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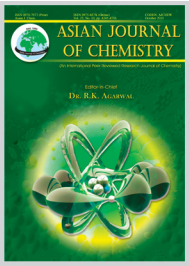

Comparison of Volatile Oxidation Products of Lard from Native Breed and Three-way Crossbred Breed Under Accelerated Oxidative Conditions

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The volatile composition of thermally oxidized lard from different sources was investigated. Volatile oxidation products extracted by headspace-solid phase micro-extraction were analyzed by gas chromatography-mass spectrometer. Fatty acid compositions in the lard of Enshi No.1 native breed (ENL) and three-way crossbred breed (TCL) were also determined. The contents of volatile oxidation products were higher in the ENL than in the TCL. Aldehydes were detected to be the most abundant volatile oxidation compounds in the two lard sources, representing from 72.37 to 73.77 %. Lard source had a significant ($P < 0.05$) influence on the major oxidation products of aldehydes, such as heptanal, octanal, nonanal, (E)-2-heptenal, (E)-2-octenal, (E, E)-2, 4-heptadienal, (E, E)-2, 4-decadienal, 2-undecenal and so on. Only slight differences in other lipid oxidation products were observed.

Key Words: Lipid oxidation, Lard, Breeds, Volatile compounds, SPME.

INTRODUCTION

Lipid oxidation plays an important role in the formation of the characteristic flavour of cooked meat and meat products[1]. There is no doubt that the meat species differences in flavour are largely explained by differences in lipid-derived volatile components. Lipid deterioration provides volatile compounds which give fatty aromas to cooked meat and determine some of the aroma differences between meats from different species[2]. In fact, the degree of oxidation in oil and fat has a direct relationship with quality criteria of food industry. Generally, unsaturated fatty acids are easier to be oxidized than saturated ones. In the presence of oxygen, unsaturated fatty acids undergo decomposition even at low temperatures[3]. Primary oxidation products, although tasteless and odorless, will act as precursors of desirable flavour compounds[4]. Actually, the most abundant and important flavour groups originate from secondary lipid oxidation products. Alcohols, aldehydes, ketones, acids, hydrocarbons and lactones are major compounds in further oxidation and decomposition of lipid[5]. At high concentrations, these compounds contribute undesirable flavour characteristics generally classified as rancid, sharp, cardboardy and pungent[6].

Many methods based on the concentration of primary or secondary products have been developed to detect the degree of lipid oxidation[4,7]. Peroxide value (POV), conjugated dienoic acid (CDA) and *p*-anisidine value (*p*-AV) are common measurements of lipid oxidation. However, because hydroperoxides exist only transiently and decompose rapidly into secondary products[8], the measurement of volatile compounds has become a well accepted indicator of lipid oxidation. In order to characterize volatile lipid oxidation products, solid phase micro-extraction-MS is the method of choice[9]. Ideally, SPME is a unique sample preparation technique, which eliminates most disadvantages to extracting organics, including high energy costs, solvent contamination, damage of the heat-sensitive oil products and excessive preparation time. It has been widely applied to many types of foods due to its simplicity and rapidness, such as fermented foods, cheese, fruit juices and edible oils including corn, soybean, olive, sunflower and rapeseed oils[10,11].

Also, this technique has proven to be an effective tool for detecting fat derived flavour compounds. A method based on headspace-solid phase micro-extraction coupled to GC-MS has been used for the analysis of volatile compounds formed from lipid oxidation of fish muscle[8]. Yasuhara and Shibamoto[12] studied the volatile compounds in the headspace of heated pork fat and found that the major components produced were hexanal, heptanal and pentanal. Boylston *et al.*[6] compared the volatile lipid oxidation products of Wagyu and domestic sources of beef. There were no significant differences between the volatile oxidation compounds when the beef was analyzed

immediately following cooking. However, after storing at 3 ºC for 3 days, levels of lipid-derived volatile compounds were higher in the Wagyu breeds than in other domestic beef sources. The effect of breed on the flavour development is debatable. Elmore *et al*.[13] studied the volatile and fatty acid compositions of grilled beef from animals fed either silage or concentrates. Fed diets had a significant effect on the contents of volatiles and fatty acid compositions, but only slight effects of breed were observed.

