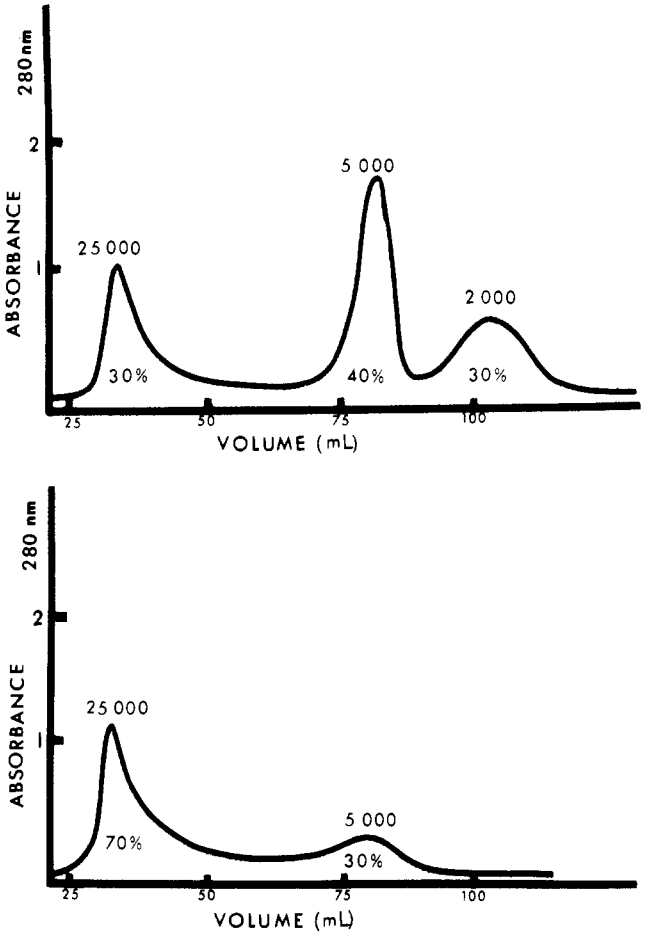

Figure 1. Sephadex G-25 filtration of two liquid protein products.

molecular weight of 5000, and 30% with a molecular weight of 25 000 or more. The second product contained 30% of the peptides with a molecular weight of 5000 and 70% with a molecular weight of 25 000 or more. Some of these high molecular weight fractions were collected and subjected to further gel filtration. The results indicated that the polypeptides were as large as 100 000. The variability in peptide length probably reflects differences in the degree of hydrolysis achieved by the various manufacturing processes.

Amino acid profiles of the completely hydrolyzed liquid protein products are shown in Table I. The amounts of amino acids found in the samples differed considerably from the amounts declared on the label. For the 17 products that had label declarations, analyses showed from 0% of the label declaration for tyrosine to 424% of the label declaration for methionine.

The quantitative label declaration on the liquid protein products stated that the only source of calories in the products was derived from protein, in contrast to the ingredient statement which indicated that the product contained glycerol or sorbitol or both. Analytical results showed that 6 of 17 liquid protein products contained from 0.1 to 5.9 g of glycerol/30 mL of product. The latter would add 23.6 cal/serving. Nine of the liquid protein products contained sorbitol. The lowest level was 0.3 g/30 mL; the highest level was 7.5 g/30 mL of sample, which would contribute an additional 30 cal/serving. Three products contained neither glycerol nor sorbitol. None of the products contained xylitol.

The data available at this time do not provide any evidence of a relationship between the composition of liquid protein products and the deaths of persons ingesting them.

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Mass and Nuclear Magnetic Resonance Spectra of Some Alkylpyrazines

The mass spectra and NMR spectra of 11 alkylpyrazines were measured. The alkylpyrazines were synthesized from dihydropyrazine and the corresponding aldehyde or ketone. The major mass spectral fragmentation of the alkylpyrazines underwent McLafferty rearrangement. The compounds which showed similar mass fragmentation patterns were easily distinguished by NMR spectra.

