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Analysis of Volatiles in Porcine Liver Pâtés with Added Sage and Rosemary Essential Oils by Using SPME-GC-MS

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The effect of the addition of two natural antioxidant extracts (sage and rosemary essential oils) and one synthetic (BHT) on the generation of volatile compounds in liver pâtés from Iberian and white pigs was analyzed using SPME-GC-MS. Lipid-derived volatiles such as aldehydes [hexanal, octanal, nonanal, hept-(*Z*)-4-enal, oct-(*E*)-2-enal, non-(*Z*)-2-enal, dec-(*E*)-2-enal, deca-(*E*,*Z*)-2,4-dienal] and alcohols (pentan-1-ol, hexan-1-ol, oct-1-en-3-ol) were the most abundant compounds in the headspace of porcine liver pâtés. Pâtés from different pig breeds presented different volatiles profiles due to their different oxidation susceptibilities as a probable result of their fatty acid profiles and vitamin E content. Regardless of the origin of the pâtés, the addition of BHT successfully reduced the amount of volatiles derived from PUFA oxidation. Added essential oils showed a different effect on the generation of volatiles whether they were added in pâtés from Iberian or white pigs because they inhibited lipid oxidation in the former and enhanced oxidative instability in the latter. SPME successfully allowed the isolation and analysis of 41 volatile terpenes from pâtés with added sage and rosemary essential oils including α-pinene, β-myrcene, 1-limonene, (*E*)-caryophyllene, linalool, camphor, and 1,8-cineole, which might contribute to the aroma characteristics of liver pâtés.

KEYWORDS: Liver pâtés; fatty acids; lipid-derived volatiles; volatile terpenes; rosemary; sage; BHT

INTRODUCTION

The study of volatiles in meat and meat products has reached high importance because of the interesting diversity of information given by this type of analysis. For example, the study of the aroma characteristics of a foodstuff as analyzed by its volatiles profile allows the achievement of objective and valuable information (*1*). Many researchers have established close relationships between volatiles profiles and the aroma characteristics of different meat products, shedding light on the mechanisms of generation of volatile compounds (*1*−*5*). Besides, the deterioration of meat and meat products during storage or manipulation can be also evaluated by analyzing volatiles generated as a result of enzymatic, microbial, or biochemical alteration phenomena (*2*, *6*, *7*). Oxidation of lipids is considered to be one of the most important causes of quality degradation in meat and fat products (*8*). Nevertheless, the degradation of lipids during meat cooking and the manufacture of meat and fat products is considered to be necessary to achieve a desirable and specific aroma, because lipid-derived volatiles, such as aldehydes, ketones, and alcohols, are important odor active compounds due to their low molecular weight and low threshold values (*1*).

Liver pâté is a traditional fat product with an increasing demand in European countries such as France, Denmark, and Spain (*9*). This product is highly prone to oxidation due to its high fat content, the presence of large amounts of iron, and the relatively low occurrence of natural antioxidants that justifies the addition of exogenous substances with antioxidant activity (*10*, *11*). Although synthetic antioxidants with phenolic structures, such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), and propyl, octyl, and dodecyl gallates (PG, OG, and DG, respectively), are easily available and largely used in the food industry, the presence of such synthetic compounds in foods has been linked to health risks generally referred to carcinogenic potential (*12*). Consequently, alternative substances with proved antioxidant activity such as sage (*Salvia officinalis*) and rosemary (*Rosmarinus officianalis*) extracts have been successfully introduced to control oxidative deterioration in several types of foodstuffs (*13*−*16*). On the other hand, the origin of the raw material (back fat, liver, and meat) used for the manufacture of liver pâtés determines the physicochemical characteristics and oxidative status exhibited by the elaborated products (*10*). In this sense, liver pâtés from extensively reared Iberian pigs are fairly different from those manufactured with raw material from intensively reared white pigs (*10*). These dissimilarities could have a direct influence on the generation of volatiles because several factors such as the fatty acid composition and non-heme iron (NHI) and tocopherol contents are closely related to the intensity of the oxidation phenomena and the characteristics of the oxidation products (*3*, *7*, *8*).

The purposes of the current work were to analyze the volatile compounds generated in the headspace of liver pâtés as affected by natural (sage and rosemary essential oils) and synthetic (BHT) added antioxidants as well as to evaluate the differences

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between liver pâtés manufactured with raw material from extensively reared Iberian pigs and those manufactured with raw material from intensively reared white pigs regarding their volatiles profiles.

MATERIALS AND METHODS

Animals, Feeds, and Sampling. Raw material [back fat, liver, and muscle (quadriceps femoris)] from two different origins were considered for the manufacture of liver pâtés. Raw material from free-range-reared Iberian pigs ($n = 7$) were obtained from the carcasses after the pigs were slaughtered at ~150 kg of live weight. Raw material from an industrial crossbreed of lean pigs ($n = 7$) (Large White-Landrace × Large White) was obtained after slaughter at 90 kg of live weight. Iberian pigs were extensively reared under traditional schemes and fed with natural resources (acorns and grass), whereas white pigs were intensively reared in a livestock farm and fed a concentrated diet (*10*). Acorn (moisture, 46.10%; fat, 5.50%; protein, 4.31%) analysis (*17*) showed the following fatty acid profile (expressed as percentage of total fatty acids analyzed): palmitic acid (C16:0), 11.82%; stearic acid (C18:0), 0.56%; oleic acid (C18:1), 67.28%; linoleic acid (C18:2), 18.70%; linolenic acid (C18:3), 0.25%. The grass (moisture, 89.24%; fat, 6.26%; protein, 4.34%) fatty acid profile was as follows: C16:0, 13.95%; C18:0, 1.99%; C18:1, 5.24%; C18:2, 11.42%; C18:3, 57.80%. The analysis of the concentrate feed (moisture, 10.42%; fat, 2.94%; protein, 18.28%) revealed the following fatty acid profile: C16:0, 19.86%; C18:0, 8.63%; C18:1, 32.84%; C18:2, 32.83%; C18:3, 2.45%. After slaughter, livers, muscles, and back fat were vacuum packaged and kept frozen (−80 °C) until the manufacture of the liver pâtés (<3 weeks).