However, the effect of breed on flavour development is debatable and the application of SPME in volatile oxidation products analysis from different lard sources is rare in the current literature. This paper compared the volatile oxidation products and fatty acid compositions of different lard sources. Two lard sources, the Enshi native breed and the three-way crossbred breed were compared.

EXPERIMENTAL

Enshi No.1 native pig is a typical domestic breed in Hubei Province, naturally raised by domestic farmers and fed diets based on natural green fodder (*e.g.*, corn, potato, coarse cereals, *etc*.), spring on mountain rather than feed additive. The prime features of native pigs include high fecundity, strong stress resistance, slow growth, less lean meat percentage and better meat quality. The three-way crossbred pig is a representative breed, which has a high utilization of fodder, faster growth, high amount of lean meat and a great demand in the market. Fresh lard from the native breed in Enshi Autonomous Prefecture (ENL) and the three-way crossbred breed (TCL) were purchased from Zhongbai warehouse in Wuhan, Hubei province. They were placed in two brown glass jars (protected against light) flushed with N_2 gas respectively and stored frozen (-18 ºC) until needed for experiments.

Standards of *n*-alkanes (C_8-C_{20}) were purchased from Fluka Chemical Co. (Germany). The following 6 compounds were used as external standards and used for the method evaluation: 2-Pentyl-furan (99.7 %), (E)-2-decenal (95 %) were obtained from Tokyo Chemical Co. Ltd., (Japan). (E,E)-2,4-decadienal (95 %) and 1-octen-3-ol (98 %) were purchased from Aldrich-Chemie (Steinheim, Germany), while hexanal (98 %) and octanal (95 %) were obtained from Riedel-de Haen AG (Germany). Methyl esters of myristic acid, palmitic acid, palmitoleic acid, stearic acid, oleic acid and linoleic acid were purchased from Nu-Chek Prep Inc (Elysian, MN).

Oxidation procedure: Fresh lard was mildly oxidized by oxygen under controlled conditions, which could reach lower *p*-anisidine value and higher peroxide value. Fifty grams of lard were placed in a 500 mL three-neck round-bottom flask. The flask was connected to a water reflux condenser, a vent pipe and a sealed plug, respectively. The reaction vessel was immersed into an oil bath, flushed with a stable flow of 0.025 m^3/h of O_2, heated at 120 (± 2) ºC and stirred continuously on a magnetic stirrer. Heating continued for another 2 h after the required temperature was reached. Each experiment was performed in triplicate.

Analysis of fatty acid composition: Fatty acid composition was analyzed by GC-FID. Fatty acid methyl esters were prepared by using a modification of those described previously using KOH /methanol reagent[14]. Briefly, lard was mixed with 2 mL hexane and converted into fatty acid methyl esters (FAMEs) by using 0.4 M KOH in methanol. Derivatized samples were filtered through a 0.2 µm membrane filter (Whatman) and the obtained solutions were analyzed by GC-FID.

Fatty acid methyl esters were analyzed on a HP6890 gas chromatograph with a DB-5 column (30 m × 0.32 mm × 0.25 µm). The GC column was programmed from 100 ºC to 170 ºC at 10 ºC /min and then increased to 200 ºC at a rate of 5 ºC/min, held for 2 min and then raised to 220 ºC at 2 ºC /min (held for 1 min). Nitrogen was the carrier gas with a split ratio of 10:1. The injection volume was 1 µL. Fatty acids were identified by known external standards. The area percent of each fatty acid was calculated by dividing its peak area by the total peak area of the fatty acids identified.

Extraction of volatile compounds by SPME: Volatile compounds were extracted using a manual SPME device equipped with a 50/30 µm DVB/CAR/PDMS fiber (Supelco, Bellfonte, PA, USA). The SPME fiber was conditioned for 1 h in the GC injection port at 270 ºC prior to use as recommended by the manufacturer. After thawing, 5 mL lard samples were added to a 20 mL vial containing a micro stirring bar. The vial was capped with a PTFE septum and placed in a 80 ºC water bath on a magnetic stirrer and the sample was equilibrated for 15 min under stirring (400 rpm) at the required temperature before SPME sampling. The fiber was then inserted into the headspace for 0.5 h. Selection of extraction temperature, equilibrium time and extraction time for SPME-GC/MS was based on our previous experience. After extraction, desorption was done at 250 ºC in the GC injection port for 5 min. Each analytical sample was measured in triplicate. In order to evaluate the possibilities of the method for the linearity and recovery of the standard compounds under study, the experiments were carried out.