Since the development of the direct combination of capillary gas chromatography and mass spectrometry (GC–MS), the comprehensive analysis of flavor extracts from certain foods has become practical. Mass spectral

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Table I. Spectral Data and Odor Descriptions of Alkylpyrazines

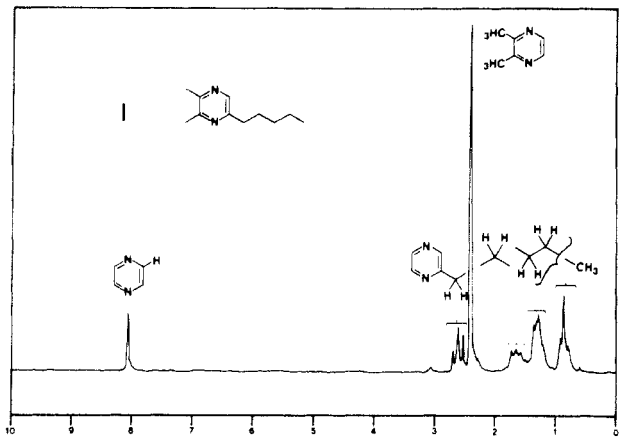
	compd	mass spectra	NMR spectra	odor descriptions
I	2,3-dimethyl-5-*n*-pentylpyrazine	42 (14), 80 (18), 94 (8), 122 (100), 135 (58), 149 (36), 163 (10), M = 178 (24)	δ 0.85 (t, 3 H, *J* = 6 Hz), 1.15-1.45 (m, 4 H), 1.45-1.85 (m, 2 H), 2.4 (s, 6 H), 2.62 (t, 2 H), 8.06 (s, 1 H)	sweet, smoked, caramel-like
II	2,3-dimethyl-5-isopentylpyrazine	42 (14), 80 (22), 94 (8), 122 (100), 135 (59), 149 (14), 163 (32), M = 178 (11)	δ 0.95 (d, 6 H, *J* = 5.7 Hz), 1.3-1.8 (m, 3 H), 2.45 (s, 6 H), 2.67 (t, 2 H), 8.18 (s, 1 H)	caramel-like, coffee, sweet
III	2,3-dimethyl-5-(2-methylbutyl)pyrazine	42 (18), 80 (22), 94 (9), 122 (100), 42 (18), 80 (22), 94 (9), 122 (100), 123 (46), 135 (23), 149 (39), 163 (41), M = 178 (16)	δ 0.85 (d, 3 H, *J* = 7.5 Hz), δ 0.85 (d, 3 H, *J* = 7.5 Hz), 0.88 (t, 3 H), 1.1-1.4 (m, 2 H), 1.7-2.1 (m, 1 H), 2.45 (s, 6 H), 1.5-1.7 (m, 2 H), 8.07 (s, 1 H)	sweet, smoked, caramel-like
IV	2,3-dimethyl-5-neopentylpyrazine	42 (24), 57 (78), 80 (18), 94 (8), 121 (30), 122 (100), 123 (36), 163 (45), M = 178 (27)	δ 0.93 (s, 9 H), 2.46 (s, 6 H), 2.57 (s, 2 H), 8.08 (s, 1 H)	brown sugar like, roasted
V	2,3-dimethyl-5-(1,2-dimethylpropyl)pyrazine	42 (30), 80 (9), 108 (30), 122 (86), 135 (100), 136 (94), 137 (46), 150 (16), 163 (64), M = 178 (37)	δ 0.75 (d, 3 H, *J* = 7.2 Hz), 0.93 (d, 3 H, *J* = 7.2 Hz), 1.23 (d, 3 H, *J* = 7.2), 1.7-2.2 (m, 1 H), 2.45 (s, 6 H), 2.3-2.7 (m, 1 H), 8.08 (s, 1 H)	sweet, caramel-like
