Investigations into the Effect of Supercritical Carbon Dioxide Extraction on the Fatty Acid and Volatile Profiles of Cooked Chicken

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Chicken fat was cooked to 80 °C and extracted using supercritical carbon dioxide at 40 °C and each of three pressures: 10.3, 20.7, and 31.0 MPa. Fatty acid and volatile profiles were obtained using capillary gas chromatography and GC–MS. Twenty-seven fatty acids were identified (8 saturates, 7 monounsaturates, and 12 polyunsaturates). Extraction pressure affected the relative concentration of 2 fatty acid classes and 13 individual fatty acids, all of which were 18 or more carbons in length. Monounsaturated and polyunsaturated fatty acid concentrations increased among extracts with increasing extraction pressure. Quantitated volatiles numbered 318, of which 99 were classified. The largest classes of volatiles were branched alkanes, enals, and aldehydes. The concentrations of total volatiles, branched alkanes, enals, ketones, lactones, alcohols, and halogenated and aromatic compounds were affected by treatments. All extractions yielded higher volatile concentrations than control. Among extracts, volatile concentration increased with decreasing extraction pressure.

Keywords: *SC-CO_2; fatty acids; volatiles; chicken fat*

INTRODUCTION

The preponderance of excess lipid byproducts that result from commercial slaughter operations encourages the investigation of novel techniques to obtain value-added products from these commodities. Preparation of lipid fractions with tailored fatty acid and/or volatile profiles and their inherent functionality and property differences offer numerous possibilities for use in the food industry. Supercritical fluid extraction is one technique that affords the possibility of selective fractionation.

The use of supercritical fluid extraction offers the advantages of elimination of organic solvents, less destruction of thermally labile constituents as compared to steam distillation, lowered extraction times, and easily manipulated conditions that allow for unique separations. A supercritical fluid, above its critical temperature and pressure, has solvation properties that are pressure and temperature dependent (Calame and Steiner, 1982). Such systems possess properties intermediate between those of liquids and gases. Mass transfer properties resemble those of gases, while solvation properties more closely resemble liquids; high diffusivity and low surface tension facilitate penetration of solid-like food matrices and extraction of components of interest (Brogle, 1982; Caragay, 1981; de Filippi, 1982; Hardardottir and Kinsella, 1988). Carbon dioxide (CO_2) is a popular supercritical fluid due to its similarity to organic solvents (Hyatt, 1984) and relatively low critical temperature (31.1 °C) and pressure (7.38 MPa) along with the advantages of being inexpensive, nontoxic, inert, and nonflammable. The solubility of components in supercritical CO_2 (SC-CO_2) is dependent on temperature and pressure. At a given temperature, the solvent power of a supercritical fluid increases with density as a function of pressure (Brogle, 1982), while the density of the fluid increases with decreasing temperature at constant pressure.

Rizvi *et al.* (1986) reported current commercial applications of the process in the decaffeination of coffee beans and the recovery of hops extract as well as in the recovery of spice essences. Patented supercritical fluid processes include removal of oil from oilseeds, deodorization of fats and oils, and fractionation of certain fats and oils. In animal products, supercritical fluid extraction has been used to remove cholesterol and lipids from milk fat, egg yolk, and muscle tissues (Bradley, 1989; Chao *et al.*, 1991; Froning *et al.*, 1990; Hardardottir and Kinsella, 1988; King *et al.*, 1989, 1993).

Successful fractionation of lipids in milk fat using SC-CO_2 has been demonstrated, indicating the ability to achieve molecular weight separation (Arul *et al.*, 1987). Short-chain triglycerides and fatty acids decreased in concentration among extracted fractions as the solidity of fractions increased, while long-chain triglycerides and fatty acids increased in concentration. Separations were partially attributed to differences in the molecular weight and volatility of the triglyceride components. Bhaskar *et al.* (1993) utilized continuous, countercurrent SC-CO_2 extraction to fractionate anhydrous milk fat, showing an increase in concentration of short- and medium-chain fatty acids with decreasing separation pressure while long-chain and unsaturated fatty acid concentration followed an opposite trend.