Manufacture of Liver Pâtés. The experimental pâtés were manufactured in a pilot plant. The same formulation was used for pâtés from Iberian and white pigs. The ingredients were as follows per 100 g of elaborated product: 28 g of liver, 40 g of back fat, 5 g of muscle, 23 g of distilled water, 2 g of sodium caseinate, and 2 g of sodium chloride. Sodium di- and triphosphates (0.3%), sodium ascorbate (0.05%), and sodium nitrite (0.03%) (Anvisa, Madrid, Spain) were also added. Three groups of pâtés from both Iberian and white pigs were considered depending on the addition of rosemary essential oil (1000 ppm), sage essential oil (1000 ppm), and BHT (200 ppm) dissolved in 10 mL of ethanol. Control pâtés containing no added antioxidants but 10 mL of ethanol were also prepared. According to previous studies (*15*), rosemary and sage oils (Soria Natural, Soria, Spain) exhibit at the aforementioned level their largest antioxidant activity. BHT (Sigma-Aldrich, Steinheim, Germany) was included up to the highest level permitted by Spanish law in this type of fat product. The protocol followed for the manufacture of liver pâtés was profusely explained elsewhere (*10*). The raw mixture of fat, liver, and muscle was packed in a glass container and given the thermal treatment (80 °C/30 min). The packed liver pâtés ($n = 5$ for each group) were kept frozen (−80 °C) until required for analytical experiments (<1 month).

Proximate Composition of Liver Pâtés. Moisture (*17*), total protein (*18*), and ash (*19*) were determined using official methods. The method of Bligh and Dyer (*20*) was used for determining the fat content of liver pâtés. Total iron was determined following the procedure described by Miller et al. (*21*).

Fatty Acid Profiles of Liver Pâtés. Fatty acid methyl esters (FAMEs) were prepared by acidic sterification in the presence of sulfuric acid, following the method of López-Bote et al. (*22*). FAMEs were analyzed using a Hewlett-Packard model HP-5890A gas chromatograph, equipped with a flame ionization detector (FID). The derivatives were separated on an FFAP-TPA fused-silica column (Hewlett-Packard) (30 mm long, 0.53 mm i.d., and 1.0 μm film thickness). The injector and detector temperatures were held at 230 °C. Column oven temperature was maintained at 220 °C. The flow rate of the carrier gas (N_2) was set at 1.8 mL/min. Identification of FAMEs was based on retention times of reference compounds (Sigma-Aldrich). Fatty acid composition was expressed as percent of total FAMEs.

Analysis of Volatiles from Liver Pâtés. The SPME fiber, coated with divinylbenzene/carboxen/poly(dimethylsiloxane) (DVB/CAR/PDMS) 50/30 μm, was purchased from Supelco Co. (Bellefonte, PA). This coating phase was chosen because of the high reproducibility presented and the lower coefficients of variance obtained compared to others, such as the CAR/PDMS fiber (*23*). The SPME fiber was preconditioned prior to analysis at 220 °C during 45 min. The headspace sampling technique was used as follows: 1 g of pâté was placed in 2.5 mL vials. The fiber was exposed to the headspace of the solution while the sample equilibrated during 30 min immersed in water at 60 °C. On the basis of preliminary studies, the sampling method was elected because in those conditions most of the analytes might have reached the equilibrium. Analyses were performed on an HP5890GC series II gas chromatograph (Hewlett-Packard) coupled to a mass-selective detector (Agilent model 5973). Volatiles were separated using a 5% phenyl−95% dimethyl polysiloxane column (Restek) (30 m × 0.25 mm i.d., 1.0 mm film thickness). The carrier gas was helium at 18.5 psi, resulting in a flow of 1.6 mL min^{-1} at 40 °C. The SPME fiber was desorbed and maintained in the injection port at 220 °C during the whole chromatography run. The injector port was in the splitless mode. The temperature program was isothermal for 10 min at 40 °C and then raised at the rate of 7 °C min^{-1} to 250 °C and held for 5 min. *n*-Alkanes (Sigma R-8769) were run under the same conditions to calculate the Kovats index (KI) values for the compounds. The GC-MS transfer line temperature was 270 °C. The mass spectrometer operated in the electron impact mode with an electron energy of 70 eV and a multiplier voltage of 1650 V, collecting data at a rate of 1 scan s^{-1} over a range of *m*/*z* 40 to 300. Volatile compounds were tentatively identified by comparing their mass spectra with those contained in the Wiley and NIST libraries and by comparison of their KI values with those reviewed by Kondjoyan and Berdagué (*24*). Results from the volatiles analysis are provided in area units (AU).

Data Analysis. The effects of pâté origin (Iberian or white pigs) and addition of antioxidants (BHT and sage and rosemary essential oils) on proximate composition and fatty acid profiles of liver pâtés were analyzed using an analysis of variance (ANOVA) for a four (antioxidants) × two (origins) together with the interaction following the generalized linear model (GLM) procedure of SPSS software (11.0 version). Chromatographic areas of all tentatively identified peaks were used as variables. To determinate the effect of the pâté origin and the four different added antioxidants on the generation of volatiles, an ANOVA for a four (antioxidants) × two (origins) together with the interaction was used. Tukey's tests were used when ANOVA found significant differences between treatments. Significance was defined at $p < 0.05$.

RESULTS AND DISCUSSION

Proximate and Fatty Acid Composition of Liver Pâtés. No significant differences between groups were detected for the proximate composition (**Table 1**) because all pâtés presented similar contents of moisture, fat, protein, ash, and iron. Notable differences between types of pâté were detected in the analysis of the fatty acid profile (**Table 1**). Regardless of the addition of antioxidants, pâtés from white pigs presented larger percentages of palmitic, stearic, and total saturated fatty acids (SFA) than pâtés from Iberian pigs ($p < 0.05$). On the other hand, pâtés from Iberian pigs showed a higher proportion of oleic and total monounsaturated fatty acids (MUFA) than pâtés from white pigs ($p < 0.05$). The latter presented higher percentages of polyunsaturated fatty acids (PUFA) such as linoleic acid ($p < 0.05$). As profusely discussed in a previous paper (*10*), the differences in the fatty acid profiles between Iberian and white pâtés are mainly caused by the different fatty acid compositions of the feeds given to the animals during the fattening period. Results obtained in the present work are in good agreement with those reported by other researchers in pâtés made with similar raw material (*25*). Added natural extracts of sage and rosemary had a significant effect on most fatty acids from pâtés from white pigs but, in this case, the differences between groups were less pronounced and did not show a clear