VI	2,3-dimethyl-5-(1-methylbutyl)pyrazine	42 (15), 80 (11), 108 (30), 122 (16), 135 (95), 136 (100), 137 (40), 149 (54), 163 (37), M = 178 (15)	δ 0.85 (t, 2 H), 1.1-1.3 (m, 3 H), 1.23 (d, 6 H, *J* = 7.5 Hz), 1.4-1.8 (m, 2 H), 2.43 (s, 6 H), 2.6-3.0 (m, 1 H), 8.08 (s, 1 H)	honey-like, sweet
VII	2,3-dimethyl-5-(1-ethylpropyl)-pyrazine	42 (8), 53 (8), 80 (24), 108 (8), 122 (95), 135 (100), 136 (45), 149 (95), 150 (93), 163 (54), M = 178 (41)	δ 0.93 (t, 6 H, *J* = 7.2 Hz), 1.4-1.8 (m, 2 H), 2.2-2.6 (m, 2 H), 2.43 (s, 6 H), 8.04 (s, 1 H)	honey-like, sweet
VIII	2-isobutyl-3,5,6-trimethylpyrazine	42 (38), 53 (42), 54 (32), 80 (11), 94 (30), 122 (24), 135 (51), 136 (100), 163 (54), 177 (32), M = 178 (44)	δ 0.92 (d, 6 H, *J* = 7.2 Hz), 1.9-2.4 (m, 1 H), 2.41 (s, 9 H), 2.56 (d, 2 H, *J* = 6.9 Hz)	roasted nut, sweet
IX	2-(2-methylbutyl)-3,5,6-trimethylpyrazine	42 (8), 53 (16), 94 (8), 135 (27), 136 (100), 137 (38), 164 (16), 177 (27), M = 192 (5)	δ 0.83 (d, 3 H, *J* = 7.2), 0.87 (t, 3 H), 1.1-1.5 (m, 2 H), 1.6-2.1 (m, 1 H), 2.43 (s, 9 H), 2.4-2.8 (m, 2 H)	sweet, roasted
X	2,3-dimethyl-5-(1,5-dimethyl-4-hexenyl)pyrazine	41 (10), 85 (9), 108 (15), 135 (78), 136 (100), 137 (49), 149 (32), 175 (9), 203 (8), M = 218 (34)	δ 1.23 (d, 3 H, *J* = 7.2 Hz), 1.57 (d, 6 H, *J* = 12 Hz), 1.47-1.93 (m, 4 H), 1.47-1.93 (m, 4 H), 1.47-1.18 (m, 1 H), 2.45 (s, 6 H), 2.6-2.9 (m, 1 H), 4.9-5.1 (m, 1 H), 8.08 (s, 1 H)	roasted nut
XI	2,3-dimethyl-5-(3,7-dimethyl-7-heptenyl)pyrazine	41 (8), 69 (14), 109 (14), 122 (100), 123 (35), 135 (86), 136 (32), 150 (35), 163 (18), 177 (30), 231 (9), M = 246 (38)	δ 0.95 (d, 3 H, *J* = 5.4 Hz), 1.63 (d, 6 H, *J* = 6 Hz), 1.2-1.8 (m, 6 H), 1.9-2.1 (m, 1 H), 2.45 (s, 6 H), 2.67 (t, 2 H), 5.08 (t, 1 H), 8.11 (s, 1 H)	roasted nut