Chao *et al.* (1991) removed fat and cholesterol from ground beef with SC-CO_2 and observed fractionation into low- and mid-melting components. Merkle and Larick (1993) fractionated raw beef fat and selectively extracted saturated and monounsaturated triglycerides on the basis of molecular weight and solvent density of the SC-CO_2. Polarity-based extraction and fractionation

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were demonstrated by Merkle and Larick (1994a) by the increased concentrations of mono- and polyunsaturated fatty acids, in extracts collected from beef tallow, with increasing solvent density. Maheshwari *et al.* (1992) suggested the possibility of separating fatty acids from triglycerides at SC-CO_2 densities below 700 kg/m^3 via comparison of their solubility isotherms across densities and noted that separation of fatty acids from one another is possible but likely not for similar species such as octadecenoic (18:1) and octadecadienoic acids (18:2) and tetradecanoic (14:0) and hexadecanoic (16:0) acids when temperatures near 60 °C are used.

Volatile compounds have been extracted and concentrated from milk fat with supercritical CO_2, indicating the importance of vapor pressure differences between volatile flavor components and nonvolatile triglycerides in selectivity. Among volatile compounds, concentration in the extract decreased with increasing molecular weight due to the vapor pressure effect (de Haan and de Graauw, 1990). Merkle and Larick (1994b) also demonstrated the ability to concentrate beef flavor volatiles with supercritical fluid extraction.

The purpose for this research was to examine the fractionating ability of SC-CO_2 extraction on cooked chicken lipids on the basis of the obtained fatty acid and bulk volatile profiles.

MATERIALS AND METHODS

Sample Preparation. Chicken fat was collected from a commercial processor and ground once through a 0.95 cm plate. Portions (500 g) were weighed and vacuum packaged in low-permeability bags (Cryovac Corp., Duncan, SC) and overwrapped with polyethylene-coated freezer paper. Samples were stored at −20 °C and thawed at 2−4 °C prior to use. Prior to extraction, fat was heated in a convection oven to 80 °C in 500 mL Pyrex glass beakers to melt the triglyceride portion and create a substrate mimetic of roasted chicken fat. An unextracted portion was placed in a Pyrex tube (16 mm × 125 mm), flushed with nitrogen gas, sealed with a Teflon-lined cap, frozen at −10 °C, and retained as a control.

Extraction. A Superpressure Model 46-13421-2 supercritical fluid extractor (Newport Scientific, Jessup, MD) equipped with a 69.0 MPa double-end diaphragm compressor was used to fractionate the cooked chicken fat (Merkle and Larick, 1994b). Aliquots (250 g) were immobilized between two plugs of glass wool and loaded into a 0.845 L (internal volume) stainless steel extraction vessel. Vessel temperature was maintained at 40 °C via an internal thermocouple and temperature controller with heat tape wrapped around the outside of the vessel. Stainless steel transfer lines were wrapped with insulation to maintain temperature within the system. Continuous extractions were carried out at each of three pressures, 10.3, 20.7, and 31.0 MPa, using SC-CO_2 at a flow rate of 10−15 L/min to a total flow volume of 500 L of ambient CO_2 measured using a flow totalizer. Extracts were collected in a 0.500 L Pyrex round-bottom flask upon fluid depressurization at ambient temperature. Extracts were transferred to Pyrex tubes (16 mm × 125 mm), flushed with nitrogen gas, sealed with a Teflon-lined cap, and frozen at −10 °C until analyzed.

Fatty Acid Quantitation. Extracts and an unextracted control were melted in a 60 °C water bath. Fatty acid methyl esters (FAME) were prepared for each treatment using the method of Morrison and Smith (1964) with modifications. Samples (100 mg) were placed into a 16 mm × 125 mm Pyrex tube and diluted with 10 mL of chloroform/methanol in a 2:1 ratio with 0.01% BHT added as an antioxidant. Aliquots (1.0 mL) were transferred to another 16 mm × 125 mm Pyrex tube, and boron trifluoride (12%) in methanol was added in excess (≥2 mL) for methylation. Internal standard [798.58 μg of heptadecenoic acid (17:1) in 40 μL of capillary GC/GC−MS grade hexane (Burdick & Jackson, Muskegon, MI)] was also added. Tubes were shaken to mix. Tubes were heated for 30 min in a 100 °C heating block. Saturated sodium chloride solution (4 mL) was added to tubes. Optima grade hexane (Fisher Scientific, Fair Lawn, NJ) was added (2 mL) to each tube to extract esters. Tubes were shaken for 5 min to facilitate extraction. The top solvent layer from each tube was transferred via Pasteur pipet to a 5 mL conical Pyrex vial. An additional 2 mL of hexane was added to sample tubes, and the tubes were then shaken for 5 min. The top solvent layer was removed as above and added to the original extract in the Pyrex vial. Samples were evaporated to 1 mL under a gentle stream of nitrogen gas. Sodium sulfate was added to the bottom of the vials to bind residual water. A 0.5 μL sample was injected directly onto a 30 m DB-23 fused silica capillary column with cyanopropyl substitution (J&W Scientific, Folsom, CA) with an internal diameter of 0.25 mm and a film thickness of 0.25 μm. An oven temperature program of 150−215 °C at 4 °C/min and 215−225 °C at 2 °C/min was used. Esters were analyzed using a Hewlett-Packard 5890 gas chromatograph equipped with a flame ionization detector and maintained with a head pressure of 26 psi, a helium carrier gas flow rate of 1.36 mL/min, and a split ratio of 26.7:1. Injector and detector temperatures were maintained at 230 and 240 °C, respectively. Data were analyzed using the Maxima 820 Chromatography Workstation (Millipore, Waters Chromatography Division, Milford, MA). Quantitation was achieved via comparison of peak areas to that of the internal standard.

