Volatile Sulfur-Containing Compounds Generated from the Thermal Interaction of Corn Oil and Cysteine

Carlos Macku and Takayuki Shibamoto*

Department of Environmental Toxicology, University of California, Davis, California 95616

Headspace volatile compounds produced from corn oil heated with cysteine at 180 °C were collected by a simultaneous purging and solvent extraction apparatus. Aroma components were analyzed by gas chromatography and gas chromatography/mass spectrometry. Fifty-four compounds were positively identified among over 100 gas chromatographic peaks. The identification of sulfur-containing compounds was confirmed by a flame photometric detector. The compounds identified were one aldehyde, three alcohols, five ketones, five alkanes, five alkenes, two furans, three pyridines, two pyrroles, five alkylbenzenes, eight thiophenes, four thiiranes, three thiols, one thiazole, five thiazolidines, and two trithiolanes. α,β-Unsaturated fatty aldehydes are proposed to react with hydrogen sulfide, a cysteine pyrolysis product, to form 2-alkylthiophenes.

INTRODUCTION

Volatile sulfur-containing compounds are important flavor constituents of many heated foods (Shankaranarayana et al., 1974). Sulfur-containing amino acids and proteins are the main precursors for these aroma compounds (Schutte, 1974). For example, cysteine is an important precursor for cooked-meat volatiles (Schrödter et al., 1982), and it is used as an additive to improve the flavor of processed foods (Oser and Hall, 1972).

Volatiles formed from the thermal interaction between cysteine and carbohydrates have been studied extensively (Schutte, 1974). However, few studies on the volatiles produced from the cysteine/fatty acid interaction have been reported. Severin and Ledl (1972) and Ledl and Severin (1973) studied for the first time the effect of fatty acids over the Maillard reaction using a cysteine/tributyrin/xylose model system. Recently, the effect of phospholipids in a heated cysteine/ribose mixture was investigated (Whitfield et al., 1988; Farmer et al., 1989). Zhang and Ho (1989) reported on the volatiles generated from the thermal interaction between cysteine and 2,4-decadienal, a secondary product of linoleic acid peroxidation.

In the present study, the headspace volatiles formed from the thermal interaction between cysteine and corn oil were identified and quantified in efforts to understand the formation mechanisms of volatile sulfur-containing compounds found in lipid-rich cooked foods.

MATERIALS AND METHODS

Materials. Corn oil (Mazola brand) was purchased from a local market. Cysteine was bought from Sigma Chemical Co. (St. Louis, MO). All authentic samples were obtained from reliable commercial sources.

Sample Preparation. Corn oil (100 g) and cysteine (1.0 g) were placed in a 500-mL two-neck, round-bottom flask, which was interfaced to a simultaneous purging and solvent extraction apparatus (SPE) devised by Umano and Shibamoto (1987). The mixture was stirred and kept at 180 °C for 4 h. The headspace was purged into the SPE with a purified air stream (flow rate, 10 mL/min). The dichloromethane extract was concentrated to 2 mL by fractional distillation.

Instrumental Analyses of Headspace Volatiles. The gas chromatographic (GC) retention index (Kovats, 1965) and mass spectral (MS) fragmentation pattern of each component were

compared with those of the authentic compounds for qualitative analysis. A nitrogen–phosphorus detector (NPD) and a flame photometric detector (FPD) were used to confirm the presence of nitrogen- and sulfur-containing volatile compounds, respectively.

A Hewlett-Packard (HP) 5880 gas chromatograph equipped with a 60 m × 0.25 mm i.d. DB-5 bonded-phase fused-silica capillary column (J&W Scientific, Folsom, CA), a flame ionization detector (FID), and an NPD was used for sample analysis. The GC peak areas were integrated with an HP 5880A series terminal. The same capillary column was also installed in an HP 5890 Series II gas chromatograph equipped with an FPD and interfaced to a Spectra Physics 4290 integrator. Injection temperature was 250 °C, and detector temperatures were 300 °C for the FID, 220 °C for the NPD, and 230 °C for the FPD. The oven temperature was held at 40 °C for the first 5 min and was then programmed to 160 °C at 2 °C/min. The linear helium carrier gas flow rate was 30 cm/s.