Table II. Major Fragments from the Mass Spectra of Alkylpyrazines

	McLafferty rearrangement (A, refer to Figure 2)			γ cleavage (B, refer to Figure 2)		
compd	fragment	*m/e*	intensity, %	fragment	*m/e*	intensity, %
I	R_1, R_2 = H	122	100	R_1, R_2, R_3, R_4 = H	135	58
II	R_1, R_2 = H	122	100	R_1, R_2, R_3, R_4 = H	135	60
III	R_1, R_2 = H	122	100	R_1, R_2, R_3 = H; R_4 = C_2H_5	163	41
				R_1, R_2, R_3, R_4 = H	135	23
IV	R_1, R_2 = H	122	100	R_1, R_2 = H; R_3, R_4 = CH_3	136	45
V	R_1 = H; R_2 = CH_3	136	94	R_1, R_3 = H; R_2, R_4 = CH_3	163	64
VI	R_1 = H; R_2 = CH_3	136	100	R_1, R_3, R_4 = H; R_2 = CH_3	149	54
VII	R_1 = H; R_2 = C_2H_5	150	93	R_1, R_3, R_4 = H; R_2 = C_2H_5	163	95
VIII	R_1 = CH_3; R_2 = H	136	100	R_1, R_4 = CH_3; R_2, R_3 = H	163	54
IX	R_1 = CH_3; R_2 = H	136	100	R_1, R_4 = CH_3; R_2, R_3 = H	163	16
				R_1 = CH_3; R_2, R_3 = H; R_4 = C_2H_5	177	27
X	R_1 = H; R_2 = CH_3	136	100	R_1, R_3, R_4 = H; R_2 = CH_3	149	32
XI	R_1 = H; R_2 = H	122	100	R_1, R_2, R_3, R_4 = H	135	86

and gas chromatographic data alone permit identification of many food constituents [Nonaka et al. (1967), cooked chicken; Walradt et al. (1971), roasted peanuts; Kinlin et al. (1972), roasted filbert; Mussinan and Walradt (1974), cooked pork liver; Yamaguchi and Shibamoto (1979), Captanopsis flower]. This is especially true for heterocyclic compounds such as pyrazines, thiazoles, thiophenes, and furans, which given very simple and characteristic fragmentations on mass spectra.