In separate runs, 0.5 μL injections of prepared ester solutions were injected, and response factors were calculated from triplicate runs by comparing peak areas of known quantities of ester reference standards (Nu Check Prep, Inc., Elysian, MN) to the peak area of the internal standard per the following formula:

$$\text{response factor} = [(\text{concentration of ester standard}) \times (\text{peak area of internal standard})]/[(\text{peak area of ester standard}) \times (\text{concentration of internal standard})]$$

Response factors (Table 1) were used to adjust preliminary quantitations for instrument response.

Table 1. Response Factors for Identified Fatty Acids in Supercritical Carbon Dioxide Extracts of Chicken Fat

	response factor[a]		
fatty acid[b]	saturates	mono-unsaturates	poly-unsaturates
12:0	0.223		
14:0	0.557		
15:0	0.767		
16:0	0.922		
17:0	1.017		
18:0	1.128		
20:0	1.281		
22:0	1.226		
14:1 *c*9		0.573	
16:1 *c*9		0.923	
18:1 *t*9		1.099	
18:1 *c*9		1.090	
18:1 *c*11		1.062	
20:1 *c*11		1.284	
18:2 *t*9,12			1.043
18:2 *c*9,12			1.081
18:3 *c*6,9,12			1.080
18:3 *c*9,12,15			1.106
20:2 *c*11,14			1.222
20:3 *c*8,11,14			1.610
20:4 *c*5,8,11,14			1.142
22:4 *c*7,10,13,16			1.235
22:5 *c*7,10,13,16,19			1.225
22:6 *c*4,7,10,13,16,19			1.297

[a] Response factor = [(peak area of internal standard) × (known concentration of ester)]/[(known concentration of internal standard) × (peak area of ester)]. [b] Fatty acids identified by number of carbons followed by units of unsaturation, configuration, and placement.

Fatty Acid Identification. GC–MS analysis was carried out on fatty acid methyl esters prepared as noted above from control and 10.3 MPa extracts. A 1.0 μL sample was injected directly onto a 30 m DB-23 capillary column (J&W Scientific) with an internal diameter of 0.25 mm and a film thickness of 0.25 μm. An oven temperature program of 150–215 °C at 4 °C/min and 215–225 °C at 2 °C/min was used. Methyl esters were analyzed using a Hewlett-Packard 5985 gas chromatograph–mass spectrometer with an ionization potential of 70 eV using both electrical and chemical ionization. The scan range for electrical ionization was 40–500 atomic mass units (amu) and 80–500 amu for chemical ionization with methane gas in which an ionization potential of 230 eV was used. GC–MS information data were compared to reference spectra (*NIH/EPA Chemical Information System*, 1978) and was coupled with retention time information for injected pure reference standards (Nu Check Prep) for identification of fatty acids.

Volatile Quantitation and Identification. Extracts and unextracted control were melted in a 60 °C water bath. Samples (300 mg) were placed into another tube, and 1026 ng of internal standard, 2,3,4-trimethylpentane, in GC/GC–MS hexane was added. The tube was sealed and vortexed. Aliquots (100 mg) were placed between two plugs of pesticide grade (Supelco, Inc., Bellefonte, PA) glass wool in a 9 mm × 85 mm glass tube. With the six-port sample valve in the no-flow position, the tube was positioned in an external closed inlet device (Scientific Instrument Service, River Ridge, LA). Volatiles were stripped from the sample via this method of dynamic purge and trap sampling for a total of 5 min. Inlet, valve, and carrier lines were maintained at 150, 160, and 170 °C, respectively. Volatiles were flushed onto a nonpolar 30 m DB-5 fused silica capillary column (J&W Scientific) with an internal diameter of 0.32 mm and a film thickness 1.0 μm. The column was in a Varian 3700 gas chromatograph (Varian, Palo Alto, CA) equipped with a flame ionization detector and maintained with a head pressure of 16 psi, a helium carrier gas flow rate of 5.73 mL/min, and a split ratio of 20.12:1. An oven temperature program of −30 to 290 °C at 4 °C/min was used with a 1 min hold at −30 °C. Data were analyzed via the same software as for fatty acid analyses. Quantitation of volatiles was based on relative peak areas compared to peak area of the internal standard.