An HP 5890 gas chromatograph interfaced to a VG Trio II mass spectrometer with a VG 11-250 computer data system was used for MS identification of the GC components. The column and oven conditions for GC/MS were as described for the HP 5880 gas chromatograph.

RESULTS AND DISCUSSION

A typical FID gas chromatogram of the headspace volatiles obtained from the heated corn oil/cysteine mixture is shown in Figure 1. Fifty-four compounds were positively identified and seven others were tentatively identified, as shown in Table I. Most of the hydrocarbons, oxygen-containing compounds, and nitrogen-containing compounds found in this work were also found in heated corn oil alone and in heated corn oil/glycine mixtures (Macku and Shibamoto, 1991). The sulfur-containing compounds found in the present study are reportedly generated from the pyrolysis of cysteine (Fujimaki et al., 1969; Zhang et al., 1988) and from the interaction of secondary products of fatty acid peroxidation (Frankel, 1985) with cysteine pyrolysis products (Farmer et al., 1989; Whitfield et al., 1988).

Most headspace fatty aldehydes found in the corn oil/glycine mixture (Macku and Shibamoto, 1991) were not detected in the headspace of corn oil heated with cysteine, suggesting that cysteine is more reactive than glycine. This observation correlates with the high reactivity of cysteine found in foods such as roasted coffee (Feldman et al., 1969) and cooked meat (Schrödter et al., 1982).

Thiophene (peak 15) has been reported as a cysteine pyrolysis product (Zhang et al., 1988). 2-Alkylthiophenes
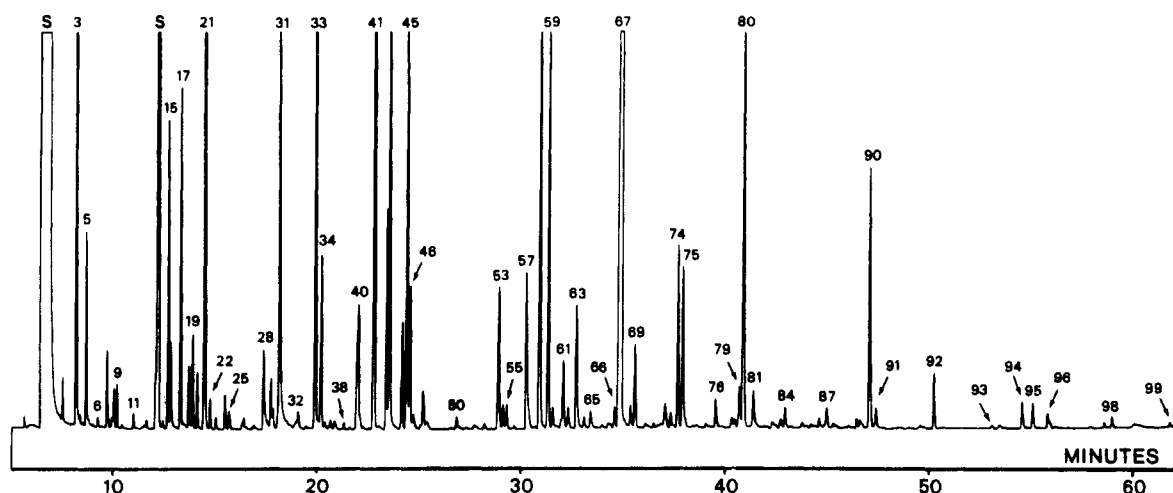

Figure 1. Typical gas chromatogram of the headspace volatiles generated from heating 100 g of corn oil with 1.0 g of cysteine. Chromatography was performed on a 60 m × 0.25 mm i.d. DB-5 bonded-phase fused-silica capillary column. Temperature program: 2 °C/min, starting at 40 °C (hold-up time = 5 min) up to 160 °C. Sample solvent (S) is a mixture of dichloromethane and cyclohexane.