Some mass spectra of these compounds listed above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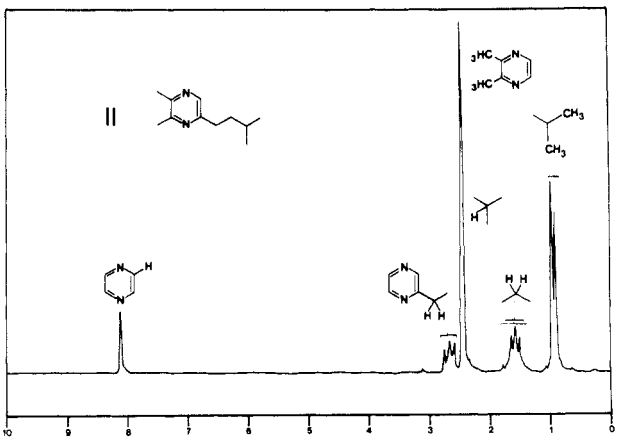
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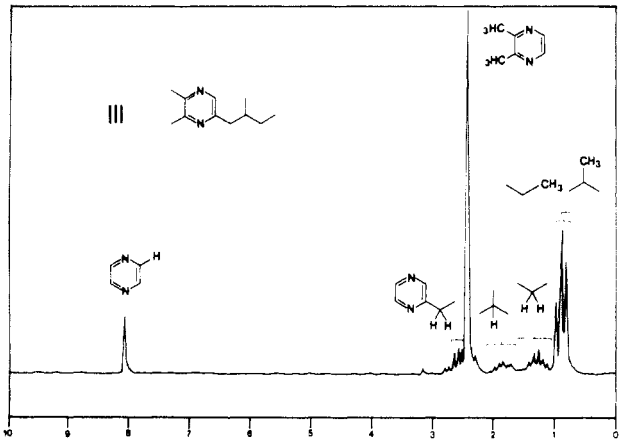
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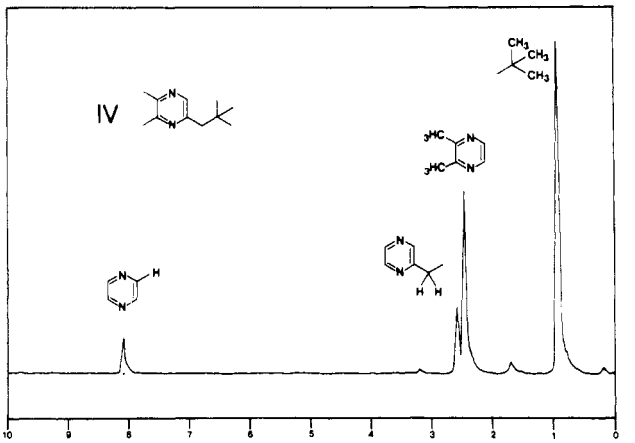
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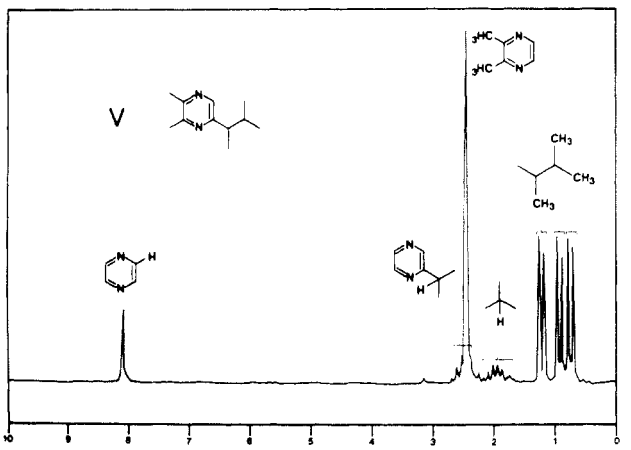