For identification via electrical ionization, 10.3 and 20.7 MPa extract samples (200 or 550 mg plus 1 mL of deionized water) were placed into a sampling tube of a Tekmar LSC-3 headspace concentrator. Samples were purged at 110 °C for 14–20 min followed by 5 min of desorption onto a 30 m DB-5 fused silica capillary column with an internal diameter of 0.32 mm and a film thickness of 1.0 μm within a Hewlett-Packard 5985 gas chromatograph–mass spectrometer. The oven temperature program was identical to that used in volatile quantitation. The ionization potential was 70 eV, and the scan range was 40–300 amu. For chemical ionization with methane gas, samples (500 mg) were purged for 14 min, desorbed for 5 min, and scanned over a 40–300 amu range with the same equipment setup and an ionization potential of 230 eV.

For more efficient concentration of higher molecular weight compounds for identification, a modified liquid/liquid extraction was used (Likens and Nickerson, 1964). Cooked fat (100 g) and deionized water (150 mL) were placed into a 500 mL Pyrex round-bottom flask with boiling chips and hooked to the long arm of a water-jacketed codistillation apparatus. The short arm was fitted to a 100 mL Pyrex round-bottom flask containing boiling chips and 75 mL of Optima grade methylene chloride (Fisher Scientific). Heating mantles beneath each flask were used to heat samples to boiling. Once both sides had begun condensing at the top of the still, codistillation was allowed for 6 h. After codistillation and cooling, remaining solvent phase containing entrapped volatiles was drained from the distillation apparatus and added to the 100 mL Pyrex round-bottom flask. A Buchi rotary evaporator was used to concentrate the solvent fraction to approximately 1 mL. The flask was washed three times with 1 mL of methylene chloride, and these volumes were added to the original 1 mL of concentrate. Combined sample was concentrated under a gentle stream of nitrogen gas to 0.1 mL prior to GC–MS analysis.

Samples (1–2 μL) from liquid/liquid extractions were injected directly onto a 30 m DB-5 capillary column in a Hewlett-Packard 5985 gas chromatograph–mass spectrometer under conditions as noted above and scanned over a 40–500 amu range. Similar conditions were used for chemical ionization with methane gas.

Volatile identifications were based on instrument retention time for previously identified compounds and GC–MS with both electrical and chemical ionization methods and comparison to NIH/EPA reference spectra (*NIH/EPA Chemical Information System*, 1978).

Experimental Design and Statistical Analysis. Extractions and fatty acid and volatile profiles were replicated three times on different batches of fat for the four treatments, three extracts and the unextracted control. Fatty acid and volatile concentrations were analyzed via analysis of variance using a randomized complete block design with replicates as blocks. Waller-Duncan *k*-ratio *t*-tests were used to separate means (*SAS/STAT User's Guide*, 1990).

Table 2. Classification of Fatty Acids Identified in Supercritical Carbon Dioxide Extracts of Chicken Fat

fatty acid	peak no.
saturates	
dodecanoic acid (12:0)[a]	1
tetradecanoic acid (14:0)	3
pentadecanoic acid (15:0)	9
hexadecanoic acid (16:0)	13
heptadecanoic acid (17:0)	20
octadecanoic acid (18:0)	25
eicosanoic acid (20:0)	40
docosanoic acid (22:0)	48
monounsaturates	
tetradecenoic acid (14:1 *c*9)	6
pentadecenoic acid (15:1)[b]	11
hexadecenoic acid (16:1 *c*9)	15
octadecenoic acid (18:1 *t*9)[b]	26
octadecenoic acid (18:1 *c*9)	27
octadecenoic acid (18:1 *c*11)[b]	28
eicosenoic acid (20:1 *c*11)	43
polyunsaturates	
hexadecadienoic acid (16:2)[b]	18
octadecadienoic acid (18:2 *t*9,12)[b]	31
octadecadienoic acid (18:2 *c*9,12)	33
octadecadienoic acid (18:2 9,11)[b]	41
octadecatrienoic acid (18:3 *c*6,9,12)	35
octadecatrienoic acid (18:3 *c*9,12,15)	37
eicosadienoic acid (20:2 *c*11,14)	45
eicosatrienoic acid (20:3 *c*8,11,14)	46
eicosatetraenoic acid (20:4 *c*5,8,11,14)	47
docosatetraenoic acid (22:4 *c*7,10,13,16)[b]	49
docosapentaenoic acid (22:5 *c*7,10,13,16,19)[b]	50
docosahexaenoic acid (22:6 *c*4,7,10,13,16,19)[b]	51

[a] Fatty acids identified by number of carbons followed by units of unsaturation, configuration, and placement (if specified), and GC peak number. [b] Tentative identification based on only one method.

