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Comparison of Static Headspace, Headspace Solid Phase Microextraction, Headspace Sorptive Extraction, and Direct Thermal Desorption Techniques on Chemical Composition of French Olive Oils

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Static headspace (SHS), headspace solid phase microextraction (HS-SPME), headspace sorptive extraction (HSSE), and direct thermal desorption (DTD) were applied to the analysis of four French virgin olive oils from Corsica. More than 60 compounds were isolated and characterized by GC-RI and GC-MS. SHS was not suited to the characterization of olive oil volatile compounds because of low sensitivity. The SPME and HSSE techniques were successfully applied to olive oil headspace analysis. Both methods allow the characterization of volatile compounds (mainly C_6 aldehydes and alcohols), which contribute significantly to the "green" flavor note of virgin olive oils. The PDMS stir bar showed a higher concentration capacity than a DVB/CAR/PDMS SPME fiber due to the higher volume of polymeric coating. DTD was a very good tool for extracting volatile and especially semivolatile compounds, such as sesquiterpenes, but requires a significant investment like that for HSSE. Finally, SPME may be a more appropriate technique for routine quality control due to its operational simplicity, repeatability, and low cost.

KEYWORDS: Static headspace (SHS); headspace solid phase microextraction (HS-SPME); headspace sorptive extraction (HSSE); direct thermal desorption (DTD); virgin olive oil

INTRODUCTION

Today, monitoring vegetable oil authenticity is of prime importance in the food industry. Many analytical procedures have been used to identify and quantify the different components that characterize vegetable oils: fatty acids, triglycerides, waxes, sterols, tocopherols, hydrocarbons, alcohols, or volatiles (*1*). C_6 aldehydes and alcohols and their corresponding esters are the most abundant volatile compounds present in olive oils and are produced enzymatically from polyunsaturated fatty acids through the "lipoxygenase pathway" (*2*, *3*).

These volatile compounds are mainly responsible for olive oil flavor. To evaluate olive oil aroma, several techniques are used (*3*, *4*). Among these extraction techniques, static headspace (SHS) is a simple and fast technique to implement because no sample preparation or solvent is needed. SHS is used in food flavor analysis for the extraction of volatile compounds of carrot samples (*5*) or for the characterization of Camembert cheeses by injecting the headspace directly into a mass spectrometer without chromatographic separation (*6*). This technique has rarely been used for the analysis of olive oil volatile compounds because of the low content of extracted compounds, often below the GC detectability threshold. However, the direct coupling of a mass spectrometer with SHS (SHS-MS) allowed the characterization of olive oils (*7*, *8*). Indeed, the authors used a statistical analysis to differentiate sources of monovarietal olive oil with 30 variables (acidity, oxidative stability, total polyphenols, bitterness intensity, etc.) in addition to the SHS-MS data (*7*). The same authors also applied this method to the detection of adulterants in olive oils, despite a low sensitivity, after a chemometric treatment, but with only the mass charge ratio obtained from the mass spectrometer (*8*).

Solid phase microextraction (SPME) is a solvent-free sample preparation technique for the extraction of volatile and semivolatile compounds, composed of a fused-silica fiber coated with different stationary phases. This method, developed by Arthur and Pawliszyn in 1990 (*9*, *10*), is used in many applications (*11*, *12*), among which is food flavor analysis (*13*, *14*). Several studies have been published on the characterization of olive oil volatile compounds by SPME, and many components have been identified (*15*−*18*).

Headspace sorptive extraction (HSSE), like the SPME method, is a recently introduced solventless enrichment technique. It is a new sampling method developed to extract volatile compounds from gaseous (headspace) or aqueous samples (stir bar sorptive extraction or SBSE) (*19*, *20*). HSSE is based on

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the sorption of analytes onto a thick film of poly(dimethylsiloxane) (PDMS) coated on a stir bar. Few studies have been published on the HSSE technique and none on olive oils. For example, the extraction of volatile components from aromatic plants was carried out by using a stir bar (*21*). The HSSE and SBSE techniques were also applied to the analysis of coffee (*22*), and SBSE was used for the characterization of chiral flavor compounds in strawberries (*23*).

Thermal desorption (TD) is already used for SBSE and HSSE techniques after the enrichment step. To our knowledge, only three studies have reported direct thermal desorption (DTD) on olive oil samples (*24*−*26*). Direct thermal extraction has been applied to several cooking olive oil samples to analyze the volatile and semivolatile compounds contributing to flavor and off-flavor. The technique can also be used for quality control, to determine origin, or for the detection of adulteration (*24*).

Comparison of various extraction techniques in food flavor analysis has already been described in the literature. SPME and SHS techniques have been compared for the analysis of alcohols and esters in beer (*27*). Both methods are highly correlated and present a high repeatability and a good linearity, but SPME can be considered as an inexpensive alternative to SHS (*27*). HSSE, SBSE, SPME, and SHS have also been applied to the analysis of the headspace of Arabica coffee and coffee brew (*22*). The PDMS stir bar showed better concentration capability than all SPME fibers or SHS, and this is entirely due to the high amount of PDMS coating.

Finally, headspace techniques have been successfully used for the quality control of olive oils, particularly for the detection of adulterants (*8*) or rancidity (oxidation) (*28*) or to determine their origin (*7*).

The aim of this work was to compare different extraction methods for the characterization of volatile and semivolatile compounds from virgin olive oils. Four French olive oils from Corsica were used in this study. First, the headspace composition was studied using three extraction techniques: static headspace, solid phase microextraction, and headspace sorptive extraction. To our knowledge, HSSE analysis of olive oils is reported for the first time. The data obtained by these three headspace techniques were then compared with those of DTD. The advantages and limitations of each method are examined, and the choice of the best extraction technique is discussed.