Analysis of volatile oxidation products: Volatile compounds were analyzed using an Agilent 6890 N GC with a split/splitless injector, coupled to a HP 5975B quadropole mass selective detector (Agilent Technologies, USA). The flow rate of helium on HP-5 column (30 m length, 0.25 mm I.D. and 0.25 µm film thickness, J & W Scientific, Folsom, CA, USA) was 1.2 mL min^{-1}. Mass spectra were obtained in electronic ionization (EI) mode at 70 eV in the 35- to 350-amu range. The ion source was set at 230 ºC and the quadrupole at 150 ºC. The injection port was in splitless mode and the injector temperature was 250 ºC. The chromatographic program was set at 40 ºC, raised to 120 ºC at 3 ºC /min, held for 2 min and then increased to 220 ºC at a rate of 5 ºC /min.

The identification of compounds was based on comparison of their mass spectra with the spectral data from MS libraries (NIST 05, WILEY 7.0). When available, volatile compounds were identified by GC retention times and mass spectral data of authentic standards. The identification was confirmed using retention indices and the value was compared with those reported in the literature. A mixture of *n*-alkanes (C_8-C_{20}) standards was run under the same conditions to calculate retention index (RI) value of each compound[15].

Quantification: For quantification purposes, selected volatile compounds' regression equations were made and their concentrations were obtained from these equations. Quantification of the monitored compounds was done using calibration

curves for known amounts of each volatile compound[3,4]. Refined soybean oil was used by spiking with pure external standards to avoid the use of solvents that might affect the SPME process. Six standards were accurately weighed and dissolved in freshly refined oil. The solution was then diluted repeatedly with the same oil to obtain the appropriate concertration. Five concentrations of each standard were prepared for calibration curves. The calibrating oils contained 0.5-5 ppm of 1-octen-3-ol, 0.2-2 ppm of 2-pentyl-furan, 2-20 ppm of hexanal and (E)-2-decenal, 10-100 ppm of octanal and (E,E)-2,4-decadienal. The standard solutions were analyzed in the same way as the lard samples. Triplicate analyses were performed on each experiment.

Statistic analysis: Data reported were obtained from triplicate samples and expressed as mean ± standard deviation. The statistical significance of the volatile oxidation compounds was evaluated by one-way ANOVA. The experimental data analyses were done with the SPSS 13.0 for Windows statistical package. Statistically, differences with P-values under 0.05 were considered significant.

RESULTS AND DISCUSSION

Fatty acid composition: Fatty acid profiles of fresh ENL and TCL are presented in Table-1. Oleic acid was the main fatty acid in the two lard sources, ranging from 36.33 to 46.34 %. Palmitic acid was the second most prevalent fatty acid (24.08-28.20 %), followed by stearic acid (13.60-17.69 %) and linoleic acid (13.13-14.77 %). The concentration of myristic acid was the lowest. Fatty acid analysis showed that difference between lard sources was evident in the oleic acid content. ENL had a 10 % higher concentration of oleic acid than TCL. Linoleic acid was the only polyunsaturated fatty acid detected. The concentration of linoleic acid in TCL (14.77 %) was slightly higher than that in ENL (13.13 %). In addition, the contents of palmitic acid and stearic acid in ENL were lower than that in TCL, representing 37.68 % and 45.89 % respectively.

TABLE-1
FATTY ACIDS PROFILE (% OF TOTAL FATTY ACIDS) OF FRESH LARD FROM ENSHI NO.1 NATIVE BREED AND THREE-WAY CROSSBRED BREED

Fatty acid composition	ENL (%)	TCL (%)	Significant
Myristic acid	0.97 ± 0.07 b	1.27 ± 0.08 a	*
Palmitic acid	24.08 ± 0.95 b	28.20 ± 1.06 a	*
Palmitoleic acid	1.89 ± 0.08 a	1.74 ± 0.10 b	*
Stearic acid	13.60 ± 0.62	17.69 ± 0.72	NS
Oleic acid	46.34 ± 1.19 a	36.33 ± 1.15 b	**
Linoleic acid	13.13 ± 0.58 b	14.77 ± 0.64 a	*

Data are expressed as means ± standard deviations (n = 3); NS, not significant; Values for each fatty acid with the different letter (a, b) are significantly different ($p < 0.05$) between ENL and TCL.