Table I. Headspace Volatiles from Heated Corn Oil with Cysteine

peak[a]	compound	peak area, %	I_{DB-5}[b]	N[c]	S[d]	RT[e]	MS[f]
3	thiirane (ethylene sulfide)	11	589		+[i]		+
5	butanal[g]	0.7	596			+	+
6	propanethiol	0.05	614		+	+	+
9	methylcyclopentane[h]	0.2	635				+
11	methylthiirane	0.06	650		+		+
15	thiophene	1.2	677		+	+	+
17	1-penten-3-ol[g]	1.3	685			+	+
18	2-pentanone[g]	0.3	690			+	+
19	1-heptene[g]	0.4	693			+	+
20	cis-dimethylthiirane	0.3	695		+		+
21	heptane[g]	5.6	700			+	+
22	2-ethylfuran[g]	0.1	705			+	+
23	cis-2-heptene	0.05	709			+	+
25	butanethiol	0.1	718		+	+	+
28	thiazole	0.4	745	+	+	+	+
29	trans-dimethylthiirane	0.2	746		+		+
31	pyridine	2.3	750	+		+	+
32	pyrrole	0.07	762	+		+	+
33	1-pentanol[g]	4.1	771			+	+
34	toluene	0.8	774			+	+
36	2-methylthiophene	0.06	780		+	+	+
38	3-methylthiophene	X[j]	786		+	+	+
39	3-hexanone[g]	X	788			+	+
40	2-hexanone[g]	0.9	793			+	+
41	octane[g]	13	800			+	+
42	cis-2-octene[g]	1.1	808			+	+
44	trans-2-octene[g]	0.6	818			+	+
45	1-ethylpyrrole	2.3	820	+		+	+
46	pentanethiol	0.8	822		+	+	+
50	2-methylcyclopentanone[g,h]	X	848				+
53	3-methylpyridine; ethylbenzene	0.8	869	+		+	+
55	2-ethylthiophene	0.1	874		+	+	+
57	2-methyl-4,5-dihydrothiazole[h]	1.1	883	+	+		+
59	2-heptanone[g]	2.4	893			+	+
61	nonane[g]	0.4	900			+	+
63	thiazolidine	NA[k]	909	+	+	+	+
64	3-ethylthiophene	0.07	912		+	+	+
65	trans-2-nonene	0.1	916			+	+
66	1,3-nonadiene[h]	0.1	930				+
67	2-methylthiazolidine	29	934	+	+	+	+
68	propylcyclohexane[g,h]	0.1	939				+
69	butylcyclopentane[g,h]	0.4	941				+
74	propylbenzene	1.0	964			+	+
75	2-propylthiophene	1.0	966		+	+	+
76	1-octen-3-ol[g]	0.2	982			+	+
79	2-octanone[g]	0.3	993			+	+
80	2-pentylfuran[g]	2.5	995			+	+
81	decane[g]	0.2	1000			+	+
84	3-propylthiophene	0.1	1019		+	+	+
85	2-ethylthiazolidine	X	1034	+	+	+	+
87	5-ethyl-4-methylthiazole[h]	0.2	1043	+	+		+
90	butylbenzene[g]	1.6	1067			+	+
91	2-butylthiophene	0.1	1069		+	+	+
92	undecane[g]	0.3	1100			+	+
93	2-propylthiazolidine	X	1135	+	+	+	+
94	cis-3,5-dimethyl-1,2,4-trithiolane	0.2	1154		+		+
95	trans-3,5-dimethyl-1,2,4-trithiolane	0.2	1161		+		+
96	pentylbenzene	0.1	1168			+	+
98	pentylpyridine	0.07	1205	+		+	+
99	2-butylthiazolidine	X	1243	+	+	+	+

[a] Peak numbers in Figure 1. [b] Kovats index values on DB-5 capillary column. [c] Detection by nitrogen–phosphorus detector. [d] Detection by flame photometric detector. [e] Identification carried out by comparing retention times with those of the authentic compounds. [f] Identification carried out by comparing mass spectra with those of the authentic compounds. [g] Previously reported in heated corn oil alone (Macku and Shibamoto, 1991). [h] Tentative identification. [i] Positive identification. [j] GC peak area less than 0.01. [k] Not available.