MATERIALS AND METHODS

SHS Analysis. The headspace of olive oils was analyzed by a Hewlett-Packard 7694 static headspace autosampler (Agilent Technologies, Palo Alto, CA). The headspace sampler was associated with a GC (HP 6890N GC system, Agilent) equipped with a flame ionization detector (FID) and coupled to a mass spectrometer [HP 5973N mass selective detector (MSD), Agilent].

The extraction conditions of the headspace sampler were as follows: oven temperature, 110 °C; loop temperature, 125 °C; transfer line temperature, 135 °C; sample equilibration time, 120 min. The extraction−injection step consisted first of pressurizing the flask [10 mL of each oil sample was placed in 20 mL vials (Agilent) sealed hermetically with a poly(tetrafluoroethylene) (PTFE)/white silicone septum and a cap] at 110 kPa for 0.3 min with helium (He). The injection loop (volume = 3 mL) was filled by depressurizing the headspace for 0.1 min, and the loop was swept with the carrier gas (loop equilibration, 0.05 min; sample injection, 0.5 min) to inject the volatile components into the chromatograph column HP-1 (50 m × 0.2 i.d., film thickness = 0.33 μm) by a transfer line (splitless mode). The carrier gas was helium, and the pressure applied at the column head was 282 kPa. This pressure was necessary because a positive pressure of 20 kPa must be applied to a cross that divides the flow of the analytes through two restrictor valves, one toward the FID and the other toward the MSD.

The oven was programmed from 60 to 250 °C at 2 °C/min and then isothermal (30 min). The FID temperature was set at 250 °C, and the temperatures of the ion source and the transfer line were 230 and 280 °C, respectively. Energy ionization was 70 eV; electron ionization mass spectra were acquired over the mass range of 35−200 atomic mass units (amu).

SPME-GC/FID and SPME-GC/MS Analysis. A manual SPME device and fibers were obtained from Supelco Co. (Bellefonte, PA). The fibers tested for extraction of the volatile components were as follows: poly(dimethylsiloxane) (PDMS) 100 μm, carboxen/PDMS (CAR/PDMS) 85 μm, carbowax/divinylbenzene (CW/DVB) 70 μm, and DVB/CAR/PDMS 50/30 μm.

Before use, fibers were conditioned as recommended by the manufacturer. The analyses were carried out with the DVB/CAR/PDMS [volume of the coating material = 0.5 μL (*22*)]. The olive oil sample (20 g) was placed in a 40 mL amber vial closed by a PTFE/silicone septum (Supelco). Before extraction, stabilization of the headspace in the vial was accomplished by equilibration for 60 min at 25 °C. Extraction was carried out at 25 °C (room temperature). A sampling time of 90 min was chosen to perform the analysis. Each analysis was carried out three times.

After exposure, the fiber was thermally desorbed into a GC and left in the injection port (equipped with a 0.75 mm i.d. inlet liner) for 4 min. The injector was set at 250 °C and operated in the splitless mode for 4 min unless otherwise stated. GC analyses were carried out using two Hewlett-Packard 5890 series II gas chromatographs, one equipped with an FID and one coupled to a Hewlett-Packard 5971A mass selective detector (quadrupole). Both were equipped with fused-silica capillary columns HP-1 (PDMS, 50 m × 0.2 mm i.d., film thickness = 0.33 μm for GC-FID and 0.5 μm for GC-MS). The carrier gas was nitrogen for GC-FID and helium for GC-MS (head pressure of both columns = 25 psi); oven temperature was programmed from 60 °C (5 min) to 250 °C at 2 °C/min and then held isothermal (20 min). The FID temperature was set at 250 °C, and the temperatures of the ion source and the transfer line were 170 and 280 °C, respectively. Energy ionization wa 70 eV; electron ionization mass spectra were acquired over the mass range of 35−350 amu. Before sampling, the fiber was reconditioned for 5 min in the GC injection port at 250 °C. The analyses were carried out in triplicate.

HSSE Analysis. The stir bar was supplied from Gerstel GmbH. A 1 cm long stir bar with a 0.5 mm PDMS coating was used [volume of the coating material = 55 μL (*22*)].

The PDMS stir bar was conditioned as described elsewhere (*19*). Sampling was realized by suspending the stir bar with a stainless steel stem (2.5 cm) in the headspace of the olive oil. The stir bar was positioned at 2 cm in the headspace volume. A sampling time of 120 min was chosen to perform the analysis. Other HSSE extraction conditions were the same as described under SPME.

After sampling, the stir bar was thermally desorbed in a Gerstel TDS-2 (thermal desorption system), and a CIS-4 PTV injector (cooled injection system, programmed temperature vaporization; filled with Tenax TA, Gerstel GmbH) was used for cryofocusing the analytes prior to transfer onto the analytical column. Liquid nitrogen was used to cool the CIS-4 to −50 °C during thermal desorption of the trap. For trapping the more volatile analytes, the CIS-4 liner was filled with Tenax TA (Gerstel GmbH). The stir bar was transferred into an empty glass thermal desorption tube (177 mm, 4 mm i.d., and 6 mm o.d., Gerstel GmbH) and desorbed in TDS.

Splitless thermal desorption was realized by ramping the TDS from 20 to 250 °C at 60 °C/min and holding the upper temperature for 7 min (transfer line to 300 °C). The CIS-4 was cooled at −50 °C and then set at 280 °C at 12 °C/s and maintained to 280 °C for 7 min (the trap was heated under a stream of helium carrier gas, 50 mL/min). GC and GC-MS conditions were the same as described for SHS experiments except for the GC oven, which was programmed from 60 °C (5 min) to 110 °C at a rate of 2 °C/min and a second rate at 10 °C/min to 250 °C (30 min), and the injection was in split mode (solvent venting, purge flow to split vent, 10 mL/min); for GC-MS electron ionization mass spectra were acquired over the mass range of 35−350 amu. Each