RESULTS AND DISCUSSION

Fatty Acid Quantitation and Identification. Fifty-one compounds were quantitated with 27 fatty acids (9 tentative) and 2 phenolic compounds (tentative) identified. Eight fatty acids were saturated, ranging from 12 to 22 carbons in chain length; 7 were monounsaturated, ranging from 14 to 20 carbons; and 12 were polyunsaturated, ranging from 16 to 22 carbons (Table 2). Katz *et al.* (1966) and Dawson *et al.* (1990) identified all of these fatty acids (excluding isomeric designations within a series) save pentadecenoic acid (15:1) and docosanoic acid (22:0) in neutral lipid fractions from skin and depot fat and mechanically deboned chicken meat, respec-

tively. Dawson *et al.* (1990) noted docosanoic acid (22:0) in phospholipid fractions only. Pentadecenoic acid (15:1) has been noted by Shams El-Din and Ibrahim (1990) in lipid extracts from chicken breast muscle but not in the corresponding thigh muscle extracts. Long-chain $n-3$ polyunsaturates, which may originate from marine oils present in poultry diets (Phetteplace and Watkins, 1990), are known to affect lipid profiles in edible tissues. Poultry, which are nonruminant species, were shown to have a propensity for directly incorporating dietary fatty acids into the carcass lipids (Jen *et al.*, 1971; Schuler and Essary, 1971; Watkins, 1991). Two phenols, 2,6-di-*tert*-butyl-4-methylphenol (BHT) and 2-*tert*-butyl-6-methylphenol, were also tentatively identified.

Fatty acids found in the greatest concentration in all treatments were octadecenoic (C18:1 *c*9), hexadecanoic (C16:0), octadecadienoic (C18:2 *c*9,12), hexadecenoic (C16:1 *c*9), and octadecanoic (C18:0) (Table 3). The predominance by concentration of octadecenoic and hexadecanoic acid is common for poultry products (Jen *et al.*, 1971). By class, the monounsaturated fatty acids were predominant in concentration in all treatments followed by saturated and polyunsaturated fatty acids (Figure 1).

As a class, monounsaturates and polyunsaturates differed significantly by treatment. Among treatments, total fatty acid concentration was lowest for the 10.3 MPa extract. This trend results from the lower density and solvent strength of the supercritical fluid at lower pressure, which inhibits extraction capabilities. The unextracted control yielded the highest concentration of monounsaturated fatty acids, and concentration decreased among extracts as extraction pressure decreased (Figure 1). The enhanced polarity of monounsaturated and polyunsaturated fatty acids and, thus, the triacylglycerides containing them, required greater solvent strength for extraction. Because solvent strength decreases as a function of density with decreasing extraction pressure at constant temperature, the concentration of these classes of fatty acids in the extracts decreased with decreasing extraction pressure among extracts. Similar results were achieved by Merkle and Larick (1994a) and Bhaskar *et al.* (1993). Ratios of saturated to unsaturated (S/U), saturated to monounsaturated (S/M), and saturated to polyunsaturated (S/P) fatty acids (Table 4) indicate the inability of any of the extraction pressure treatments to extract unsaturated fatty acids on a level that equals their natural abundance in the unextracted control. Among extracts, S/U, S/M, and S/P ratios increase with decreasing extraction pressure, showing the greater ability of higher pressure treatments to extract triacylglycerides containing the more polar unsaturated fatty acids (Table 4).

Extraction pressure influenced the concentration of 6 unknown compounds and 13 individual identified fatty acids, of which 2 were saturated, 3 monounsaturated, and 8 polyunsaturated. Most individual saturates were sufficiently nonpolar and volatile to be quite soluble in SC-CO_2, even at 10.3 MPa. The only saturates affected by treatment were octadecanoic (18:0) and docosanoic (22:0) acids, which are of relatively high molecular weight and low volatility. The increased polarity of individual mono- and polyunsaturates allowed for more noticeable effects of pressure and solvating ability.