Table 1. Proximate and Fatty Acid Composition of Liver Pâtés from White and Iberian Pigs with Added BHT and Sage and Rosemary Essential Oils

	control		BHT		sage		rosemary			*p* value[a]		
	white	Iberian	white	Iberian	white	Iberian	white	Iberian	SEM[b]	A	O	A×O
moisture[3]	50.51	48.42	50.45	49.1	50.74	49.15	49.69	49.25	0.21	0.774	0.090	0.461
fat[c]	31.82	33.37	32.56	32.01	32.99	33.38	33.7	32.57	0.11	0.858	0.214	0.532
protein[d]	10.04	10.34	10.23	9.81	10.68	10.00	10.36	9.90	0.23	0.391	0.977	0.197
ash[c]	2.78	2.69	2.77	2.76	2.65	2.76	2.85	2.93	0.03	0.102	0.790	0.557
total iron[d]	45.19	50.59	47.27	50.82	49.99	49.04	52.65	50.93	1.46	0.838	0.620	0.821
fatty acids[e]												
C14:0	1.11d	1.12cd	1.12cd	1.14bc	1.17a	1.13cd	1.19a	1.15b	0.00	0.000	0.001	0.000
C16:0	22.65b	20.69cd	22.63b	20.80c	23.26a	20.61d	23.27a	20.62d	0.19	0.000	0.000	0.000
C18:0	13.40b	10.58c	13.44b	10.58c	13.91a	10.50cd	13.88a	10.42d	0.25	0.000	0.000	0.000
Σ SFA[f]	37.98b	32.87cd	38.09b	32.99c	39.11a	32.72d	39.12a	32.69d	0.46	0.000	0.000	0.000
C16:1	2.44a	2.00d	2.42ab	2.00d	2.39c	1.98de	2.40bc	1.96e	0.03	0.000	0.000	0.026
C18:1	43.57c	53.43ab	42.96d	53.38b	42.1f	53.59a	42.39e	53.54ab	0.86	0.000	0.000	0.000
C20:1	1.08d	1.83bc	1.04d	1.79c	0.98e	1.86ab	0.98e	1.90a	0.07	0.004	0.000	0.000
Σ MUFA[f]	47.58c	57.52ab	46.92d	57.42b	45.96f	57.62a	46.24e	57.65a	0.87	0.000	0.000	0.000
C18:2	12.23b	7.71cd	12.54a	7.69d	12.57a	7.82c	12.45a	7.75cd	0.38	0.000	0.000	0.000
C18:3	0.61a	0.49b	0.63a	0.49b	0.62a	0.51b	0.63a	0.50b	0.01	0.097	0.000	0.315
C20:4	0.70a	0.56b	0.78a	0.55b	0.75a	0.57b	0.74a	0.53b	0.02	0.000	0.000	0.093
Σ PUFA[f]	14.40b	9.63c	14.99a	9.57c	14.90a	9.65c	14.90a	9.64c	0.42	0.000	0.000	0.000

[a] *p* values for the studied factors: A, antioxidant; O, pâté origin; A×O, interaction antioxidant × pâté origin. [b] Standard error of the mean. [c] Grams per /100 g of pâté. [d] Micrograms of iron per gram of pâté. [e] Results are expressed as means in percent of methyl esters from total analyzed. In the same row, means with different letters are statistically different. [f] SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids.

pattern. The addition of BHT did not affect the fatty acid profiles of liver pâtés (**Table 1**).

Focusing on the generation of volatiles, PUFA are, contrary to MUFA, very prone to oxidation, leading to the generation of residual substances and unpleasant odors in meat and fat products (*8*). Thus, considerable attention has been given to fatty acids in relation to the generation of volatiles as long as the pathways for the formation of volatile compounds from lipid oxidation are considered to be fairly specific for each fatty acid (*26*).

Analysis of Volatiles from Liver Pâtés. Tables 2 and **3** summarize GC-MS data obtained from the analysis of volatile compounds from liver pâtés. From the total of volatile compounds detected in the extracts, 93 of them were tentatively identified (good match of MS and/or coincidence of KI). To appropriately discuss the results, essential oils-derived terpenes are separately presented in a table from other volatile compounds mainly derived from the liver pâté ingredients (muscle, liver, and adipose tissue).

Analysis of Volatiles Related to Ingredients. Table 2 shows volatile compounds derived from the main ingredients of liver pâté (liver, muscle, and adipose tissue) categorized into nine classes. Lipid-derived volatiles such as aldehydes [hexanal, octanal, nonanal, hept-(*Z*)-4-enal, oct-(*E*)-2-enal, non-(*Z*)-2-enal, dec-(*E*)-2-enal, deca-(*E*,*Z*)-2,4-dienal] and alcohols (pentan-1-ol, hexan-1-ol, oct-1-en-3-ol) were the most abundant compounds in the headspace of liver pâté. A relatively high amount of esters was also detected, whereas ketones, furans, and hydrocarbons were minor components. Results from the present study agree with those obtained in previous works focused on the study of headspace volatiles from cooked pork, oxidized liver, and canned liver sausage (*7*, *27*, *28*). The off-flavors produced as a consequence of the thermal treatment of lipid-rich foods, such as liver pâté, are mainly derived from the autoxidation of lipids (*29*). Moreover, liver pâté exhibits relatively high levels of non-heme iron derived from the liver tissue (*10*) that could promote oxidation phenomena in the manufactured product. In fact, some of the volatile compounds detected in the present study have been described as indicators of lipid decomposition and contributors to the overall off-flavor of oxidized liver (*28*). Great importance has been given to hept-(*Z*)-4-enal due to its low threshold level (0.04 ppb) and has been linked to fishy and unpleasant flavors (*28*, *30*). Some alkadienals such as hepta-(*E*,*E*)-2,4-dienal or nona-2,4-dienal have been associated with the oxidative deterioration of PUFA and have been linked to unpleasant characteristics in cooked liver, with "fishy" notes for the former and "rancid" odors for the latter (*28*, *31*). The oxidation of unsaturated fatty acids undergoes the formation of some other volatile compounds such as non-(Z)-2-enal related to "cardboard"-like odor and deca-(*E*,*Z*)-2,4-dienal) and deca-(*E*,*E*)-2,4-dienal associated with "rancid" and "warmed-over" flavors (*28*, *31*).

Regardless of the addition of antioxidants, liver pâtés from white pigs (control group) showed, compared to those from Iberian pigs, a higher number of lipid-derived volatiles because pentan-2-one, but-3-en-2-one, pentanal, hepta-(*E*,*E*)-2,4-dienal, hexa-2,4-dienal, deca-(*E*,*Z*)-2,4-dienal, and deca-(*E*,*E*)-2,4-dienal were not detected in the headspace of Iberian pâtés. Furthermore, pâtés from white pigs presented significantly ($p < 0.05$) higher chromatographic areas of certain compounds closely related to lipid oxidation and off-flavors such as heptan-1-ol (0.67 vs 0.17 AU), oct-3-en-1-ol (1.86 vs 0.52 AU), octan-1-ol (1.60 vs 0.50 AU), hex-(*E*)-2-en-1-ol (1.03 vs 0.59 AU), heptanal (2.14 vs 0.51 AU), buten-2-enal (2.97 vs 0.62 AU), octanal (3.25 vs 0.65 AU), nonanal (9.95 vs 4.05 AU), oct-(*E*)-2-enal (3.32 vs 0.65 AU), nona-2,4-dienal (2.19 vs 0.51 AU), non-(Z)-2-enal (4.74 vs 0.79 AU), and dec-(*E*)-2-enal (4.65 vs 0.29 AU). Differences between types of pâté are remarkably high for hexanal (white, 21.77 AU; Iberian, 2.66 AU; $p < 0.05$), which has been widely used on meat products as an indicator of lipid oxidation (*1*, *3*, *7*). These results are in agreement with those obtained in a previous work in which the oxidation stability of liver pâtés as measured by TBA-RS was studied (*10*). A higher proportion of MUFA and lower proportion of PUFA in pâtés from Iberian pigs and the presence of significantly ($p < 0.05$) higher amounts of vitamin E in muscles, livers, and adipose tissues from Iberian pigs, compared to those from white pigs (*10*), could partly explain the results obtained in this study.