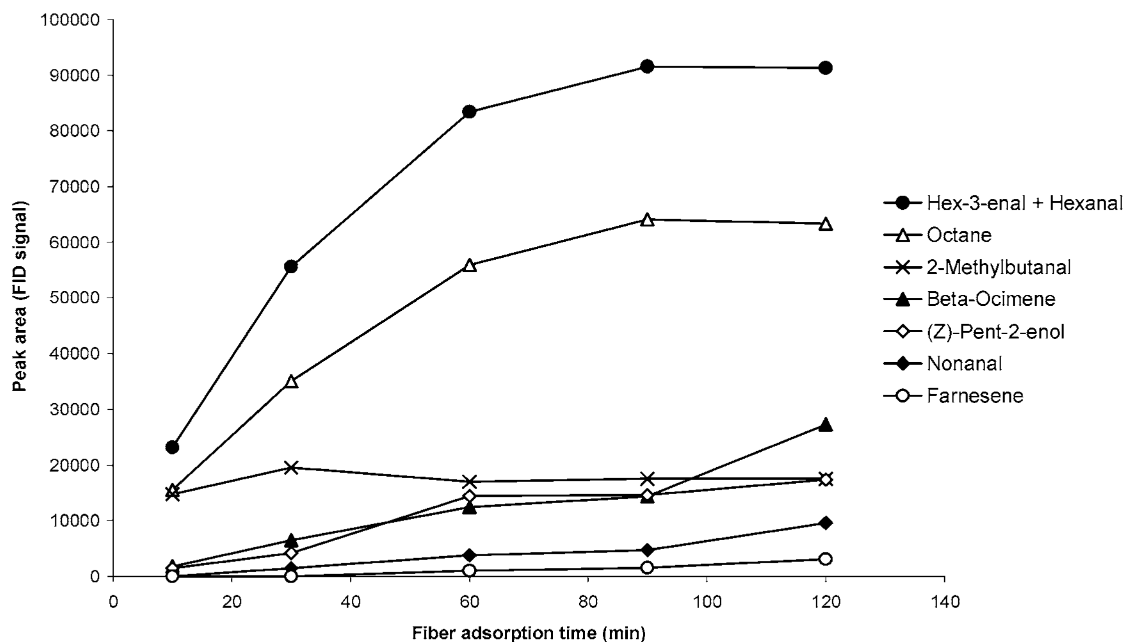

Figure 1. Adsorption time profiles of some olive oil volatile compounds (Sabine variety) using a DVB/CAR/PDMS fiber.

extraction was repeated three times and, before sampling, the stir bar was reconditioned for 20 min in the TDS at 250 °C.

Thermal Desorption Analysis. The olive oil (5 μL) was placed with a microliter syringe (10 μL, Gerstel GmbH) on glass wool inserted into an empty glass thermal desorption tube (previously thermally desorbed with glass wool at 300 °C for 30 min). Splitless thermal desorption was realized by ramping the TDS from 20 to 80 °C at 60 °C/min and holding the upper temperature for 20 min. The other conditions were the same as for HSSE experiments. Between each analysis, the thermal desorption tube was changed and cleaned. As previously, each experiment was carried out in triplicate, and blank runs were carried out periodically during the study.

Component Identification. Identification of the components in each olive oil was based on (a) their GC retention indices (RI) on an apolar column, determined relative to the retention times of a series of *n*-alkanes (C-5 to C-28; retention times determined for each experiment, SPME and HSSE 20 s at 50 °C and DTD 0.1 μL; the other sampling conditions were the same as described above), using linear interpolation with those of authentic compounds or literature data (*29*), or (b) computer matching with the reference mass spectra of the Wiley 6 library and comparison of spectra with those of our laboratory library (mass spectra library built up from pure substances). Several structures were also confirmed by standard compound injection. All chemicals were purchased from Fluka or Sigma-Aldrich (Saint Quentin Fallavier, France). To make analytical data comparable, the peak area of each identified compound in olive oil samples was percent normalized, and mean percentage values were calculated by triplicate analyses for SPME, HSSE, and DTD experiments.

Olive Oil Samples. Four French extra virgin olive oil samples, extracted from olives of Sabine, Cailletier, Picholine, and Koroneiki varieties cultivated in Speloncato, certified as authentic by David Bichon (Speloncato, Corsica, France), were used for the investigation in this study. The fruits were harvested in January 2003. The samples were stored at 4 °C in the dark between each analysis.

RESULTS AND DISCUSSION

Four virgin French olive oils were analyzed by different extraction techniques: static headspace, solid phase microextraction, headspace sorptive extraction, and direct thermal extraction. Each experiment was carried out three times. More than 60 volatile and semivolatile compounds were isolated and characterized by GC-RI and GC-MS analysis.

SHS Analysis. *Optimization of Exposure Temperature and Time*. Many analyses were carried out by SHS on olive oil samples. Different temperatures were tested (40−110 °C) for several exposure times (30−120 min). The injection conditions were also studied with different times for sample injection, loop fill, or loop equilibration. However, the results of analysis of the four olive oils by this extraction technique show a very low sensitivity. Indeed, almost no compound was extracted by this technique or under the GC detectability threshold.

In addition, thermo-oxidation of the virgin olive oil can occur during the headspace generation time of 120 min at 110 °C. This process is responsible for the evolution of volatile compounds by oxidation of unsaturated fatty acids, and in particular for the appearance of off-flavors (*28*).

SPME Analysis. *Fiber Choice and Optimization of Adsorption Time*. Headspace solid phase microextraction was used to characterize the volatile compounds present in the four virgin olive oils produced in January 2003. The comparison of the performance of the four fibers (PDMS, CAR/PDMS, CW/DVB, and DVB/CAR/PDMS) was made using a sample of olive oil (Sabine variety). The fibers tested clearly showed that the signal obtained with the DVB/CAR/PDMS fiber was the most suitable for the analysis of olive oil volatiles, compared with PDMS, CAR/PDMS, and CW/DVB fibers. Indeed, the best results were obtained with the DVB/CAR/PDMS fiber. To determine the optimal adsorption time of the fiber to the sample headspace, the DVB/CAR/PDMS fiber was exposed for time periods of 10, 30, 60, 90, and 120 min. A sampling time of 90 min was chosen to perform the analysis (see **Figure 1**).