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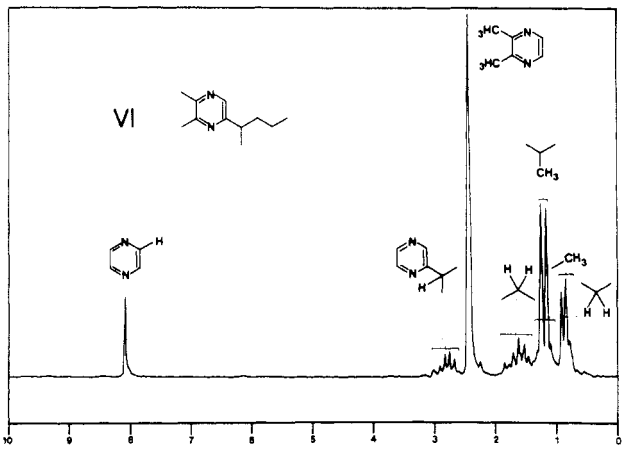
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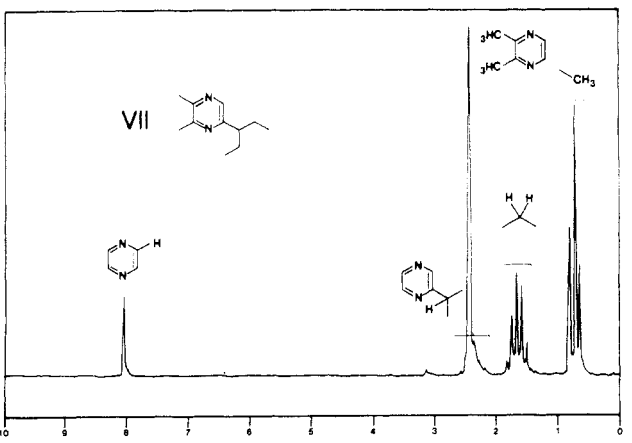
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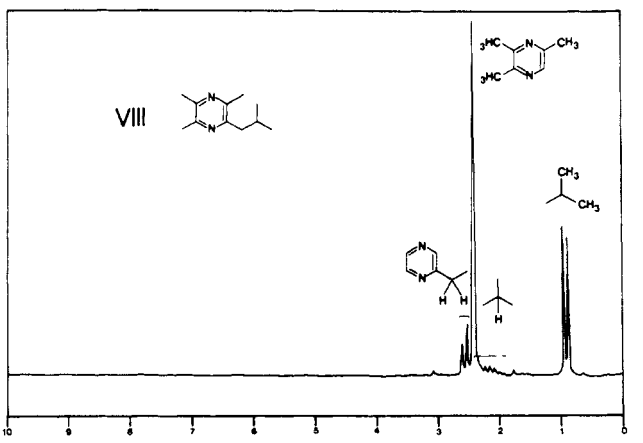
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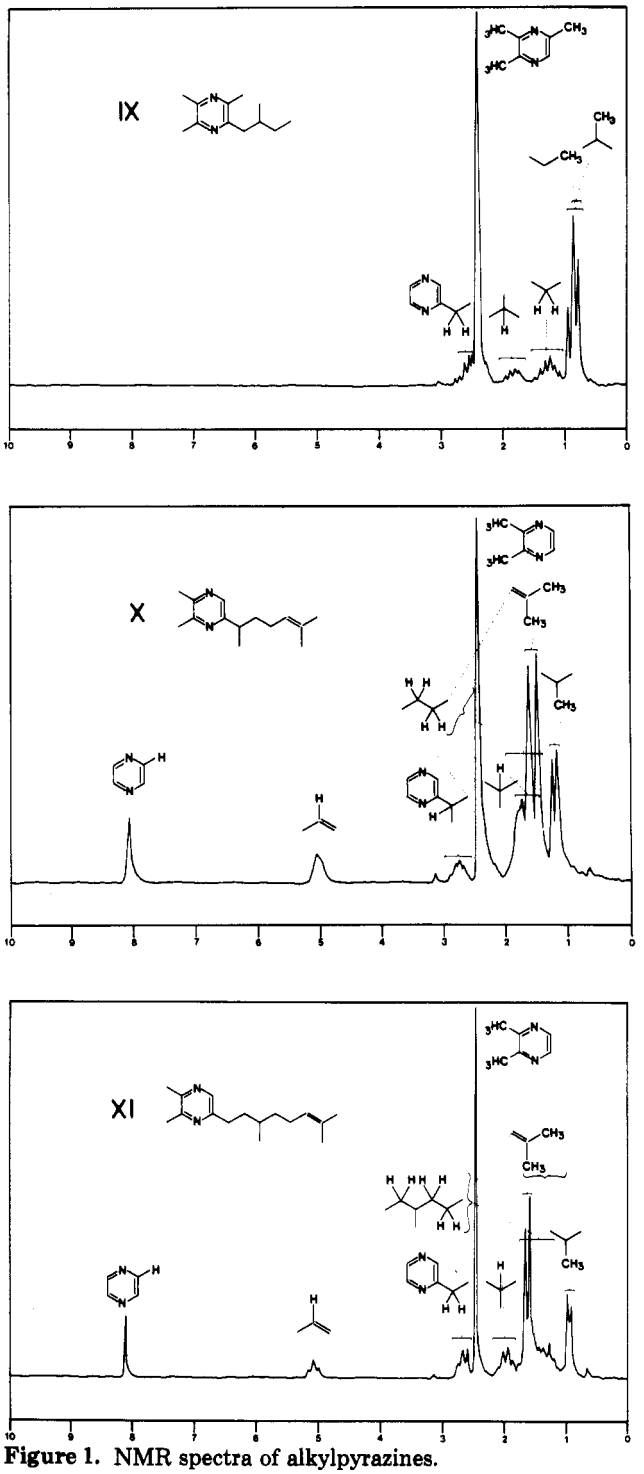