On the other hand, the large differences between types of pâtés in terms of fatty acid profiles could affect the aromatic characteristics of pâtés as long as the oxidative decomposition of oleic acid leads to the formation of volatile compounds

Table 2. Volatile Compounds Detected in the Headspace of Liver Pâtés from White and Iberian Pigs with Added BHT and Sage and Rosemary Essesntial Oils[a]

	control		BHT		sage		rosemary			*p* value[b]			
compound	white	Iberian	white	Iberian	white	Iberian	white	Iberian	SEM	A	O	A×O	MI[c]
Alcohols													
pentan-1-ol	2.85a	1.50abc	1.10abc	0.90bc	2.64ab	0.14c	1.68abc	0.20c	0.20	0.015	0.000	0.060	MS, KI
hexan-1-ol	2.31bc	1.29bc	2.96b	1.08bc	7.50a	0.48c	1.50bc	0.57c	0.38	0.000	0.000	0.000	MS, KI
heptan-1-ol	0.67b	0.17de	0.43bcd	0.17de	1.08a	0.40cd	0.66bc	0.00e	0.06	0.000	0.000	0.004	MS,KI
oct-1-en-3-ol	1.86ab	0.52d	0.59cd	0.26d	2.33a	0.43d	1.32bc	0.58cd	0.13	0.000	0.000	0.000	MS, KI
2-ethylhexan-1-ol	0.83b	0.52b	0.59b	0.53b	1.52a	0.56b	1.11ab	0.84ab	0.07	0.007	0.001	0.003	MS, KI
hexa-2,4-dien-1-ol	0.49a	0.25b	0.00c	0.00c	0.41ab	0.00c	0.38ab	0.00c	0.03	0.000	0.000	0.000	MS
octan-1-ol	1.60a	0.50cd	1.06b	0.48cd	0.90bc	0.80bcd	0.45d	0.43d	0.07	0.000	0.000	0.000	MS, KI
2,5-dimethylcyclohexanol	1.46a	0.70b	0.92b	0.48bc	1.51a	0.00c	1.74a	0.00c	0.11	0.010	0.000	0.000	MS
3-methylbutan-1-ol (nitrate)	0.44b	0.21c	0.31bc	0.28bc	0.71a	0.00d	0.00d	0.00d	0.04	0.000	0.000	0.000	MS
nonan-1-ol	0.76ab	0.56b	0.60b	0.00c	0.94a	0.00c	0.89a	0.00c	0.06	0.000	0.000	0.000	MS
hex-(*E*)-2-en-1-ol	1.03a	0.59bc	0.60bc	0.48c	0.89ab	0.00d	1.06a	0.00d	0.07	0.000	0.000	0.000	MS
Acids													
acetic acid	0.00b	0.00b	0.00b	0.00b	0.82a	0.00b	0.64a	0.00b	0.06	0.000	0.000	0.000	MS, KI
Esters													
acetic acid ethyl ester	0.00b	0.00b	0.00b	0.00b	0.51a	0.00b	0.51a	0.00b	0.04	0.000	0.000	0.000	MS, KI
hexanoic acid ethyl ester	1.86abc	2.00ab	1.95ab	0.91cd	2.28a	0.67d	1.26bcd	0.46d	0.12	0.000	0.000	0.003	MS, KI
heptanoic acid ethyl ester	0.73ab	0.60bc	0.75ab	0.32c	0.95a	0.40c	0.61bc	0.50bc	0.04	0.090	0.000	0.003	MS, KI
octanoic acid ethyl ester	6.37a	6.92a	7.18a	5.88ab	3.03bc	2.59c	1.90c	1.84c	0.41	0.000	0.506	0.560	MS, KI
nonanoic acid ethyl ester	3.27a	1.06b	3.16a	3.45a	2.97a	1.30b	1.19b	0.79b	0.20	0.000	0.000	0.001	MS, KI
decanoic acid ethyl ester	10.60b	6.18cd	6.87c	14.27a	4.89cd	4.68cd	3.73d	4.41cd	0.59	0.000	0.081	0.000	MS
dodecanoic acid ethyl ester	3.15a	1.00b	1.24b	4.35a	1.30b	1.64b	0.71b	1.08b	0.21	0.000	0.052	0.000	MS
tetradecanoic acid ethyl ester	3.29ab	0.89c	1.13c	4.97a	1.52bc	1.63bc	1.01c	0.71c	0.26	0.000	0.305	0.000	MS
hexadecanoic acid ethyl ester	3.80ab	1.15c	1.34c	5.10a	1.27c	1.72bc	1.28c	1.08c	0.27	0.000	0.303	0.000	MS
Ketones													
cycloex-2-en-1-one	0.57c	0.54c	0.00c	0.58c	3.27b	0.25c	4.30a	0.00c	0.25	0.000	0.000	0.000	MS, KI
pentan-2-one	0.70b	0.00c	0.11c	0.00c	1.27a	0.00c	0.75b	0.00c	0.08	0.000	0.000	0.000	MS