Headspace Composition by SPME. Compounds isolated and identified by SPME-GC/FID and SPME-GC/MS were mainly aldehydes with 52.9−63.1% of the total FID area such as but-2-enal, 2-methylbutanal, hexanal, or (*E*)-hex-2-enal and alcohols (10.8−22.2%) such as penten-3-ol, (*Z*)-pent-2-enol, (*Z*)-hex-3-enol, or (*E*)-hex-2-enol as well as monoterpene (β-ocimene) and sesquiterpene (farnesene) (**Table 1**). We proposed the structure of seven isomeric unsaturated hydrocarbons (3,4-diethylhexa-1,5-diene, 3-ethylocta-1,5-diene, and deca-3,7-diene, known as pentene dimers) that were characterized in the volatile fraction of the four French olive oils by comparison of mass spectra

Table 1. Compounds Extracted by HS-SPME in the Four Olive Oils

		Sabine[c]		Cailletier		Picholine		Koroneiki		
compound[a]	RI[b]	%[d]	SD[e]	%	SD	%	SD	%	SD	refs
acetic acid*	557	1.5	0.1	0.9	0.2	0.5	0.0	0.8	0.2	*16, 31*
ethyl acetate*	596	1.3	0.1	1.1	0.3	0.2	0.0	0.5	0.1	*16, 31*
but-2-enal[†,°]	618	1.7	0.2	0.8	0.2	0.4	0.0	1.1	0.2	*16*
3-methylbutanal*	628	1.0	0.1	0.4	0.2	0.4	0.0	0.6	0.1	*3, 16*
2-methylbutanal*	638	1.2	0.1	0.4	0.2	0.4	0.0	0.5	0.2	*3, 16*
penten-3-one°	657	0.7	0.1	0.5	0.1	1.8	0.0	1.1	0.2	*16, 31*
penten-3-ol°	660	1.8	0.1	1.1	0.1	nd[f]		nd		*16, 31*
pentan-3-one*	670	3.8	0.2	2.4	0.2	1.4	0.2	2.5	0.3	*16, 31*
heptane*	700	0.5	0.2	0.4	0.2	nd		0.1	0.2	*16*
3-methylbutanol*	719	1.2	0.1	0.4	0.1	0.2	0.0	0.3	0.0	*16, 31*
(*E*)-pent-2-enal°	724	1.1	0.1	0.7	0.2	0.5	0.0	0.5	0.1	*16, 31*
(*Z*)-pent-2-enol°	751	0.8	0.1	0.8	0.1	0.3	0.0	0.7	0.1	*16, 31*
hex-3-enal[†,°]	773	5.2	0.3	4.7	0.0	3.1	0.2	2.6	0.1	*16, 31*
hexanal*										*16, 31*
octane*	800	3.7	0.2	3.2	0.0	0.8	0.0	2.2	0.1	*16, 24*
(*E*)-hex-2-enal*	826	42.7	1.2	49.0	2.1	58.1	1.9	47.2	1.9	*16, 31*
(*Z*)-hex-3-enol*	837	4.5	0.3	5.1	0.2	5.5	0.4	7.8	0.2	*16, 31*
(*E*)-hex-2-enol*	845	4.4	0.2	5.4	0.2	2.3	0.2	7.4	0.1	*16, 31*
hexanol*	848	3.9	0.7	5.5	0.2	2.5	0.5	6.0	0.3	*16, 31*
p-xylene°	855	2.1	1.3	3.0	0.1	1.6	1.0	0.6	0.1	*3, 16*
hexa-2,4-dienal[†,°]	872	0.1	0.0	0.1	0.0	tr[g]		0.1	0.0	*3, 16*
3,4-diethylhexa-1,5-diene[†,h,°]	894	0.4	0.0	0.3	0.0	0.4	0.0	0.3	0.0	*16, 30*
3,4-diethylhexa-1,5-diene[†,h,°]	898	0.3	0.0	0.4	0.0	0.4	0.1	0.3	0.0	*16, 30*
isobutyl isobutyrate°	900	0.2	0.0	nd		0.6	0.0	nd		*16, 31*
3-ethylocta-1,5-diene[†,h,°]	931	1.7	0.1	1.5	0.0	1.6	0.1	1.5	0.1	*16, 30*
3-ethylocta-1,5-diene[†,h,°]	938	1.7	0.1	1.4	0.0	2.1	0.1	1.6	0.1	*16, 30*
(*Z*)-hex-3-enyl acetate*	981	0.1	0.0	0.2	0.0	0.1	0.0	1.9	0.0	*16, 31*
deca-3,7-diene[†,h,°]	983	0.5	0.0	0.5	0.0	0.7	0.0	0.7	0.0	*16, 30*
deca-3,7-diene[†,h,°]	986	0.9	0.0	0.9	0.0	1.0	0.0	1.0	0.0	*16, 30*
deca-3,7-diene[†,h,°]	987	1.0	0.0	0.7	0.0	1.5	0.1	0.8	0.0	*16, 30*
β-ocimene[†]	1029	0.9	0.0	0.7	0.0	0.8	0.0	0.1	0.0	*16, 24*
nonanal*	1071	0.3	0.0	0.4	0.0	0.2	0.0	0.3	0.0	*3, 16*
(*Z*)-4,8-dimethylnona-1,3,7-triene°	1092	0.1	0.0	0.1	0.0	0.1	0.0	0.4	0.0	*16, 24*
sesquiterpene	1350	0.5	0.0	0.2	0.0	0.1	0.0	nd		
farnesene[†,°]	1471	nd		0.1	0.0	0.1	0.0	nd		*3, 16, 24*
unknowns [*n* (%)]		39 (8.2)		31 (6.7)		41 (10.3)		31 (8.5)		

[a] Order of elution and percentages of components are given on apolar column (HP-1). [b] Retention indices as determined on HP-1 column using the homologous series of *n*-alkanes. [c] Olive variety. [d] Peak area percent (percent normalized areas) determined by HS-SPME-GC/FID analysis (mean values of three replicates). [e] Standard deviation. [f] Compound not detected. [g] Trace (<0.1%). [h] Attempt at identification. † Correct isomer not characterized. *Structure confirmed by standard compound injection. °Compound identified by literature data (RI and MS).

