | 5.17–5.54 | Thyme, medicine, Wood[2] |
| 61 | γ-Cadinene | RI,MS | 474 | 1768 | 1504 | | | Wood[2] |
| 62 | Cadina-1,4-diene | RI,MS | 482 | 1778 | 1515 | 0.30–0.49 | 0.95–1.32 | Spice, fruit[2] |
| 63 | Calamenene | RI,MS | 503 | 1799 | 1534 | – | 0.10–0.24 | |
| 64 | Germacrene B | RI,MS | 503 | 1800 | 1535 | 0.20–0.25 | 2.94–4.71 | |
| 65 | α-Calacorene | RI,MS | 535 | 1893 | 1519 | 0.22–0.31 | 0.27–0.36 | Wood[2] |
| *Oxygenated sesquiterpenes* | | | | | | | | |
| 66 | Caryophyllene oxide | S,RI,MS | 579 | 1953 | 1558 | 0.07–0.11 | 1.40–1.66 | Herb, sweet, spice[2] |
| 67 | Torreyol | RI,MS | 618 | 2041 | 1604 | 0.30–0.41 | 0.82–1.04 | |
| 68 | Elemol | S,RI,MS | 630 | 2066 | – | 0.02–0.03 | 0.00–0.66 | Green, wood[2] |
| 69 | Spathulenol | RI,MS | 650 | 2104 | 1571 | 0.17–0.27 | 2.18–2.29 | Herb, fruit[2] |
| 70 | *t*-Cadinol | RI,MS | 673 | 2153 | 1636 | 0.09–0.15 | 0.75–0.79 | |
| 71 | *t*-muurulol | RI,MS | 680 | 2168 | 1649 | 0.11–0.22 | 2.46–2.50 | Herb, weak spice[2] |
| 72 | β-Eudesmol | S,RI,MS | 695 | 2203 | 1655 | 0.03–0.06 | 0.58–0.68 | Sweet, wood[2] |
| 73 | α-Cadinol | RI,MS | 700 | 2210 | 1651 | 0.24–0.40 | 1.83–1.95 | Herb, wood[2] |
| | *Other compounds* | | | | | | | |
| 74 | 6-Methyl-5-hepten-2-one[f] | S,RI,MS | 247 | 1312 | 989 | 0.02–0.06 | – | |
| 75 | Benzaldehyde | RI,MS | 362 | 1516 | 959 | 0.05–0.09 | – | Almond, burnt sugar[2] |
| 76 | Cinnamaldehyde[f] | MS | 599 | 1978 | 1582 | 0.08–0.15 | – | Cinnamon, paint[2] |
| 77 | Manool | S,RI,MS | 851 | 2489 | 2057 | – | 0.18–0.26 | |

[a] Identification method.
[b] Retention indices based on fatty acid methyl esters (Supelcowax-10).
[c] Kovats indices on Supelcowax-10.
[d] Kovat's indices on HP-5.
[e] Percent amount of volatile compounds in juniper berries samples, calculated o on the basis of chromatographic peak areas.
[f] Detected in juniper berries' headspace but not in the gin samples' headspace (17).
[g] Tentatively identified by KI or $RI_{fame}$.
[h] Tentatively identified by mass spectra.
[i] Identified by comparison with standard compounds.
[1] http://www.crec.ifas.ufl.edu.
[2] http://www.flavornet.org/flavornet.html.

agreement with the results obtained by SDE and in accordance with previous results obtained by other authors (Barjaktarović et al., 2005; Kallio & Jünger-Mannermaa, 1989; Shahmir et al., 2003).

The number of compounds detected by the present SPME (seventy four) method is comparable with the number of compounds extracted by SDE technique (sixty eighth), as shown in Table 1, and it is much higher than the number of compounds detectable by static headspace analysis (Chatzopoulou & Katsiotis, 2006). The most relevant difference between volatile profiles obtained by SPME and SDE was the uptake of the less volatile compounds. SDE allowed detecting a higher number of compounds with poor volatility, such as the diterpenoid manool (Table 1). In addition, the percent areas of both oxygenated and not oxygenated sesquiterpenes observed by SDE was higher than those given by SPME (Table 1). Nevertheless, given that poorly volatile compounds such as sesquiterpenoids and diterpenoids are scarcely involved in the sensory perception, their relevance from the point of view of the quality control of juniper berries intended for gin aromatization should be low.

Moreover, the SPME extraction temperature was significantly lower than the temperature needed for the analysis of juniper volatiles by distillation and by static headspace analysis (Angioni et al., 2003; Chatzopoulou & Katsiotis, 2006; Gonny et al., 2006; Shahmir et al., 2003). Thus, the SPME method avoided the risk of possible chemical alterations of heat-sensitive compounds.