but-3-en-2-one	1.11a	0.00b	0.00b	0.00b	1.22a	0.00b	1.08a	0.00b	0.09	0.000	0.000	0.000	MS, KI
Aldehydes													
pentanal	4.16a	0.00c	1.48bc	0.00c	4.76a	0.00c	3.10ab	0.00c	0.36	0.038	0.000	0.038	MS
hexanal	21.77a	2.26b	2.68c	0.25c	21.19a	0.29c	16.96b	0.31c	1.52	0.000	0.000	0.000	MS, KI
3-(methylthio)propanal	1.15a	1.16a	1.38a	0.88b	0.00c	0.00c	0.00c	0.00c	0.10	0.000	0.003	0.000	MS
heptanal	2.14a	0.51bc	0.99b	0.36bc	2.20a	0.11c	1.13b	0.46bc	0.13	0.002	0.000	0.000	MS, KI
buten-2-enal	2.97a	0.62bc	0.98b	0.53bc	0.80bc	0.12c	0.70bc	0.28c	0.14	0.000	0.000	0.000	MS, KI
benzaldehyde	3.60a	1.19cd	2.94ab	3.05ab	2.11bc	0.48d	2.22bc	0.56d	0.19	0.000	0.000	0.000	MS, KI
octanal	3.25b	0.65c	0.59c	0.42c	5.28a	0.38c	4.42ab	0.44c	0.32	0.000	0.000	0.000	MS, KI
benzeneacetaldehyde	1.75a	0.72b	0.69b	1.51a	0.85b	0.31b	0.87b	0.42b	0.08	0.000	0.002	0.000	MS, KI
nonanal	9.95a	4.05b	3.02b	2.82b	11.88a	0.89b	11.51a	0.85b	0.77	0.001	0.000	0.000	MS
hept-(*Z*)-4-enal	1.41c	0.42c	0.47c	0.51c	11.07a	0.63c	7.37b	0.40c	0.64	0.000	0.000	0.000	MS, KI
hepta-(*E*,*E*)-2,4-dienal	1.09a	0.00b	0.00b	0.00b	0.97a	0.00b	0.94a	0.00b	0.09	0.000	0.000	0.000	MS, KI
pent-4-enal	0.27c	0.32c	0.68bc	0.39c	5.03a	0.30c	2.04b	0.26c	0.27	0.000	0.000	0.000	MS
oct-(*E*)-2-enal	3.32b	0.65c	0.40c	0.32c	7.23a	0.00c	1.06c	0.19c	0.40	0.000	0.000	0.000	MS, KI
nona-2,4-dienal	2.19b	0.51c	0.47c	0.00c	3.99a	0.00c	3.66a	0.00c	0.26	0.000	0.000	0.000	MS, KI
non-(*Z*)-2-enal	4.74b	0.79cd	0.67cd	0.11cd	7.56a	0.00d	1.69c	0.00d	0.43	0.000	0.000	0.000	
hexa-2,4-dienal	0.61c	0.00c	0.00c	0.00c	4.90a	0.00c	1.98b	0.00c	0.27	0.000	0.000	0.000	MS, KI
dec-(E)-2-enal	4.65b	0.29c	0.38c	0.34c	8.99a	1.09c	2.07bc	0.19c	0.51	0.000	0.000	0.000	MS, KI
deca-(*E*,*E*)-2,4-dienal	1.43ab	0.00c	0.35c	0.00c	2.26a	0.00c	0.67bc	0.00c	0.14	0.000	0.000	0.000	MS, KI
deca-(*E*,*Z*)-2,4-dienal	3.67b	0.00c	0.40c	0.00c	8.64a	0.00c	3.48b	0.00c	0.50	0.000	0.000	0.000	MS, KI
Hydrocarbons													
decane	2.30b	1.73bc	3.30a	1.35cd	1.73bc	0.59de	1.38c	0.45e	0.15	0.000	0.000	0.003	MS
non-1-en-3-yne	0.00c	0.00c	0.00c	0.00c	3.19a	0.00c	1.78b	0.99bc	0.19	0.000	0.000	0.000	MS
Furans													
dihydrofuran-2-one	0.62b	0.00c	0.00c	0.00c	1.45a	0.00c	1.28a	0.00c	0.09	0.000	0.000	0.000	MS, KI
2-pentylfuran	0.66bc	0.33c	0.29c	0.16c	3.93a	0.45bc	1.62b	0.10c	0.21	0.000	0.000	0.000	MS, KI
2,5-dihydrofuran	0.94bc	0.37c	0.58c	0.00c	6.41a	0.00c	2.47b	0.00c	0.35	0.000	0.000	0.000	MS, KI
Nitrogen Compounds													
2-methylpyridine	0.00c	0.00c	0.00c	0.00c	0.83a	0.64ab	0.70ab	0.50b	0.06	0.000	0.000	0.043	MS
Others													
1-ethyl-2,3-dimethylbenzene	0.00c	0.00c	0.00c	0.00c	1.49a	0.00c	0.42b	0.00c	0.08	0.000	0.000	0.000	MS, KI
1-methyl-4-methylethyl-benzene	0.00c	0.00c	0.00c	0.00c	80.22ab	96.69a	75.30b	73.66b	6.76	0.000	0.266	0.186	MS, KI
BHT	0.00b	0.00b	1584.65a	1679.90a	0.00b	0.00b	0.00b	0.00b	190.93	0.000	0.356	0.292	MS