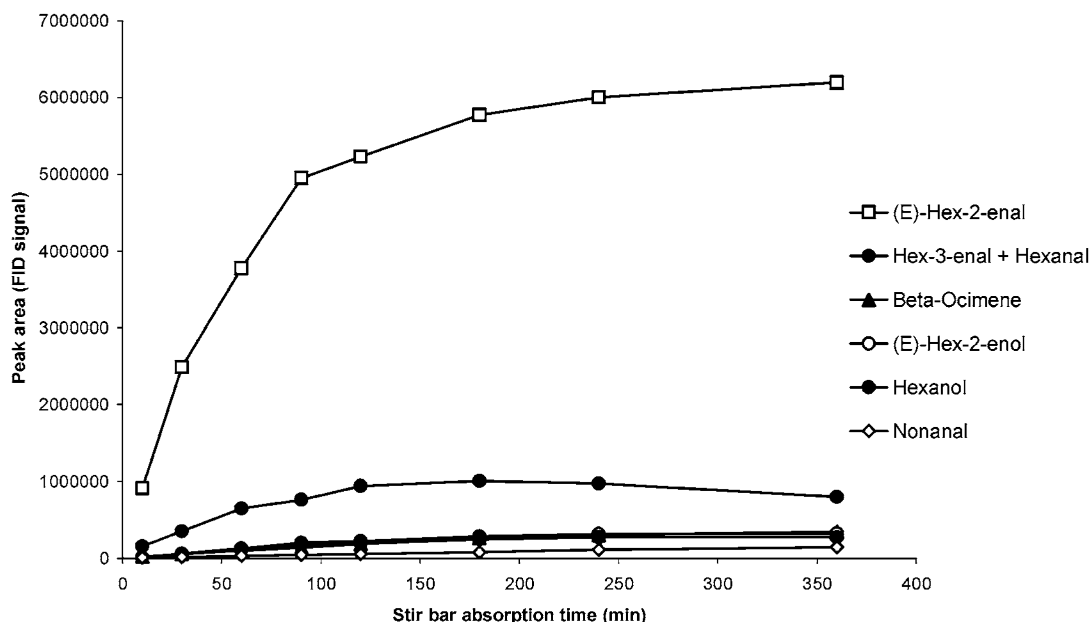

Figure 2. Absorption time profiles of some olive oil volatile compounds (Picholine variety) using a PDMS stir bar.

and order of elution, in line with Angerosa et al. (*30*). These authors also propose, on the basis of previous literature data, a biochemical pathway for these seven compounds deriving from the enzymatic transformation of fatty acids (*30*). (*E*)-Hex-2-enal was the principal compound extracted by SPME in virgin olive oil headspace, and the great majority of the identified components were previously reported in the literature as constituents of olive oil aroma (*16*).

Comparison of SPME and SHS Methods. SPME and SHS have the same advantages previously described. However, SPME is a good extraction technique on olive oil headspace, with a better sensitivity compared to SHS. This is even more

Table 2. Compounds Extracted by HSSE in the Four Olive Oils

		Sabine[c]		Cailletier		Picholine		Koroneiki		
compound[a]	RI[b]	%[d]	SD[e]	%	SD	%	SD	%	SD	refs
ethyl acetate*	598	1.0	0.0	2.2	0.0	0.3	0.0	nd[f]		*16, 31*
2-methylbutanal*	632	2.2	0.0	0.9	0.1	1.3	0.0	1.5	0.0	*3, 16*
penten-3-ol°	651	1.5	0.0	1.1	0.0	1.6	0.0	nd		*16, 31*
pentan-3-one*	659	7.3	0.1	5.5	0.0	4.7	0.1	7.5	0.1	*16, 31*
3-methylbutanol*	708	3.2	0.0	1.0	0.0	0.6	0.1	2.0	0.0	*16, 31*
(*Z*)-pent-2-enol°	740	1.9	0.0	1.5	0.0	0.9	0.2	1.6	0.0	*16, 31*
hex-3-enal†,°	{ 769	9.4	0.8	11.5	0.2	11.2	0.2	9.1	0.1	*16, 31*
hexanal*										*16, 31*
octane*	800	1.9	0.0	1.9	0.1	0.6	0.1	1.3	0.0	*16, 24*
(*E*)-hex-2-enal*	826	46.6	0.9	50.4	0.2	60.3	0.5	45.3	0.3	*16, 31*
(*Z*)-hex-3-enol*	834	3.3	0.1	2.7	0.0	2.9	0.0	5.5	0.0	*16, 31*
(*E*)-hex-2-enol*	845	3.9	0.2	3.7	0.1	1.2	0.0	5.9	0.1	*16, 31*
hexanol*	847	4.4	0.1	5.8	0.1	2.0	0.0	6.4	0.2	*16, 31*
hexa-2,4-dienal†,°	876	0.1	0.0	0.1	0.0	0.1	0.0	0.1	0.0	*3, 16*
isobutyl isobutyrate°	899	nd		nd		0.3	0.0	nd		*16, 31*
(*E*)-hept-2-enal°	927	0.5	0.0	0.4	0.0	0.2	0.0	0.2	0.0	*3, 16*
3-ethylocta-1,5-diene†,h,°	932	1.0	0.0	0.9	0.0	0.9	0.0	0.8	0.0	*16, 30*
3-ethylocta-1,5-diene†,h,°	939	0.9	0.0	0.8	0.0	1.2	0.0	0.8	0.0	*16, 30*
hepta-2,4-dienal†,°	966	0.2	0.0	0.1	0.0	0.1	0.0	0.1	0.0	*3, 16*
(*Z*)-hex-3-enyl acetate*	985	nd		nd		nd		1.4	0.0	*16, 31*
deca-3,7-diene†,h,°	988	0.4	0.0	0.4	0.0	0.4	0.0	0.5	0.0	*16, 30*
deca-3,7-diene†,h,°	991	1.6	0.0	1.2	0.0	1.6	0.0	1.2	0.0	*16, 30*
deca-3,7-diene†,h,°	993	nd		0.5	0.0	nd		0.7	0.0	*16, 30*
β-ocimene†	1024	1.1	0.0	0.8	0.0	0.7	0.0	0.1	0.0	*16, 24*
guaiacol°	1057	0.1	0.0	nd		nd		0.1	0.0	*24, 32*
methyl benzoate*	1065	0.1	0.0	tr[g]		nd		nd		*3, 16*
nonanal*	1067	0.4	0.0	0.5	0.0	0.3	0.0	0.5	0.0	*3, 16*
phenyl ethyl alcohol°	1086	tr		nd		nd		0.1	0.0	*16, 24*
(*Z*)-4,8-dimethylnona-1,3,7-triene°	1090	0.2	0.0	0.1	0.0	0.1	0.0	0.3	0.0	*16, 24*
sesquiterpene	1370	0.1	0.0	tr		nd		nd		
α-copaene	1376	0.3	0.0	0.1	0.0	0.1	0.0	tr	0.0	*16, 24*
farnesene†,°	1486	0.2	0.0	0.4	0.0	0.3	0.0	0.2	0.0	*3, 16, 24*
α-muurolene°	1494	0.1	0.0	tr		tr		nd		*3, 24*
unknowns [*n* (%)]		*37* (6.1)		*44* (5.5)		*45* (6.1)		*38* (6.8)		