In conclusion, the application of SPME to the analysis of juniper berries intended for gin aromatization enabled an extensive number of volatile and semivolatile compounds to be identified. These represented most of the compounds documented in gin samples (Vichi et al., 2005). A high content percentage was due to monoterpenoids, whose analysis could be important in the evaluation of sensory characteristics of juniper. In fact, monoterpenoids can have a heavy impact on gin aroma because of their high volatility. Moreover, the volatile composition of juniper determined by SPME was in reasonable agreement with results previously obtained using other extraction techniques (Angioni et al., 2003; Barjaktarović et al., 2005; Kallio & Jünger-Mannermaa, 1989; Shahmir et al., 2003). This non-invasive technique operated at low extraction temperatures, which implies a lower risk of thermal alteration of the sample. Furthermore, the SPME analysis can be performed in a short time and at a low cost. Both of these factors are required as they enable a large number of samples to be analysed. Therefore, these results suggest the suitability of the proposed method to assess the volatile composition of juniper berries used as principal flavouring ingredient in gin production.

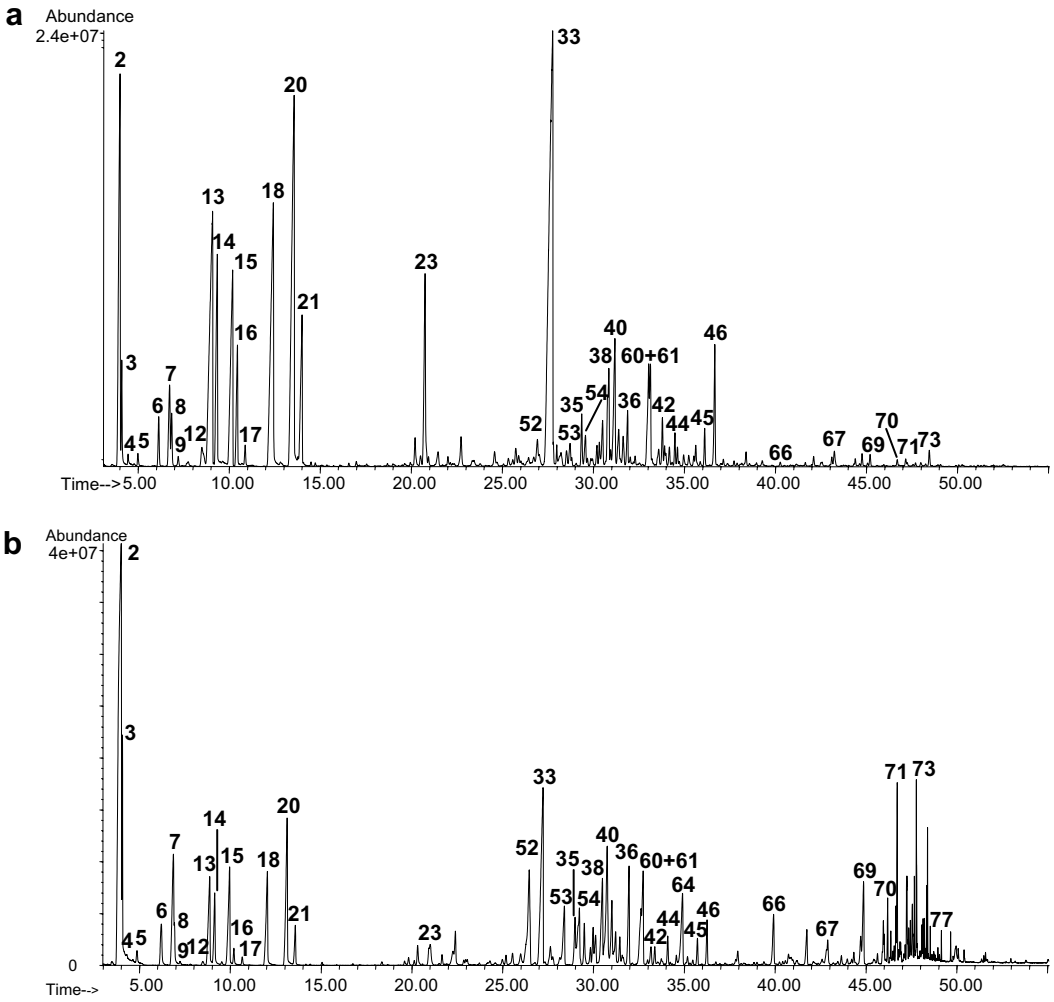


Fig. 1. GC–MS chromatographic profiles of *Juniperus communis* L. berries obtained by SPME (a) and SDE (b) extraction. The separation was carried out on a Supelcowax-10 capillary column. The identification numbers correspond to those reported in Table 1.

## Acknowledgements

This study was supported by the Generalitat de Catalunya, project 2005SGR00156; by the Ministerio de Ciencia y Tecnología (MCYT), project AGL2005-03451/ALI; and by the Secretaría de Estado de Educación y Universidades (Ministerio de Educación y Ciencia), Ref. SB2003-0118.

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