[a] Values are means (area units × 10^6) of five analyses. In the same row, means with different letters are statistically different. [b] *p* values for the studied factors: A, antioxidant; O, pâté origin; A×O, interaction antioxidant × pâté origin. [c] Method of odentification: MS, mass spectrum comparison using Wiley and NIST libraries; KI, Kovats index in agreement with literature values.

Table 3. Volatile Terpenes Detected in the Headspace of Liver Pâtés from White and Iberian Pigs with Added Sage and Rosemary Essential Oils[a]

	sage		rosemary				
compound	white	Iberian	white	Iberian	SEM	*p* value[b]	MI[c]
			Monoterpene Hydrocarbons				
α-thujene	5.27b	7.56a	2.45c	2.41c	0.54	0.000	MS
α-pinene	83.22c	107.82bc	211.17ab	251.65a	20.84	0.002	MS, KI
β-1-pinene	69.16	64.62	72.63	44.46	5.92	0.354	MS
β-2-pinene	1.13b	0.78b	1.66a	1.72a	0.11	0.000	MS
δ-3-carene	1.46a	0.84b	0.56b	0.58b	0.11	0.001	MS, KI
α-fenchene	8.64	8.91	8.36	9.66	0.56	0.885	MS, KI
camphene	84.58	102.23	58.56	61.67	6.86	0.066	MS, KI
γ-terpinene	1.46a	1.16a	0.00b	0.00b	0.16	0.000	MS
β-terpinene	14.80a	14.59a	2.27b	1.53b	1.89	0.002	MS
α-terpinene	1.48b	1.76ab	2.18ab	2.32a	0.12	0.041	MS
β-myrcene	62.85ab	89.01a	48.00b	51.33b	5.53	0.021	MS
tricyclene	3.46a	2.49ab	2.08b	1.81b	0.20	0.005	MS, KI
1-limonene	301.23a	391.69a	119.20b	122.88b	30.91	0.000	MS, KI
(*E*)-ocimene	3.55b	4.74a	0.00c	0.00c	0.47	0.000	MS
β-ocimene	5.29	6.75	6.10	6.82	0.25	0.099	MS
α-terpinolene	2.85c	5.39a	3.35bc	4.13b	0.25	0.000	MS, KI
isoterpinolene	6.54b	9.04a	7.96ab	6.74b	0.32	0.008	MS
allo-cimene	20.07a	21.17a	0.00b	0.00b	2.45	0.000	MS, KI
			Sesquiterpene Hydrocarbons				
α-cubenene	20.07a	21.17a	0.00b	0.00b	0.13	0.000	MS, KI
farnesol	1.44b	2.11a	0.00c	0.00c	0.25	0.000	MS, KI
α-ylangene	2.40a	1.90b	0.00c	0.00c	0.09	0.000	MS
α-copaene	0.00b	0.00b	0.75a	0.76a	0.24	0.000	MS, KI
α-gurjunene	0.89a	0.85a	0.00b	0.00b	0.11	0.000	MS, KI
junipene	2.09a	2.32a	0.00b	0.00b	0.26	0.000	MS
(*E*)-caryophyllene	24.62b	27.72b	56.40a	60.50a	3.89	0.000	MS, KI
β-selinene	6.15	7.20	5.82	6.15	0.22	0.134	MS
α-elemene	0.00b	0.00b	2.13a	2.11a	0.25	0.000	MS
δ-cadinene	1.46ab	1.12b	1.86a	1.91a	0.11	0.014	MS, KI
			Terpenoid Alcohols				
linalool	73.70a	81.88a	37.63b	37.17b	5.07	0.000	MS, KI
p-menth-3-en-1-ol	5.34a	5.03a	1.36b	1.57b	0.45	0.000	MS, KI
α-terpineol	3.14	2.79	3.30	2.87	0.15	0.630	MS
endo-borneol	28.87	30.40	32.63	31.87	0.84	0.432	MS, KI
terpinene-4-ol	13.20a	14.24a	7.95b	7.84b	0.77	0.000	MS
p-cymen-8-ol	2.87a	2.76a	0.59b	0.59b	0.27	0.000	MS, KI
			Terpenoid Esters				
linalyl acetate	161.76a	152.26a	6.48b	7.25b	17.64	0.000	MS
linalyl propionate	64.24ab	73.73a	52.69b	55.09b	2.46	0.002	MS
endo-bornyl acetate	16.13	16.53	19.80	21.10	1.16	0.365	MS, KI
geranyl propionate	0.00c	0.00c	1.02b	1.17a	0.22	0.000	MS, KI
			Terpenoid Carbonyls				
camphor	1007.11a	1026.35a	496.87b	499.16b	64.74	0.000	MS, KI
			Other Terpenoids				
α-phellandrene epoxide	26.88a	34.90a	0.00b	0.00b	3.68	0.000	MS, KI
1,8-cineole	766.18b	665.81b	1012.46a	1009.49a	58.50	0.046	MS, KI

[a] Values are means (area units × 10^6) of five analyses. In the same row, means with different letters are statistically different in Tukey's test. [b] Statistical significance. [c] Method of identification: MS, mass spectrum comparison using Wiley and NIST libraries; KI, Kovats index in agreement with literature values.

associated with pleasant notes, described as "floral" and "sweet" (*32*), whereas the aromatic notes of linoleic and PUFA-derived volatiles have been described as intense "grass-like" and related to rancidity in cooked meat and porcine liver (*28*, *33*). Consistent with results from fatty acid profiles, the ratio between oleic-derived volatiles (octanal, nonanal, octan-1-ol) and linoleic-derived volatiles [hexanal, oct-(*E*)-2-enal, non-(*Z*)-2-enal] was found to be significantly higher in pâtés from Iberian pigs (Iberian, 1.41; white, 0.50; $p < 0.05$), suggesting a more pleasant aromatic profile in the latter. The high content of oleic acid and its oxidation-derived aldehydes in meat products from Iberian pigs has been related to essential quality traits (*7*, *34*, *35*).

The addition of BHT and essential oils of sage and rosemary on liver pâtés had a significant effect on the generation of most volatiles (**Table 2**). Regardless of the origin of the pâté, the addition of BHT successfully inhibited the development of oxidative deterioration because the presence of major lipid-derived volatiles in the headspace of pâtés decreased when this antioxidant was added. The antioxidant effect of BHT on meat products has been largely reported in the scientific literature (*14*, *15*). In the present work, the addition of BHT had the most evident effect on those pâtés with higher oxidative instability (pâtés from white pigs). Compared to the control group, pâtés from white pigs with added BHT presented significantly ($p < 0.05$) smaller amounts of oct-3-en-1-ol (1.86 vs 0.52 AU), octan-1-ol (1.60 vs 1.06 AU), pentan-2-one (0.70 vs 0.11 AU), pentanal (4.16 vs 1.48 AU), hexanal (21.77 vs 2.26 AU), heptanal (2.14 vs 0.99 AU), buten-2-enal (2.97 vs 0.98 AU), octanal (3.25 vs 0.59 AU), nonanal (9.95 vs 3.02 AU), oct-(*E*)-2-enal (3.32 vs 0.40 AU), non-(*Z*)-2-enal (4.74 vs 0.67 AU), nona-2,4-dienal (2.19 vs 0.47 AU), dec-(*E*)-2-enal (4.65 vs 0.38 AU), deca-(*E*,*E*)-2,4-dienal (1.43 vs 0.35 AU), and deca-(*E*,*Z*)-2,4-dienal (3.67 vs 0.40 AU), among others.

On the other hand, the addition of sage and rosemary essential oils had a significant effect on the generation of major volatile compounds, but this effect was different depending on whether they were added on Iberian or white pâtés. In fact, the interaction between "origin of pâté" and "antioxidant" was found to be significant for most volatiles (**Table 2**). In agreement with previous research on several meats and meat products (*13*, *14*), the addition of sage and rosemary oils had an antioxidant effect on pâtés from Iberian pigs as long as smaller amounts of hexanal, nonanal, and other lipid-derived volatiles were detected in the headspace of treated pâtés when compared to the control counterparts. In contrast, the addition of sage and rosemary essential oils in pâtés from white pigs had an opposite behavior, significantly increasing ($p < 0.05$) the formation of volatiles generated from PUFA and associated with "fishy" and unpleasant flavors in liver products (*28*) such as hept-(*Z*)-4-enal (control, 1.41 AU; sage, 11.88 AU; rosemary, 11.51 AU), pent-4-enal (control, 0.27 AU; sage, 5.03 AU; rosemary, 2.04 AU), hexa-2,4-dienal (control, 0.61 AU; sage, 4.90 AU; rosemary, 1.98 AU), nona-2,4-dienal (control, 2.19 AU; sage, 3.99 AU; rosemary, 3.66 AU), and deca-(*E*,*Z*)-2,4-dienal (control, 3.67 AU; sage, 8.64 AU; rosemary, 3.48 AU). Although the antioxidant activity of plant phenolics extracts is generally recognized (*36*), the pro-oxidant properties of these substances have also been described, being able to generate reactive oxygen species and damage lipids, proteins, and other cellular components (*37*, *38*). Results from the present work suggest that the activity of essential oils of sage and rosemary is dependent on the compositional characteristics of the food matrix. In fact, the activity of plant phenolics on food systems has been considered to be influenced by the presence of other active substances in the food matrix (*15*, *38*). Food systems, and particularly liver pâtés, are very complex in the number and type of chemicals in the mixture, and a particular combination of these compounds might behave differently from the individual components. In this sense, Wong et al. (*13*) and Fang and Wada (*39*) reported possible interactions between phenolic compounds from sage and rosemary essential oils and vitamin E, resulting in different activities depending on the individual amounts of these substances in the food system. Significant differences ($p < 0.05$) were found between Iberian and white pigs regarding vitamin E content in muscles (6.18 vs 1.94 mg/kg of muscle), livers (7.93 vs 3.49 mg/kg of liver), and adipose tissues (19.67 vs 1.21 mg/kg of adipose tissue) used for the manufacture of liver pâtés (*10*). The presence of a certain amount of an endogenous antioxidant (vitamin E) in the raw material and manufactured product might influence the activity of exogenous active extracts, leading to antioxidant or pro-oxidant effects.