[a] Order of elution and percentages of components are given on apolar column (HP-1). [b] Retention indices as determined on HP-1 column using the homologous series of *n*-alkanes. [c] Olive variety. [d] Peak area percent (percent normalized areas) determined by HSSE-GC/FID analysis (mean values of three replicates). [e] Standard deviation. [f] Compound not detected. [g] Trace (<0.1%). [h] Attempt at identification. † Correct isomer not characterized. *Structure confirmed by standard compound injection. °Compound identified by literature data (RI and MS).

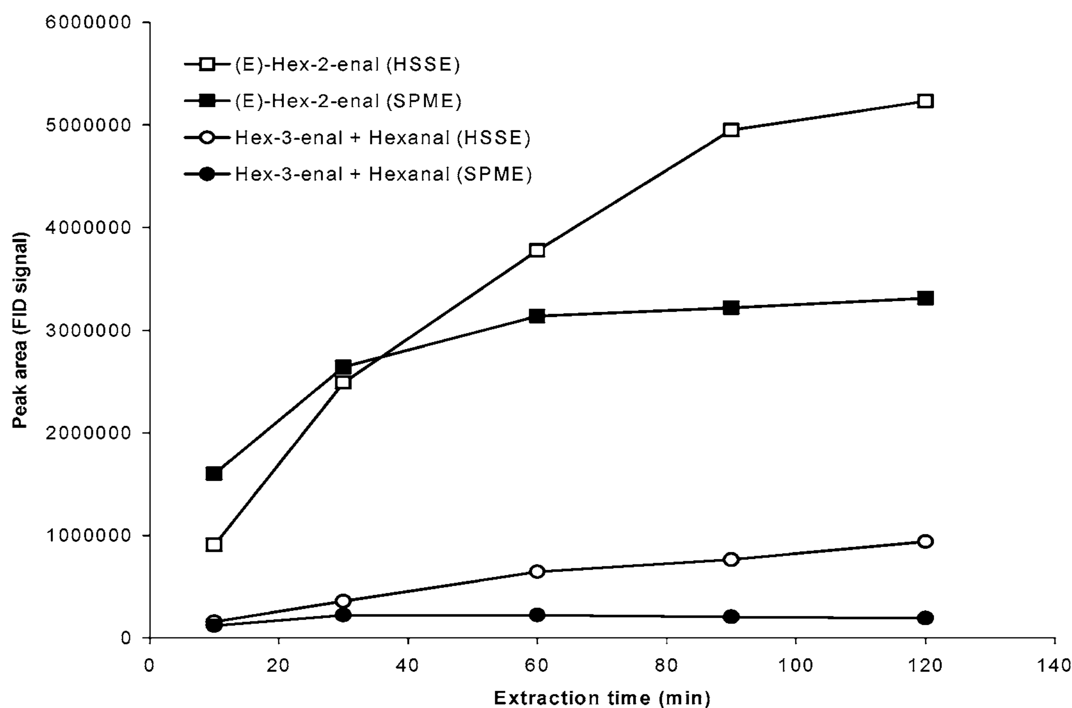

Figure 3. Comparison of performances of the SPME and HSSE extraction techniques for olive oil headspace components (Picholine variety) based on their peak areas (FID signal).

obvious for an identical extraction temperature (25 °C). In addition, good reproducibility was noted during the analysis, with relatively weak standard deviations.