Generally, ENL contained a higher proportion of monounsaturated fatty acids (mainly oleic acid) and lower saturated fatty acids (mainly palmitic acid and stearic acid) compared with TCL, while the polyunsaturated fatty acid content was similar. Elmore *et al.*[13] studied the differences on the fatty acid compositions of grilled beef from two cattle breeds and found that the ratio of polyunsaturated fatty acids to saturated fatty acids was higher in the Holstein-Friesian cattle compared with the Aberdeen Angus cattle but no other effects of breed were observed. It has been reported that, many of the fatty acid derived volatiles are related to the relative amounts of oleic acid and linoleic acid[16]. Hence, it is necessary to compare the variation of volatile oxidation compounds in ENL and TCL, owing to the different composition of fatty acids in lard samples.

Linearity and recovery of volatiles: The 6 standard compounds found in oxidized lard were analyzed to determine the linearity and recovery of HS-SPME. Five concentrations of each analyte were prepared for plotting standard calibration curves. The volatile compounds selected with the ranges of linearity and performance parameters of calibration curves are summarized in Table-2.

The linear range covered the volatile compound concentrations expected in the samples since good regression coefficients (R^2) were obtained. As shown in Table-2, (E, E)-2,4-decadienal showed the best linearity ($R^2 = 0.998$). On the other hand, the least linearity was obtained for 2-pentyl-furan ($R^2 = 0.976$). Jelen *et al.*[3] investigated 14 volatile lipid oxidation compounds including hexanal, heptanal, 1-octen-3-ol, octanal and (E, E)-2, 4-decadienal extracted by HS-SPME and confirmed the availability of SPME method for each volatile analysis. Heptanal showed the best linearity ($R^2 = 0.998$) and pentanal obtained the least linearity ($R^2 = 0.918$).

Recoveries were performed to test the accuracy of the method. As shown in Table-2, the average recoveries of standard compounds ranged from 82.35 to 109.52 %. The results demonstrated that the method was applicable for the analysis of volatile compounds from thermally oxidized fatty acids.

Volatile compounds during oxidation: Gas chromatography-mass spectrometry data of volatile compounds isolated from oxidized lard using SPME are summarized in Table-4. The volatile oxidation compounds identified in the study were primarily aldehydes, aliphatic acids, alcohols, ketones and other volatiles. These compounds represented groups of characteristic fatty acid secondary oxidation products, resulting

TABLE-2
THE CONCENTRATION RANGE, REGRESSION EQUATIONS, R^2, RECOVERY FOR THE STANDARD COMPOUNDS

Compounds	Concentration range (μg/L)	Regression equation	R^2	Recovery (%)
Hexanal	1.63-16.30	y = 6803656 x + 4506067	0.986	82.35
1-Octen-3-ol	0.42-4.17	y = 15699581 x - 2136787	0.990	95.28
2-Pentyl-furan	0.18-1.77	y = 64458340 x + 9717866	0.976	109.52
Octanal	4.11-82.20	y = 1440842 x + 3559738	0.993	86.52
(E)-2-Decenal	1.69-16.92	y = 18859313 x - 2376673	0.991	92.65
(E,E)-2,4-Decadienal	8.72-87.20	y = 30835112 x - 4904490	0.998	96.46

Y the volatile compound peak area, X the volatile compound concentration

mostly from the oxidation of oleic acid, linoleic acid and linolenic acid[9]. Most of the identified compounds had been previously reported in the literature as volatile lipid oxidation components[17,18]. Among these volatiles, 6 compounds were identified using chemical standards (Table-3) and others were identified tentatively.

TABLE-3
QUANTIFICATION OF MAJOR VOLATILE COMPOUNDS BASED ON THE STANDARD CALIBRATION CURVE (μg/L)

Volatile oxidation compounds	Flushed with oxygen content (μg/L)	
	ENL	TCL
Hexanal	6.84 ± 0.67	7.18 ± 0.83
1-Octen-3-ol	1.95 ± 0.35	2.04 ± 0.33
2-Pentyl-furan	1.18 ± 0.16	1.20 ± 0.15
Octanal	73.95 ± 6.69	64.94 ± 7.05
(E)-2-Decenal	15.77 ± 1.17	14.64 ± 1.27
(E,E)-2,4-Decadienal	37.35 ± 3.34	42.55 ± 3.66

Results are the means ± standard deviation (n = 3).