Figure 1. NMR spectra of alkylpyrazines.

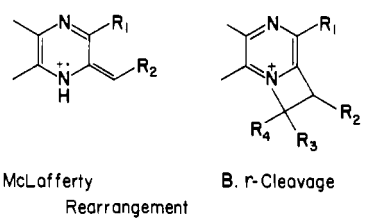
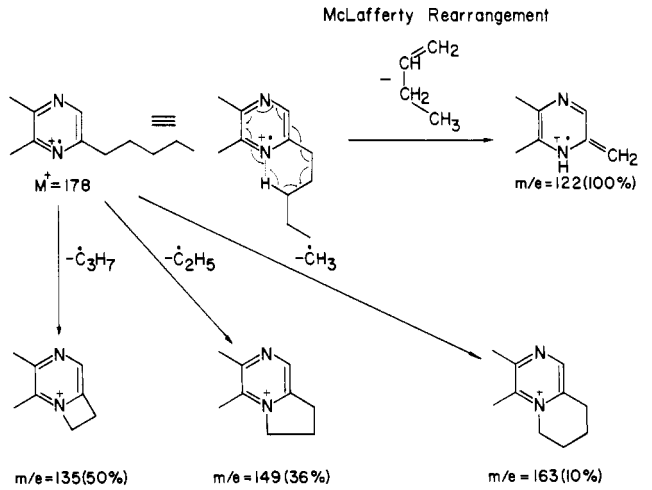


Figure 2. Fragments produced from McLafferty rearrangement and γ cleavage (refer to Table II).

have been published: alkylpyrazines by Bondarovich et al. (1967); bicyclic pyrazines by Walradt et al. (1971), Pittet et al. (1974), and Vitzthum and Werkhoff (1975); pyrrolo[1,2-a]pyrazines by Flament et al. (1977); alkylthiazoles by Buttery et al. (1973); thiophenes by Stoll et al. (1967),

Scheme I



Kato et al. (1973), and Mussinan and Walradt (1974).

Among the heterocyclic compounds, pyrazines have been characterized as the chemicals which contribute the unique roasted or toasted flavor to cooked foods. They exist in almost all cooked or processed foods (Maga and Sizer, 1973) and sometimes comprise nearly half of the volatiles obtained from a meat extract (Mussinan and Walradt, 1974).

Recently, we have developed a new method of pyrazine synthesis and reported some new pyrazines along with their MS, NMR, and IR data (Masuda et al., 1980). In this study, mass spectra and NMR spectra of 11 alkylpyrazines, which have not yet been found in foods, are reported. We hope that these data may be utilized in the search for new flavor components of foods.

EXPERIMENTAL SECTION

Synthesis of Pyrazines. All pyrazines used for this experiment were prepared by the method described by Masuda et al. (1980). All starting chemicals were obtained from reliable commercial sources.

All pyrazine samples were purified by preparative GLC (Perkin-Elmer Model 900) prior to instrumental analysis.

Mass Spectrometer. A Hitachi Model RMU-6M mass spectrometer equipped with a Hitachi Model M-6010 and 10 11/A data system was used under the following conditions: ionization voltage, 70 eV; emission current, 80 μA, ion acceleration voltage, 3100 V; ion source temperature, 200 °C.

NMR Spectrometer. NMR spectra were obtained with a Varian Model EM-390 nuclear magnetic resonance spectrometer (90 MHz) in carbon tetrachloride with tetramethylsilane as an internal standard.

RESULTS AND DISCUSSION

Table I lists the mass spectra of the pyrazines by using the method of presentation of Herz et al. (1971) and NMR spectra of the pyrazines by using the presentation method of Walradt et al. (1971). The odor descriptions of the pyrazines are also shown in Table I.