HSSE Analysis. *Optimization of Absorption Time.* To determine the optimal absorption time of the PDMS stir bar to the sample headspace, the stir bar was exposed for time periods

Table 3. Olive Oil Compounds Extracted by Direct Thermal Desorption

		Sabine[c]		Cailletier		Picholine		Koroneiki		
compound[a]	RI[b]	%[d]	SD[e]	%	SD	%	SD	%	SD	refs
hex-3-enal†,°	774	2.3	0.1	3.5	0.4	2.8	0.2	1.6	0.1	*16, 31*
hexanal*										*16, 31*
octane*	801	0.6	0.1	0.6	0.1	nd[f]		0.7	0.2	*16, 24*
(*E*)-hex-2-enal*	824	17.5	0.1	23.8	2.9	21.0	0.9	23.3	1.1	*16, 31*
(*Z*)-hex-3-enol*	834	1.9	0.1	1.7	0.1	1.3	0.1	4.5	0.3	*16, 31*
(*E*)-hex-2-enol*	846	2.0	0.1	2.0	0.5	0.5	0.0	3.7	0.1	*16, 31*
hexanol*	847	4.8	0.3	7.0	0.6	1.4	0.2	9.3	0.7	*16, 31*
styrene*	870	tr[g]		nd		nd		nd		*3, 16*
(*E*)-hept-2-enal°	926	0.6	0.2	0.6	0.2	0.3	0.0	0.5	0.0	*3, 16*
3-ethylocta-1,5-diene†,[h]°	930	0.9	0.3	0.9	0.2	0.8	0.0	0.8	0.0	*16, 30*
3-ethylocta-1,5-diene†,[h],°	937	0.6	0.0	0.6	0.1	0.7	0.0	0.7	0.0	*16, 30*
hepta-2,4-dienal†,°	963	0.4	0.0	nd		nd		0.5	0.0	*3, 16*
(*Z*)-hex-3-enyl acetate*	981	nd		nd		nd		2.1	0.0	*16, 31*
deca-3,7-diene†,[h],°	983	0.4	0.0	0.5	0.1	0.3	0.0	0.8	0.1	*16, 30*
deca-3,7-diene†,[h],°	986	1.6	0.1	1.4	0.2	1.4	0.0	1.6	0.1	*16, 30*
deca-3,7-diene†,[h],°	989	nd		0.6	0.1	nd		1.3	0.1	*16, 30*
benzyl alcohol*	1000	tr		nd		nd		1.0	0.1	*16, 24*
β-ocimene†	1031	2.4	0.1	1.9	0.2	1.4	0.1	0.4	0.0	*16, 24*
guaiacol°	1052	0.5	0.1	nd		nd		0.4	0.0	*24, 32*
methyl benzoate*	1060	0.3	0.0	nd		nd		nd		*3, 16*
nonanal*	1074	3.4	0.3	3.6	0.3	1.7	0.3	2.4	0.2	*3, 16*
phenyl ethyl alcohol°	1080	tr		tr		tr		3.6	0.1	*16, 24*
(*Z*)-4,8-dimethylnona-1,3,7-triene°	1097	0.7	0.0	0.4	0.0	0.2	0.0	1.7	0.1	*16, 24*
(*E*)-non-2-enal°	1130	0.3	0.0	0.3	0.0	0.2	0.0	nd		*16, 32*
p-ethylphenol°	1136	1.0	0.0	tr		0.2	0.0	0.7	0.0	*24*
ethyl benzoate*	1141	0.6	0.0	0.3	0.0	0.2	0.0	0.2	0.0	*3, 24*
methyl salicylate*	1164	0.4	0.0	0.3	0.0	0.6	0.0	0.3	0.0	*3, 24*
p-vinylphenol°	1180	0.4	0.0	tr		0.1	0.0	0.4	0.0	*24*
(*Z*)-dec-2-enal°	1227	1.3	0.1	1.6	0.1	0.8	0.2	2.2	0.3	*24*
4-ethylguaiacol°	1243	0.2	0.0	tr		nd		tr		*32*
deca-2,4-dienal†,°	1258	0.4	0.1	0.3	0.0	0.3	0.1	0.4	0.1	*16, 24*
4-vinylguaiacol°	1272	tr		tr		nd		tr		
deca-2,4-dienal†,°	1277	0.9	0.1	0.5	0.1	0.7	0.2	0.8	0.2	*16, 24*
methyl anisate°	1283	tr		nd		0.2	0.0	0.2	0.0	*24*
sesquiterpene	1364	2.3	0.1	1.1	0.1	0.4	0.0	0.8	0.1	
α-copaene	1369	10.0	0.2	4.0	0.3	1.7	0.1	0.8	0.0	*16, 24*
p-hydroxyphenyl ethyl alcohol°	1379	0.2	0.0	0.1	0.0	nd		0.1	0.0	*24*
6-pentyl-α-pyrone[h],°	1406	0.2	0.0	0.1	0.0	nd		0.4	0.1	
sesquiterpene	1422	0.5	0.0	0.2	0.1	0.1	0.0	0.4	0.1	
sesquiterpene	1464	0.5	0.0	0.3	0.0	0.4	0.0	0.4	0.0	
sesquiterpene	1471	0.3	0.0	0.2	0.0	0.1	0.0	0.4	0.0	
farnesene†,°	1479	20.1	0.7	28.8	4.0	26.1	0.6	11.1	0.4	*3, 16, 24*
α-muurolene°	1486	3.4	0.2	1.3	0.2	0.8	0.0	0.4	0.0	*3, 24*
γ-cadinene	1506	0.2	0.0	0.1	0.0	0.1	0.0	nd		*24*
sesquiterpene	1531	0.4	0.0	0.3	0.1	0.3	0.0	1.2	0.1	
sesquiterpene	1557	0.3	0.0	0.3	0.0	0.3	0.0	0.5	0.1	
sesquiterpene	1563	0.3	0.0	0.3	0.0	0.2	0.0	0.2	0.0	
tetradecanol[h],°	1654	2.6	0.0	1.5	0.3	12.2	0.3	0.7	0.1	
methyl palmitate°	1898	0.1	0.0	nd		nd		nd		
ethyl palmitate*	1966	tr		nd		nd		nd		*3*
methyl oleate°	2077	tr		nd		nd		nd		*3*
unknowns [*n* (%)]		*15* (12.2)		*18* (9.0)		*25* (20.2)		*24* (16.5)		

[a] Order of elution and percentages of components are given on apolar column (HP-1). [b] Retention indices as determined on HP-1 column using the homologous series of *n*-alkanes. [c] Olive variety. [d] Peak area percent (percent normalized areas) determined by DTD-GC/FID analysis (mean values of three replicates). [e] Standard deviation. [f] Compound not detected. [g] Trace (<0.1%). [h] Attempt at identification. † Correct isomer not characterized. *Structure confirmed by standard compound injection. °Compound identified by literature data (RI and MS).

of 10, 30, 60, 90 120, 180, 240, and 360 min. A sampling time of 120 min was chosen to perform the analysis (see **Figure 2**).