No differences existed between treatments for concentrations of individual identified fatty acids of chain length less than 18 carbons, indicating that the supercritical fluid possessed sufficient solvent strength at all pressures evaluated to extract the triacylglycerides containing shorter chain, lower molecular weight acids. The threshold for noticeable differences in extraction capabilities for this system may be at 18 carbons as 8 of 9 identified variants of an 18-carbon acid were affected by treatments. Only octadecenoic acid (18:1 *t*9) was unaffected, likely because it was present in all treatments at the lowest concentration among monounsaturates with 18 carbons (8.84–10.83 mg/g of extract)

Table 3. Fatty Acid Concentrations of Supercritical Carbon Dioxide Extracts of Chicken Fat

peak no.[a]	retention time	concentration[b] (mg/g) at 0.0 MPa	10.3 MPa	20.7 MPa	31.0 MPa
1	3.24	0.458	1.056	0.852	0.774
2	3.81	43.815	57.518	49.819	47.270
3	4.80	9.287	16.288	15.607	12.610
4	4.94	0.379	0.819	0.622	0.496
5	5.08	0.320	0.504	0.600	0.347
6	5.23	2.464	4.373	4.266	3.509
7	5.41	0.759	0.970	0.647	0.692
8	5.79	0.000	0.096	0.000	0.000
9	5.83	0.718	0.917	0.879	0.840
10	6.04	0.000	0.191	0.000	0.000
11	6.44	0.245	0.287	0.264	0.257
12	6.65	0.000	0.302	0.000	0.185
13	7.09	219.994	213.580	240.548	229.847
14	7.36	3.598	4.032	4.470	4.384
15	7.48	55.941	60.505	66.642	63.213
16	7.63	0.657	0.648	0.749	0.766
17	7.75	0.411[ab]	0.375[b]	0.477[a]	0.464[a]
18	8.01	1.801	1.788	2.144	2.074
19	8.23	0.217	0.271	0.274	0.310
20	8.39	1.474	1.176	1.388	1.433
21	8.67	0.430	0.572	0.455	0.370
22	8.77	0.212	0.381	0.384	0.194
23 (IS)	8.88	80.012	81.074	84.668	80.205
24	9.48	0.224	0.243	0.282	0.289
25	9.84	58.779[a]	39.320[c]	50.686[b]	51.896[b]
26	10.11	10.782	9.012	10.831	8.839
27*	10.30	360.290[a]	262.440[b]	333.174[a]	343.186[a]
28*	10.37	19.449[a]	14.237[b]	17.937[a]	18.250[a]
29	10.45	3.614[a]	2.374[b]	3.220[ab]	3.250[ab]
30*	10.55	0.830[a]	0.536[b]	0.735[a]	0.742[a]
31	10.67	1.424[a]	0.932[b]	1.323[a]	1.396[a]
32	10.85	1.522	1.143	1.229	1.475
33*	11.04	165.359[a]	128.287[b]	162.585[a]	166.270[a]
34	11.18	0.336	0.246	0.346	0.348
35	11.45	1.604[a]	1.280[b]	1.672[a]	1.668[a]
36	11.62	0.252	0.175	0.222	0.221
37*	11.97	8.244[a]	6.437[b]	8.290[a]	8.539[a]
38	12.45	1.139[a]	0.816[b]	1.129[a]	1.096[ab]
39	12.56	0.306	0.223	0.314	0.311
40	12.88	0.744	0.330	0.386	0.467
41	13.15	1.429[a]	0.778[b]	1.001[b]	1.033[b]
42	13.22	0.668[a]	0.291[b]	0.374[b]	0.492[ab]
43*	13.30	3.218[a]	1.533[c]	2.247[b]	2.403[b]
44	13.90	0.544[a]	0.331[b]	0.429[ab]	0.378[b]
45*	14.13	1.446[a]	0.793[c]	1.012[b]	1.107[b]
46	14.60	1.246	0.988	1.092	1.232
47	14.89	1.509	1.244	1.422	1.462
48	15.91	1.217[a]	0.917[b]	1.057[ab]	1.163[a]
49	18.22	0.247	0.096	0.079	0.157
50*	19.35	0.593[a]	0.339[b]	0.450[ab]	0.464[ab]
51	19.65	0.664[a]	0.407[b]	0.539[ab]	0.578[a]
SFA[c]		292.670	273.586	311.403	299.029
MUFA[c]		452.389[a]	352.386[b]	435.362[a]	439.658[a]
PUFA[c]		185.566[a]	143.369[b]	181.608[a]	185.979[a]
total		990.860	842.396	995.150	988.745

[a] Refer to Table 2 for peak identifications. [b] Means in the same row with the same letter do not differ significantly ($P \leq 0.05$), *($P \leq 0.01$). [c] SFA, saturated; MUFA, monounsaturated; PUFA, polyunsaturated.