Mass Spectra. Pyrazines which possess an *n*-propyl or longer side chain (containing γ-hydrogen) undergo McLafferty rearrangement. In general, this gives the base peak for the pyrazines studied in this report (Sample et al., 1967). Scheme I shows a typical fragmentation of compound I. Another important fragmentation of alkylpyrazines is γ cleavage of side chains which are ethyl or longer. The resultant fragments are stabilized by ring formation (Spiteller, 1966). These fragments are not base

peaks but can be counted as major peaks (intensity, 10–95%). The fragments produced from McLafferty rearrangement and γ cleavage are shown in Figure 2 (refer to Table II). The strong characteristic peak at m/e 57 (78%) in compound IV is due to the stable *tert*-butylcarbonium ion (Grubb and Meyerson, 1963).

NMR Spectra. NMR spectra of 11 alkylpyrazines and their interpretations are shown in Figure 1. Compounds I, II, and III show very similar fragmentation patterns in their mass spectra. These can easily be distinguished from each other by using NMR spectra of their alkyl substitutes. Compound I shows a typical long aliphatic chain [δ 0.85 (3 H, t, $-CH_3$), 1.15–1.45 (4 H, m, $-CH_2-CH_2-$), 1.45–1.85 (2 H, m, $-CH_2-$)]. Compound II shows a terminal isopropyl group [δ 0.95 [6 H, d, $-CH(CH_3)_2$], 1.3–1.8 [3 H, m, $-CH(CH_3)_2$ and $-CH_2-CH(CH_3)_2$]]. Compound III exhibits neither of these.

The pyrazines can generally be identified by mass spectra alone on the basis of their typical fragmentation patterns. Sometimes, however, it is difficult to identify the structure of side chains by MS alone. In these cases, NMR spectra can be used to confirm the structure of the side chains. The hydrogen atoms on an α-carbon (a carbon atom next to the pyrazine ring) are deshielded to δ 2.5. The chemical shifts of the other hydrogens on the alkyl groups are the same as those of the individual hydrocarbons except for the methyl groups on the α-carbon, which are deshielded to δ 1.23 (compounds V, VI, and X).

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Influence of Polyunsaturation on Thermal Decomposition of Saturated Triacylglycerols

The typical oxidative products of saturated triacylglycerols could not be detected when heated in the presence of ethyl linoleate. The qualitative patterns of the volatiles produced from the heated mixtures were essentially the same as those obtained from the unsaturated ester when heated alone. The major products typical of the autoxidation of saturated tyiacylglycersols were practically absent. However, the relative amounts of specific decomposition products of linoleate autoxidation were altered when heated in the presence of saturated triacylglycerols.

Frying fats as well as food lipids undergo chemical decomposition when food is exposed to heat in the presence of oxygen. Such reactions are responsible for both desirable and undesirable changes in the flavor and nutritional quality of food. A great deal of information is available regarding the oxidative reactions of pure saturated and unsaturated fatty acid systems. However, the effect of the presence of one substrate fatty acid on the oxidative decomposition of another is not clear. Only one study could be found in which the volatile oxidation products from mixtures of specific triacylglycerols were investigated (Selke et al., 1980). Samples containing triolein and trilinolein produced volatiles which could be ascribed to each substrate. On the other hand, heated mixtures of tristearin–trilinolein produced no observable volatiles that could be related to the saturated triglyceride.

The present work was undertaken to study the influence of heat on saturated and unsaturated fats when coexisting in the same reaction medium. Model systems consisting of unsaturated ethyl esters and saturated triacylglycerols were used.

EXPERIMENTAL SECTION

Tricaproin, trilaurin, trimyristin, and ethyl linoleate were purchased commercially and purified by cold finger distillation as described previously (Nawar et al., 1969). One-gram samples of the saturated triacylglycerol alone or the unsaturated ethyl esters alone or mixtures containing 1 g of each were heated in 250-mL round-bottom flasks fitted with ground glass stoppers at 180 °C for 1 h. The methods used for separation and identification of the decomposition products were the same as those previously reported (Crnjar et al., 1980).

RESULTS AND DISCUSSION

Gas chromatographic analyses of heated trilaurin, ethyl linoleate, and the trilaurin–ethyl linoleate mixture are

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