Comparison of HSSE and SPME Methods. HSSE analyses of French olive oils show that the same compounds were extracted and identified as in SPME experiments (32 compounds identified against 34 in SPME, see **Table 2**). Indeed, the compound present in greater quantities in the four samples was always (*E*)-hex-2-enal, and we also found the same aldehydes, alcohols, or hydrocarbons, which were previously characterized. Moreover, the stir bar allowed us to extract other sesquiterpenes such as α-copaene or α-muurolene in addition to farnesene.

Comparison of Sampling Performances of HSSE and SPME Methods. **Figure 3** shows the performances of the HSSE sampling technique compared with SPME after 60 min of extraction for the olive oil headspace components based on their peak area (FID signal). The analyses were realized with the same detector on an HP 6890N gas chromatograph. When the stir bar is introduced into the headspace, the liquid/vapor balance is necessarily broken, contrary to SPME (the needle perforates the septum without breaking the balance) for which better performances were observed compared to HSSE in <1 h of extraction. The speed of kinetic extraction can also play a

significant role (bulk retention for HSSE against surface adsorption for SPME). However, beyond 1 h, the PDMS stir bar always shows a higher concentration capacity than the SPME fiber. This performance is entirely due to the higher volume of PDMS coating (55 μL against 0.5 μL for DVB/CAR/PDMS fiber) (*22*).

In the same way, the results obtained by HSSE were much better compared to those obtained in SPME with PDMS coating [volume of the coating material = 0.6 μL (*22*)] during our tests and already described in the literature (comparison of behavior of four fibers coating in SPME and in particular PDMS fiber) (*16*). However, the stir bar was coated with only PDMS, and more coatings for SPME, in particular DVB/CAR/PDMS, could be very effective in studying volatile compounds from olive oils.

The performances of the two extraction techniques, SPME (DVB/CAR/PDMS) and HSSE (PDMS), were "comparable" on the selectivity of extracted compounds. Like SPME, the HSSE analyses showed good reproducibility, but the HSSE technique required the use of a thermal desorption system requiring a more significant investment. However, the stir bar can be thermally desorbed directly into a GC injector liner (*22*).

DTD Analysis. *Optimization of Desorption Conditions*. Many tests were carried out by DTD on olive oil samples. Different desorption temperatures were tested (40, 80, 120, 160, 200, 240, and 280 °C) for two exposure times (10 and 20 min). A desorption temperature of 80 °C for 20 min was chosen to perform the analysis. In addition, the tests implemented at different extraction temperatures (until 280 °C) enabled us to extract esters such as ethyl oleate, fatty acids such as oleic acid, or some hydrocarbons such as squalene.

Comparison of DTD, HSSE, and SPME Methods. The fourth extraction technique tested in this study was direct thermal desorption. This technique allows us to extract more than 60 volatile and semivolatile compounds with a very low quantity of olive oil (5 μL) without any sample preparation (**Table 3**). C_6 aldehyde (19.8−27.3%) and C_6 alcohol (3.2−17.5%) compounds were characterized by DTD-GC/FID and DTD-GC/MS, which had already been identified in SPME and HSSE analysis. However, the semivolatile components were preferentially extracted and represented the main identified compounds: several sesquiterpenes, such as farnesene and α-copaene for the most abundant, or esters derived from the fatty acids, such as methyl palmitate, ethyl palmitate, or methyl oleate. These esters cannot be logically extracted by headspace extraction techniques. Compared to the literature data (*24*−*26*), more semivolatile compounds (mainly sesquiterpenes) were extracted in the four virgin olive oils. DTD is thus a technique perfectly suited to the extraction of semivolatile compounds. The method is also quite reproducible, like that of SPME and HSSE. However, method optimization requires the choice of a desorption temperature >25 °C (a temperature of 80 °C was chosen) to perform the extraction effectively compared to SPME and HSSE. Moreover, there are more risks of oxidation beyond 100 °C.

Comparison of Chemical Composition for the Four French Olive Oils. The differences among varieties (Sabine, Cailletier, Picholine, and Koroneiki) were mainly quantitative, because most compounds were present in all of the olive oils analyzed. The variety Koroneiki is known to have a more significant concentration of esters (*31*) such as (*Z*)-hex-3-enyl acetate, detected in only this olive oil. A direct comparison of the chemical composition of olive oils in the literature data is difficult because of the great variability of the volatile compositions, which depend on several parameters: ripeness stage, extraction technique, or analytical method (*16*, *18*).

In this paper, the characterization of virgin olive oil volatile and semivolatile compounds has been obtained using different extraction techniques. These methods allowed us to identify 61 compounds with a broad range of molecular weights in the four French olive oils from Corsica. SHS was not suited to the characterization of olive oil volatile compounds because of low sensitivity. The recently introduced SPME and HSSE have been successfully applied to olive oil headspace analysis. Both methods enable the characterization of volatile compounds (mainly C_6 aldehydes and alcohols), which contribute significantly to the "green" flavor note of virgin olive oils. The PDMS stir bar showed a higher concentration capacity than the DVB/CAR/PDMS SPME fiber due to the higher volume of coating. DTD was a very good tool for extracting semivolatile compounds from olive oils. However, SPME may be a more appropriate technique for routine quality control due to its operational efficiency (simplicity and repeatability) and low cost.

This technique, coupled with a statistical analysis, will now be applied to many olive oil samples from southern Europe to carry out a varietal distinction.

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