As shown in Table-4, a total of 44 volatile compounds were identified in oxidized lard, including 13 aldehydes, 6 acids, 9 alcohols, 6 ketones, 6 hydrocarbons, 3 esters and 1 furan. In total, 35 and 38 volatile compounds were identified in oxidized ENL and TCL respectively and their total peak areas were 1848.64×10^6 and 1796.91×10^6. It has been reported that there are large amounts of aldehydes in the headspace of cooking oils. Aldehydes were the predominant volatiles of cooking oil[18]. As indicated in Table-4, higher aldehydes were the largest group of volatile compounds in oxidized lard based on peak areas, most of which were reported before as the volatile oxidation constituents of oxidized pork phospholipids[17]. These aldehydes, including 4 alkanals, 6 alkenals and 3 dienals, represented from 72.37 % (TCL) to 73.77 % (ENL) of the total volatile compounds. They have great importance as oxidation products because of their contribution to the aroma of oxidized oils and foods containing fat[19]. Among these aldehydes, (E)-2-decenal, 2-undecenal, (E)-2-octenal, nonanal, octanal, (E, E)-2, 4-decadienal and (E)-2-heptenal were the predominant in oxidized lard. Um *et al.*[20] identified that (E)-2-nonenal, (E)-2-decenal and 2-undecenal were present in high concentrations in the heated beef fat. Moreover, these aldehydes have been found to contribute oily and tallowy flavour to beef.

Significant differences on the content of volatile oxidation products as influenced by lard source were noted (Table-4). Drumm and Spanier[21] demonstrated that the rate of formation of lipid oxidation products was dependent on the specific compounds, which were formed at a faster rate than the other oxidation products. The significant differences between two lard sources were dependent upon the specific 13 volatile compounds, including 11 aldehydes and 2 acids. However, there were no significant differences for the remaining volatile oxidation products between two lard sources. Based on peak areas, heptanal, (E)-2-heptenal, octanal, (E, E)-2, 4-heptadienal, (E)-2-octenal, nonanal, (E)-2-nonenal, (E, E)-2, 4-nonadienal, (E)-2-decenal, (E, E)-2, 4-decadienal and 2-undecenal were the main volatile components responsible for differences between two oxidized lard samples. Hexanal was one of the main volatile oxidation products, accounting for 6.84 and 7.18 μg/L (Table-3) in ENL and TCL, respectively. Hexanal is a typical oxidation volatile from linoleic acid, described as the source of fatty aromas by Stanke[22], which has been commonly monitored for measuring lipid oxidation in foods. (E, E)-2, 4-Decadienal arises from decomposition of linoleic acid hydroperoxides[16] and (E)-2-heptenal is also a typical volatile oxidation product from linoleic acid[7]. The content of unsaturated fatty acid plays an important role in the generation of lipid-derived flavour compounds. Torres *et al.*[16] reported that nonanal, (E)-2-decenal and 2-undecenal had significant positive correlations with oleic acid content, but hexanal and (E, E)-2, 4-decadienal were positively correlated with linoleic acid. Heptanal, octanal and nonanal originate mainly from oleic acid[23]. These alkanals are very important, because they contribute to a pleasant fruity aroma at low concentration; instead, they give unpleasant, sharp and pungent attributes if present at higher concentration[24].

Specifically, the ENL had significantly higher ($P < 0.05$) contents of heptanal, octanal, nonanal, (E, E)-2, 4-heptadienal, (E)-2-nonenal, (E, E)-2, 4-nonadienal, (E)-2-decenal and 2-undecenal than the TCL, but lower ($P < 0.05$) contents of (E)-2-heptenal, (E)-2-octenal and (E, E)-2, 4-decadienal. This result coincided with the fatty acid composition of lard. It has been reported that the productions of volatile oxidation compounds are greatly influenced by the fatty acid composition of oils[3]. The concentration of oleic acid in ENL was 10 % higher than that in TCL, while linoleic acid was 1.64 % lower. The higher content of oleic acid in ENL could give rise to increased levels of compounds formed in decomposition of oleic acid. Differences in the contents of volatile oxidation products were attributed to different fatty acid composition in the lard samples.