(peaks 36, 55, 75, 91) have been found in several fatty acid/cysteine model systems (Whitfield et al., 1988; Farmer et al., 1989; Zhang and Ho, 1989). These compounds are probably formed from the interaction between fatty aldehydes and hydrogen sulfide (van der Ouweland et al., 1989), a cysteine pyrolysis product. 2-Ethylthiophene was thermally generated from a mixture containing 2-hexenal and hydrogen sulfide (Boelens et al., 1974). In our study, 2-propylthiophene (peak 75), a seven-carbon atom molecule, was the alkylthiophene produced in greatest amount (Table I). Its concentration correlates with the amount of 2-heptenal, which was the most prominent α,β-unsaturated volatile aldehyde found in heated corn oil (Macku and Shibamoto, 1991). A proposed formation mechanism for these heterocyclic compounds is shown in Figure 2. A nucleophilic attack by the hydrogen sulfide molecule on the α,β-unsaturated aldehyde replaces the oxygen atom with a sulfur atom. The resulting sulfur-containing radical, formed by hydrogen abstraction at the γ-carbon, rearranges its electrons and reacts with a surrounding hydroxyl radical to undergo dehydration and thiophene ring formation.

Thiirane or ethylene sulfide (peak 3) is a cysteine pyrolysis product (Sakaguchi and Shibamoto, 1978). It has also been found in cysteine/glucose model systems exposed to UV light (Sheldon and Shibamoto, 1988). Ethylene sulfide and propylene sulfide (peak 11) were reported in cooked foods such as heated beef (MacLeod and Seyyedain-Ardebili, 1981), heated soya protein, casein, and fish (Qvist and von Sydow, 1974).

Propanethiol, butanethiol, and pentanethiol (peaks 6, 25, and 46, respectively) are probably the products of the
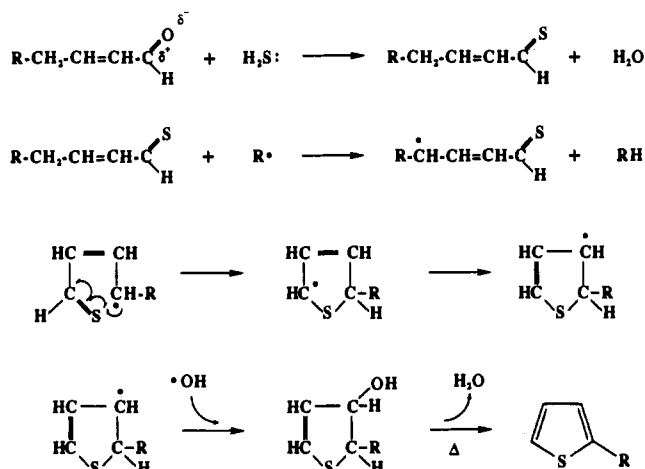
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Figure 2. Suggested mechanism for the thermally formed 2-alkylthiophenes found in the headspace of corn oil/cysteine mixture.

reactions between hydrogen sulfide and propanol, butanol, and pentanol (Chang et al., 1978), respectively.

With the exception of 2-methylthiazolidine (peak 67), only trace amounts of 2-alkylthiazolidines were found in the present study (peaks 63, 85, 93, 99). 2-Methylthiazolidine, which is the main component of the heated corn oil/cysteine mixture, is reportedly a main product of cysteine pyrolysis (Fujimaki et al., 1969). However, other 2-alkylthiazolidines are probably the reaction products of saturated fatty aldehydes and cysteamine, a cysteine decarboxylation product (Sakaguchi and Shibamoto, 1978).

3,5-Dimethyl-1,2,4-trithiolanes (peaks 94, 95) are thermal degradation products of cysteine (Zhang et al., 1988). These compounds were reported to have a typical boiled-beef aroma (Chang et al., 1968).

Even though the content of sulfur-containing amino acids found in food proteins is small in relation to other amino acids (Cheftel and Cuq, 1985), cysteine is an important source of key flavor compounds. The amounts of sulfur-containing aroma compounds produced during cooking and food processing are usually minute. However, these compounds are very noticeable because of their low odor thresholds (Maga, 1975).

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