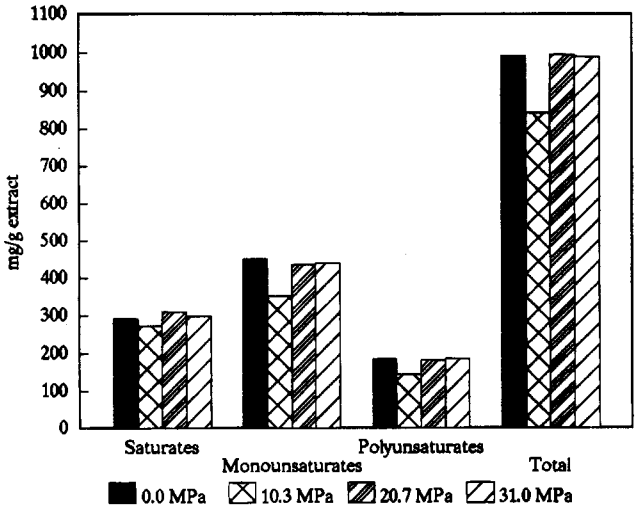

Figure 1. Concentrations of fatty acid classes in SC-CO_2 extracts of chicken fat.

Table 4. Ratios of Saturates/Unsaturates (S/U), Saturates/Monounsaturates (S/M), and Saturates/Polyunsaturates (S/P) in Supercritical Carbon Dioxide Extracts of Chicken Fat

	pressure			
ratio	0.0 MPa	10.3 MPa	20.7 MPa	31.0 MPa
S/U	0.459	0.552	0.505	0.478
S/M	0.647	0.776	0.715	0.680
S/P	1.577	1.908	1.715	1.608

and most likely to be exhaustively extracted under all extraction conditions.

Work performed by Maheshwari *et al.* (1992) showed an increase in solubility of dodecanoic (12:0) and tetradecanoic (14:0) free fatty acids with increasing extraction pressure at constant temperature. Data presented here indicate that these more volatile fatty acids, which were likely extracted as a portion of a triacylglycerol molecule and not in their unbound form, were concentrated in the lower pressure extract (Table 3) where competing molecules were less soluble in the extraction medium. While the inherent solubility of these compounds likely increases with extraction pressure, the molecules have more competition for solvent phase in this system at higher pressures.

Volatile Quantitation and Identification. Quantitated volatile compounds numbered 318, of which 99 were grouped into 12 compound classes—enals, aldehydes, ketones, lactones, alkanes, branched alkanes, alcohols, acids, alkenes, halogenateds, aromatics, and heteroatomics (Table 5). Thermal breakdown of fatty acids and oxidation of unsaturated lipids can account for compounds representative of all volatile classes (Ellis *et al.*, 1961; Hoffman, 1962; MacLeod and Seyyedain-Ardebili, 1981) except aromatics and halogenated alkanes, which likely derive from the breakdown of aromatic amino acids in connective tissue or feed constituents and entrapped water, respectively. The largest numbers of classified volatiles were branched alkanes (38), enals (12), and aldehydes (9).

By concentration, branched alkanes, aldehydes, and enals were predominant (Table 6) in all treatments followed by alkenes in all but one treatment. Lactone concentration exceeded that of alkenes only in the 10.3 MPa extract. Treatments affected total volatile concentration, which was higher for all extracts versus control and, among extracts, increased with decreasing extraction pressure. Total volatiles were concentrated over 12-fold in 10.3 MPa extract versus control. Similarly, concentrations of enals, ketones, lactones, branched alkanes, alcohols, and halogenated and aromatic compounds were affected by treatment, with concentration