Representing 12.65 % (ENL) to 13.69 % (TCL) of the total compounds identified, 6 acids were identified in the oxidized lard, which included hexanoic acid, heptanoic acid, 2-heptenoic acid, octanoic acid, 2-octenoic acid and nonanoic acid. Aliphatic acids appear to be the ubiquitous volatiles and derived from the decomposition of primary oxidation products. Heptanoic acid has been identified as a minor volatile compound in oxidized methyl linoleate. 2-Octenoic acid could be derived directly from (E, E)-2, 4-decadienal[25]. In general, the total fatty acids levels in TCL were significantly higher than ENL. Among these acids, only hexanoic and nonanoic acid were significantly different in the two lard sources. The ENL had significantly higher ($P < 0.05$) content of nonanoic acid, but lower ($P < 0.05$) content of hexanoic acid. It seems probable that the volatile unsaturated fatty acids were derived from oxidation of corresponding aldehydes which were derived initially from long-chain unsaturated fatty acids. The higher ($P < 0.05$) content of nonanal in the ENL could result to increased level of nonanoic acid and lower content of hexanal gave rise to decreased level of hexanoic acid. The presence of fatty acids is generally related to the flavour of oils and food products containing fat. Ha and Lindsay[25] postulated that many volatile fatty acids present in beef fat contributed to tallow-like flavour in deep-fried potatoes.

Alcohols, ketones and other compounds (including hydrocarbons, esters and furans) represented together 13.42 % and 14.04 % of the total volatile compounds in ENL and TCL respectively. However, the contents of these compounds were

TABLE-4
VOLATILE COMPOUNDS OF LARD OXIDIZED BY OXYGEN AS INFLUENCED BY LARD SOURCE

Volatile oxidation compounds	RI	Total peak area ($\times 10^{-6}$)		Significant
		ENL	TCL	
Aldehydes				
Hexanal	800	51.01 ± 4.60	53.34 ± 5.70	NS
(E)-2-Hexenal	852	n.d.	0.42 ± 0.10	NS
Heptanal	902	56.10 ± 6.36 a	44.31 ± 6.73 b	*
(E)-2-Heptenal	956	61.74 ± 7.79 b	82.49 ± 9.11 a	*
Octanal	1004	110.11 ± 9.63 a	97.12 ± 10.20 b	*
(E,E)-2,4-Heptadienal	1013	32.08 ± 4.64 a	26.60 ± 4.55 b	**
(E)-2-Octenal	1059	166.47 ± 15.49 b	178.02 ± 16.04 a	*
Nonanal	1106	114.25 ± 11.85 a	102.76 ± 11.46 b	*
(E)-2-Nonenal	1161	76.56 ± 9.89 a	69.24 ± 10.34 b	*
(E,E)-2,4-Nonadienal	1218	40.34 ± 6.01 a	34.22 ± 5.56 b	*
(E)-2-Decenal	1264	294.93 ± 22.10 a	273.57 ± 23.87 b	*
(E,E)-2,4-Decadienal	1319	110.25 ± 10.32 b	125.28 ± 11.27 a	*
2-Undecenal	1368	249.88 ± 19.16 a	213.06 ± 20.35 b	*
Sub total		1363.72 a	1300.43 b	*
Acids				
Hexanoic acid	1020	116.70 ± 11.03 b	155.70 ± 15.39 a	*
Heptanoic acid	1112	32.79 ± 4.75	27.08 ± 4.06	NS
2-Heptenoic acid	1155	2.45 ± 0.57	4.21 ± 0.86	NS
Octanoic Acid	1211	18.53 ± 3.04	12.93 ± 2.15	NS
2-Octenoic acid	1244	0.86 ± 0.21	n.d.	NS
Nonanoic acid	1301	65.58 ± 7.88 a	44.35 ± 5.66 b	*
Sub total		236.91 b	244.27 a	*
Alcohols				
3,7-Dimethyl-1-octanol	979	2.21 ± 0.51	4.58 ± 0.93	NS
1-Octen-3-ol	982	28.39 ± 5.47	29.91 ± 5.08	NS
1-Octanol	1093	59.01 ± 9.41	62.27 ± 8.27	NS
Phytol	1276	2.68 ± 0.64	n.d.	NS
2-Tetradecanol	1311	0.41 ± 0.10	3.81 ± 0.86	NS
1-Heptadecanol	1444	n.d.	0.20 ± 0.057	NS
2-Pentadecanol	1447	0.64 ± 0.18	0.013 ± 0.004	NS
2-Dodecanol	1490	n.d.	0.088 ± 0.024	NS
2-Heptadecanol	1548	0.66 ± 0.16	0.054 ± 0.012	NS
Sub total		94.00	100.93	NS
Ketones				
1-Hepten-3-one	980	4.38 ± 0.76	n.d.	NS
trans-3-Nonen-2-one	1141	7.72 ± 1.22	10.17 ± 1.63	NS
1-Hexen-3-one	1182	n.d.	2.04 ± 0.44	NS
2-Decanone	1194	9.92 ± 1.57 a	9.49 ± 1.54 b	NS
2-Tridecanone	1505	1.85 ± 0.47	n.d.	NS
2-Pentadecanone	1685	n.d.	2.78 ± 0.67	NS
Sub total		23.87	24.48	NS
Others				
5-Methyl-2-heptene	827	5.12 ± 1.0	n.d.	NS
1-Nonene	891	9.29 ± 1.78	1.59 ± 0.34	NS
2-Pentyl-furan	991	85.33 ± 10.50	86.89 ± 9.43	NS
Pentyl hexanoate	1288	2.68 ± 0.64	4.62 ± 0.96	NS
γ-Nonalactone	1374	18.73 ± 2.97	18.45 ± 3.39	NS
Dodecyl-oxirane	1410	0.18 ± 0.042	5.37 ± 1.00	NS
Pentadecane	1497	3.95 ± 0.86	5.53 ± 1.06	NS
Octyl hexanoate	1544	n.d.	2.06 ± 0.45	NS
Tetradecy-oxirane	1562	4.86 ± 0.95	0.40 ± 0.11	NS
Hexadecane	1598	n.d.	1.89 ± 0.48	NS
Sub total		130.14	126.80	NS
Total		1848.64 a	1796.91 b	*