Analysis of Terpenes from Sage and Rosemary Extracts. SPME allowed the isolation and analysis of 41 terpenes derived from sage and rosemary essential oils (**Table 3**). Monoterpene hydrocarbons such as α-pinene, camphene, *β*-myrcene, and 1-limonene, sesquiterpene hydrocarbons such as α-cubenene and (*E*)-caryophyllene, and oxygen-derivative terpenes such as alcohols (linalool, endo-borneol, terpinene-4-ol), esters (linalyl acetate, linalyl propionate), carbonyls (camphor), and ethers (1,8-cineole) were the most abundant. Most of these compounds have been previously reported as volatile components of sage and rosemary essential oils and isolated in the headspace of several spiced foods (*4*, *40*, *41*). As expected, no differences were detected between white and Iberian pâtés as long as the same formulation was used for all of them. Although most volatile terpenes were detected in both groups, significant differences were detected between sage and rosemary essential oils with regard to their terpene profile. Compared to that from pâtés with added sage extract, headspace from pâtés with added rosemary presented significantly ($p < 0.05$) higher amounts of α-pinene, *β*-2-pinene, and 1,8-cineole, whereas the former showed higher amounts of *γ*-terpinene, *β*-terpinene, 1-limonene, *allo*-cimene, and most abundant terpenoids such as linalool and their esterified derivatives, *p*-menth-3-en-1-ol, terpinene-4-ol, *p*-cymen-8-ol and camphor, among others. However, the main differences between essential oils were detected for sesquiterpene hydrocarbons because pâtés with added sage extract showed relatively high amounts of certain compounds such as α-cubenene, farnesol, α-ylangene, α-gurjunene, and junipene, which were not detected in pâtés with added rosemary extract. Other compounds such as α-copaene and α-elemene were detected only in pâtés with added rosemary extract. Several of the volatile terpenes detected are recognized odorants and are commonly used in the food industry as flavor and fragrance ingredients (*40*). Volatile terpenes such as α-pinene, 1,8-cineole, and linalool have been related to "spices, pine needles", "medicinal, cough syrup", and "flowers, carnation" odors, respectively, and have been reported as contributors to the aroma of spiced cooked sausages (*4*). In the absence of olfactometry or sensory assessment of pâtés, the contribution of these compounds to the overall aroma of pâtés remains unknown, and, therefore, the attitude of consumers toward pâtés with odor notes referred to such aromatic herbs would be a future work of interest. On the other hand, using deodorized extracts of these plants would be also an interesting option to achieve antioxidant effects in meat and fat products without including unexpected aroma components (*42*).

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LITERATURE CITED

(1) Mottram, D. S. Flavour formation in meat and meat products: a review. *Food Chem.* **1998**, *62*, 415−424.

(2) Vergnais, L.; Masson, F.; Montel, M. C.; Berdagué, J. L.; Talon, R. Evaluation of solid-phase microextraction for analysis of volatile metabolites produced by Staphylococci. *J. Agric. Food Chem.* **1998**, *46*, 228−234.

(3) Elmore, J. S.; Mottram, D. S.; Enser, M.; Wood, J. D. Effect of the polyunsaturated fatty acid composition of beef muscle on the profile of aroma volatiles. *J. Agric. Food Chem.* **1999**, *47*, 1619−1625.

(4) Chevance, F. F. V.; Farmer, L. J. Identification of major volatile odor compounds in frankfurters. *J. Agric. Food Chem.* **1999**, *47*, 5151−5160.

(5) Chevance, F. F. V.; Farmer, L. J. Release of volatile odor compounds from full-fat and reduced-fat frankfurters. *J. Agric. Food Chem.* **1999**, *47*, 5161−5168.

(6) Vinauskiene, R.; Venskutonis, P. R.; Posthumus, M. A. Changes of volatile compounds in poultry meat during storage. In *Proceedings of the 48th Congress of Meat Science and Technology*; 2002; pp 292−293.

(7) Estévez, M.; Morcuende, D.; Ventanas, S.; Cava, R. Analysis of volatiles in meat from Iberian and lean pigs after refrigeration and cooking by using SPME-GC-MS. *J. Agric. Food Chem.* **2003**, *51*, 3429−3435.

(8) Morrissey, P. A.; Sheehy, P. J. A.; Galvin, K.; Kerry, J. P.; Buckley, D. J. Lipid stability in meat and meat products. *Meat Sci.* **1998**, *49*, S73−S86.

(9) Rosmini, M. R.; Perlo, F.; Pérez-Alvarez, J. A.; Pagán-Moreno, M. J.; Gago-Gago, A.; López-Santoveña, F.; Aranda-Catalá, V. TBA test by an extractive method applied to 'Paté'. *Meat Sci.* **1996**, *42*, 103−110.

(10) Estévez, M.; Morcuende, D.; Ramírez, M. R.; Ventanas, J.; Cava, R. Extensively reared Iberian pigs versus intensively reared white pigs for the manufacture of liver pâté. *Meat Sci*. **2004**, *67*, 453−461.

(11) Madhavi, D. L.; Despande, S. S.; Salunkhe, D. K. Toxicological aspects and food antioxidants. In *Food Antioxidants*; Dekker: New York, 1996; pp 267−311.

(12) Clayson, D. B.; Iverson, F.; Nera, E.; Lok, E.; Rogers, C.; Rodrigues, C. Histopathological and radioautographical studies on the forestomach of F344 rats treated with butylated hydroxyanisole and related chemicals. *Food Chem. Toxicol*. **1986**, *24*, 1171−1182.

(13) Wong, J. W.; Hashimoto, K.; Shibamoto, T. Antioxidant activities of rosemary and sage extracts and vitamin E in a model system. *J. Agric. Food Chem.* **1995**, *43*, 2707−2712.

(14) Chen, X.; Jo, C.; Lee, J. I.; Ahn, D. U. Lipid oxidation, volatiles and color changes of irradiated pork patties as affected by antioxidants. *J. Food Sci.* **1999**, *64*, 16−19.