Table 5. Identified Volatiles in Supercritical Carbon Dioxide Extracts of Chicken Fat

enals	aldehydes
2,4-hexadienal	pentanal
2-hexenal	hexanal
2(*E*)-heptenal	heptanal
2(*E*),4(*E*)-heptadienal	octanal
2-octenal	nonanal
2(*E*)-nonenal	decanal
2,4-nonadienal	tetradecanal
2(*E*)-decenal	pentadecanal
2(*E*),4(*E*)-decadienal	hexadecanal
2(*E*),4(*Z*)-decadienal	
2-undecenal	alkanes
	cyclohexane
lactones	heptane
δ-decalactone	tetradecane
γ-decalactone	
δ-dodecalactone	alcohols
δ-tetradecalactone	1-penten-3-ol
	1-nonen-3-ol
branched alkanes	tetradecanol
methylcyclopentane	hexadecanol
2,3,4-trimethylpentane	
2,3,3-trimethylpentane	acids
2,2,4-trimethylpentane	tetradecanoic acid
2-methylheptane	hexadecanoic acid
4-methyloctane	octadecanoic acid
2,6-dimethylheptane	
3,5-dimethylheptane	alkenes
2,4-dimethylheptane	1-octene
1,2,4-trimethylcyclohexane	2-octene
2,2,5-trimethylheptane	2-methyl-2-heptene
2,2,4-trimethylheptane	2-methyl-2-nonene
2,2,6-trimethylheptane	4-methyl-2-undecene
3,3,5-trimethylheptane	
3-methylnonane	halogenated
2,5- or 2,6-dimethylundecane	1,1,1-trichloroethane
3,6-dimethylundecane	bromodichloromethane
2,8-dimethylundecane	dibromochloromethane
2,2,6- or 2,2,7-trimethyldecane	dichlorobenzene
3,3,5-trimethyldecane	
2,5-dimethyldodecane	heteroaromatics
2-methyl-6-propyldodecane	2-methyltetrahydrofuran
5-propyltridecane	
	ketones
aromatics	1-penten-3-one
toluene	2-heptanone
1,3-xylene	2,3-octanedione
xylene isomer	heptadecanone
2-pentylfuran	2-pentadecanone
2,6-di-*tert*-butyl-4-methylphenol	dodecanone

Table 6. Volatile Class Concentrations of Supercritical Carbon Dioxide Extracts of Chicken Fat

	concentration[a] (ppm) at			
class	0.0 MPa	10.3 MPa	20.7 MPa	31.0 MPa
enals	1.368^{c}	15.753^{a}	10.653^{ab}	4.063^{bc}
aldehydes	1.428	16.323	11.962	6.092
ketones*	0.108^{b}	3.207^{a}	2.815^{a}	1.059^{b}
lactones*	0.165^{d}	4.432^{a}	1.773^{b}	0.848^{c}
alkanes	0.140	1.695	1.061	0.694
branched alkanes	4.258^{b}	26.791^{a}	14.451^{b}	9.097^{b}
alcohols	0.054^{b}	3.598^{a}	2.039^{ab}	0.800^{b}
acids	0.520	3.472	2.176	0.041
alkenes	0.533	4.404	3.163	2.272
halogenateds	0.061^{b}	0.700^{a}	0.542^{a}	0.397^{ab}
aromatics	0.059^{c}	1.648^{a}	1.148^{ab}	0.566^{bc}
heteroatomics	0.000	0.045	0.000	0.067
total*	9.945^{c}	123.795^{a}	76.627^{ab}	37.489^{bc}

[a] Means in the same row with the same letter do not differ significantly ($P \leq 0.05$), *($P \leq 0.01$).

factors ranging from 6 for branched alkanes to 66 for alcohols. Aldehyde, alkane, and alkene concentrations followed a similar trend but did not differ significantly ($P \leq 0.05$). Acids and heteroatomics followed no clear concentration trend across treatments and did not differ significantly, but some of these classes had higher concentrations over the control (Table 6). All pressure treatments possessed sufficient solvating ability in the SC-CO_2 to affect volatile concentration over the unextracted control. Among extractions, the volatile concentration trend is due to the supercritical fluid's decreased ability at lower pressures to solubilize less volatile and higher molecular weight volatiles, fatty acids, and triacylglycerols, enabling volatiles to be selectively extracted and concentrated. At higher pressures, more types of compounds compete for position in the supercritical fluid phase. Merkle and Larick (1994b) similarly achieved concentration of beef fat volatiles at lower pressures. Indeed, this point near the critical point of CO_2 is on the steeper portion of the solubility curve, where the most efficient separatory power is expected (Allada, 1984).

Conclusions. Extraction of cooked chicken lipids with various pressures using SC-CO_2 facilitated the fractionation of the lipids based on individual fatty acid and class concentrations and volatile class profiles. This type of fractionation is not possible using traditional extraction techniques that utilize solvent systems with a constant solvating ability. Further refinement of this process could yield the ability to produce, via fractionation, lipid-derived extracts with altered fatty acid and/or volatile profiles, useful for specific functional properties and flavor applications in food systems.

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