RI, Linear retention index on HP-5MS column; n.d., not detected; Results are the means ± standard deviation; *indicates significant difference ($p < 0.05$) between ENL and TCL; NS, not significant; Means with the different superscript (a, b) are significantly different ($p < 0.05$).

not significantly different between two lard sources. Alcohols were produced and formed as an important fraction of volatiles in the oxidation process. Among the detected alcohols, 1-octanol and 1-octen-3-ol were the most abundant. 1-Octen-3-ol generated from the oxidation of linoleic acid, accounting for 1.95 and 2.05 μg/L in ENL and TCL, respectively. 1-Octen-3-ol has a characteristic odor of mushroom and is an important contributor to off-flavour due to its low odor threshold, which was reported before as the volatile lipid oxidation product[8,23]. 2-Pentyl furan was the only furan compound identified in the two lard sources, the concentration of it was 1.18 μg/L in ENL and 1.20 μg/L in TCL. 2-Pentyl furan is a typical oxidation compound from linoleic acid. γ-Nonalactone was the only lactones detected in the present study, representing 1.01 % (ENL) and 1.03 % (TCL) of the total volatile compounds identified. Lactones are key compounds that contribute to desirable aroma. Um *et al*.[20] reported that γ-nonalactone was present in high concentrations in the heated beef fat. A series of lactones generate during the thermal oxidation of saturated and unsaturated fatty acids.

Conclusion

The volatile compounds from thermally oxidized fatty acids were monitored by HS-SPME and the effect of lard source on the content and composition of volatile oxidation products was determined. In conclusion, the HS-SPME coupled with GC-MS is a good technique for analyzing the volatile oxidation compounds formed in the lard. The most abundant volatiles detected in oxidized ENL and TML were short- and medium-chain aldehydes. Generally, the results of the study showed that the ENL source had higher total peak areas of volatile oxidation products, in comparison to the TCL source. The significant differences in the content of oxidation volatiles were mainly dependent on the specific volatiles. A majority of aldehydes generated from these fatty acids were significantly different in the two lard sources. Only slight differences in other oxidation products were observed.

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