(15) McCarthy, T. L.; Kerry, J. P.; Kerry, J. F.; Lynch, P. B.; Buckley, D. J. Evaluation of the antioxidant potential of natural food/plant extracts as compared with synthetic antioxidants and vitamin E in raw and cooked pork patties. *Meat Sci*. **2001**, *57*, 45−52.

(16) Yu, L.; Scanlin, L.; Wilson, J.; Schmidt, G. Rosemary extracts as inhibitors of lipid oxidation and color change in cooked turkey products during refrigerated storage. *J. Food Sci*. **2002**, *67*, 582−585.

(17) AOAC. Moisture content. 950.46. In *Official Methods of Analysis*, 17th ed.; Association of Official Analytical Chemists: Gaithersburg, MD, 2000.

(18) AOAC. Protein content in meat. 928.08. In *Official Methods of Analysis*, 17th ed.; Association of Official Analytical Chemists: Gaithersburg, MD, 2000.

(19) AOAC. Ashes content. 920.153. In *Official Methods of Analysis*, 17th ed.; Association of Official Analytical Chemists: Gaithersburg, MD, 2000.

(20) Bligh, E. G.; Dyer, W. J. A rapid method of total lipid extraction and purification. *Can. J. Biochem. Phys.* **1959**, *37*, 911−917.

(21) Miller, D. K.; Gómez-Basauri, J. V.; Smith, V. L.; Kanner, J.; Miller, D. D. Dietary iron in swine rations affects nonheme iron and TBARS in pork skeletal muscles. *J. Food Sci.* **1994**, *59*, 747−750.

(22) López-Bote, C. J.; Rey, A.; Sanz, M.; Gray, J. L.; Buckley, J. D. Dietary vegetable oils and α-tocopherol reduce lipid oxidation in rabbit muscle. *J. Nutr.* **1997**, *127*, 1176−1182.

(23) Machiels, D.; Istasse, L. Solid-phase microextraction evaluation of key aroma compounds in cooked beef meat as influenced by breed and diet. In *Proceedings of the 48th Congress of Meat Science and Technology*; 2002; pp 262−263.

(24) Kondjoyan, N.; Berdagué, J. L. A *Compilation of Relative Retention Indices of the Analysis of Aromatic Compounds*; Laboratoire flaveur, station de recherches sur la viande, INRA: Theix, France, 1996.

(25) Ordóñez, J. A.; D'Arrigo, M.; Cambero, M. I.; Pinc, C.; de la Hoz, L. Características de patés de hígado de cerdos alimentados con dietas ricas en PUFA n-3 y vitamina E. In *Proceedings Congreso Nacional de Ciencia y Tecnología de Alimentos*, Granada, Spain, 2003; p 184.

(26) Chen, J.; Ho, C.-T. The flavour of pork. In *Flavor of Meat, Meat Products and Seafoods*; Shahidi, F., Ed.; Blackie Academic and Professional: London, U.K., 1998; pp 61−83.

(27) Ruiz, J.; Ventanas, J.; Cava, R. New device for direct extraction of volatiles in solid samples using SPME. *J. Agric. Food Chem*. **2001**, *49*, 5115−5121.

(28) Im, S.; Hayakawa, F.; Kurata, T. Identification and sensory evaluation of volatile compounds in oxidized porcine liver. *J. Agric. Food Chem*. **2004**, *52*, 300−305.

(29) Labuza, T. P. Kinetics of lipid oxidation in foods. *Crit. Rev. Food Technol*. **1971**, *2*, 355−405.

(30) McGill, A. S.; Hardy, R.; Gunstone, F. D. Further analysis of the volatile components of frozen cold stored cod and the influence of these on flavour. *J. Sci. Food Agric.* **1977**, *28*, 200−205.

(31) Lee, H.; Kizito, S. A.; Weese, S. J.; Craig-Schmidt, M. C.; Lee, Y.; Wei, C.-I.; An, H. Analysis of headspace volatile and oxidized volatile compounds in DHA-enriched fish oil on accelerated oxidative storage. *J. Food Sci.* **2003**, *68*, 2169−2177.

(32) Specht, K.; Baltes, W. Identification of volatile flavor compounds with high aroma values from shallow-fried beef. *J. Agric. Food Chem.* **1994**, *24*, 2246−2253.

(33) Shahidi, F.; Pegg, R. B. Hexanal as an indicator of meat flavour deterioration. *J. Food Lipids* **1993**, *1*, 177−186.

(34) Ruiz, J.; Ventanas, J.; Cava, R.; Andrés, A. I.; García, C. Volatile compounds of dry cured Iberian ham as affected by the lengh of the curing process. *Meat Sci*. **1999**, *52*, 19−27.

(35) Cava, R.; Ventanas, J.; Tejeda, J. F.; Ruiz, J.; Antequera, T. Effect of free-range rearing and α-tocopherol and copper supplementation on fatty acid profiles and susceptibility to lipid oxidation of fresh meat from Iberian pigs. *Food Chem*. **2000**, *68*, 51−59.

(36) Huang, S.; Frankel, E. N.; Schwarz, K.; Aeschbach, R.; German, J. B. Antioxidant activity of carnosic acid and methyl carnosate in bulk oil and oil-in-water emulsions. *J. Agric. Food Chem*. **1996**, *44*, 951−956.

(37) Aruoma, O. I.; Halliwell, B.; Aeschbach, R.; Loliger, J. Antioxidant and pro-oxidant properties of active rosemary constituents: carnosol and carnosic acid. *Xenobiotics* **1992**, *22*, 257−268.

(38) Yen, G.-C.; Chen, H.-Y.; Peng, H.-H. Antioxidant and pro-oxidant effects of various tea extracts. *J. Agric. Food Chem*. **1997**, *45*, 30−34.

(39) Fang, X.; Wada, S. Enhancing the antioxidant effect of α-tocopherol with rosemary in inhibiting catalized oxidation caused by Fe^{2+} and hemoprotein. *Food Res. Int.* **1993**, *26*, 405−411.

(40) Ibáñez, E.; Oca, A.; de Murga, G.; López-Sebastián, S.; Tabera, J.; Reglero, G. Supercritical fluid extraction and fractionation of different pre-processed rosemary plants. *J. Agric. Food Chem*. **1999**, *47*, 1400−1404.

(41) Paleari, M. A.; Moretti, V. M.; Bersani, C.; Beretta, G.; Mentasti, G. Characterisation of a lard cured with spices and aromatic herbs. *Meat Sci.* **2004**, *67*, 549−557.

(42) Dorman, H. J. D.; Peltoketo, A.; Hiltunen, R.; Tikkanen, M. J. Characterisation of the antioxidant properties of de-odourised aqueous extracts from selected Lamiaceae herbs. *Food Chem*. **2003**, *83*, 255−262.

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