ratio (Nilsson, 1970) is in its clean resolution of colors and elimination of both overlap in red and yellow spectral bands and interference in the measurements by decomposition products.

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Isolation and Identification of Volatile Components from Wild Rice Grain (*Zizania aquatica*)

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The isolation and identification of flavor compounds characteristic of the popular gourmet item, Wild Rice, have heretofore been unreported. Wild rice volatiles were isolated using vacuum steam distillation followed by solvent extraction. The isolate was fractionated by preparative gas chromatography, and the fractions were subjected to gas chromatography–mass spectrometry using 500 ft × 0.03 in. i.d. Carbowax 20M wall-coated open-tubular columns. Identification were based upon flame ionization and alkali flame ionization detector responses, retention indices (I_E), and GC–MS analyses in conjunction with the specific detectors. This first analysis of wild rice resulted in the identification of 112 compounds reported herein.

Wild rice (*Zizania aquatica*) is one of four species of distinct types of annual grasses which grow in shallow moving water near Lake shores, shallow streams, or ponds in the upper Great Lakes region of the United States and Canada. Other species have been identified in North America (*Zizania palustris* and *Zizania texana*) and in Manchuria and the Far East (Rossman et al., 1973).

Wild rice has been harvested by American Indians for centuries as a staple of their diets. Within the past decade the grain has achieved wide recognition for its distinctive flavor by the gourmet food market and more recently has been increasingly used in rice mixes and casserole items which feature wild rice as an ingredient. Native wild stands of the grain once served as the sole supply, but these stands now contribute approximately 1 million pounds of grain annually compared to 5 million pounds from the commercial cultivation of nearly 20 000 acres of wild rice paddies (Lund et al., 1977).

Unlike other cereal grains, will rice undergoes a series of postharvest processing steps which contribute to the development of a commercially acceptable commodity. The mature kernel is harvested as a moist, metabolically active seed which is piled approximately 18 in. deep to undergo a biochemical and microbiological fermentation during which time the flavor develops and the kernel becomes darkly pigmented. Following fermentation, the grain is dried or parched to reduce the kernel moisture to approximately 7%, hulled, and separated into various grades of finished product. The flavor characteristics of commercial wild rice are dictated by variations of these processes and can be broadly described as either "tea-like", "grainy", "earthy", or "toasted". The grainy flavor and at least part of the tea-like flavor is inherent to the grain at harvest time. A lighter grassy flavor is more pronounced in immature grain while starchy flavor variations are more apparent in mature grains. The tea-like flavor is enhanced during fermentation. It is characteristic of high-quality grain and contributes to the overall, full, hearty flavor. It is frequently accompanied by varying degrees of earthy and moldy or musty flavors reflecting the product moisture level during fermentation. Variation of toasted flavor results from the specific design of parching equipment with the direct-fired surface parchers providing a more burnt and smoky character while the forced-pair parchers produce a milder toasted or roasted flavor. The unique processes involved with the production of wild rice contribute not only to its distinctive flavor, but also to the flavor variations of commercial wild rice (Lund et al., 1977).

Literature concerned with the flavor chemistry of wild rice is nonexistent and that for cereal grains is limited. Wheat has been studied most extensively (Hougen et al., 1971; McWilliams and Mackey, 1969; Okada, 1969; El-Basyouni and Towers, 1964) with the flavor chemistry of white bread receiving the greatest attention (Johnson and Sanchez, 1973; Coffman, 1967a,b; Johnson et al., 1966; Collyer, 1964; Kobayasi and Fujimaki, 1965; Zyuz'ko et al., 1973, 1974; Lorenz and Maga, 1972: Mulders, 1973a,b; Mulders and Dhont, 1972; Mulders et al., 1972, 1973). Roasted barley (mugi-cha), used in the production of dark beer and a tea-like beverage, has received attention (Shimizu et al., 1967; Shimizu et al., 1970a,b; Wang et al., 1968, 1969; Collins, 1971) while the volatile carbonyls and amines of malted barley have been identified by several investigators concerned with their contribution to the flavor of beer (Arkima and Ronkainen, 1971; Hrklicka and Dyr, 1968; Damm and Kringstad, 1964; Wagner, 1971; Baerwald Niefind, 1969; Drews et al., 1957; Palamand et al., 1969; Slaughter, 1970; Slaughter and Uvgard, 1971, 1972; Steinke and Paulson, 1964). The volatile components of cooked rice have been studied (Ayano and Furuhashi, 1970; Tanaka, 1972) with specific interest in the factors

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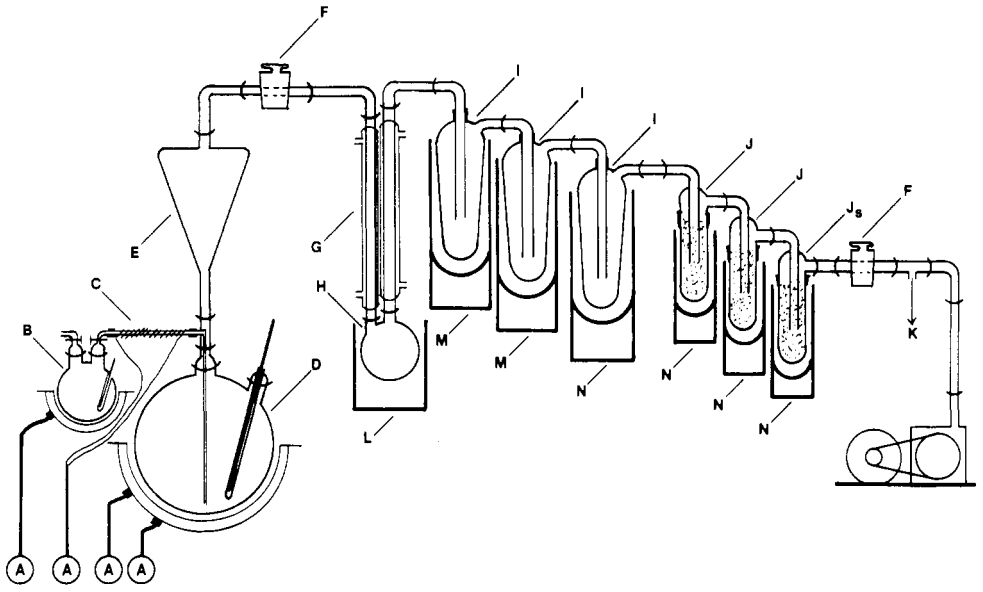

Figure 1. Vacuum steam distillation apparatus: (A) variable transformer, (B) 3-L steam generator, (C) heated transfer line, (D) 12-L sample flask, (E) foam trap, (F) isolation valve, (G) cold water condenser, (H) 3-L cold trap, (I) 2-L cold traps, (J) glass bead filled cold traps, (K) to manometer, (L) dry ice bath, (M) ethanol–dry ice bath, (N) liquid nitrogen bath.

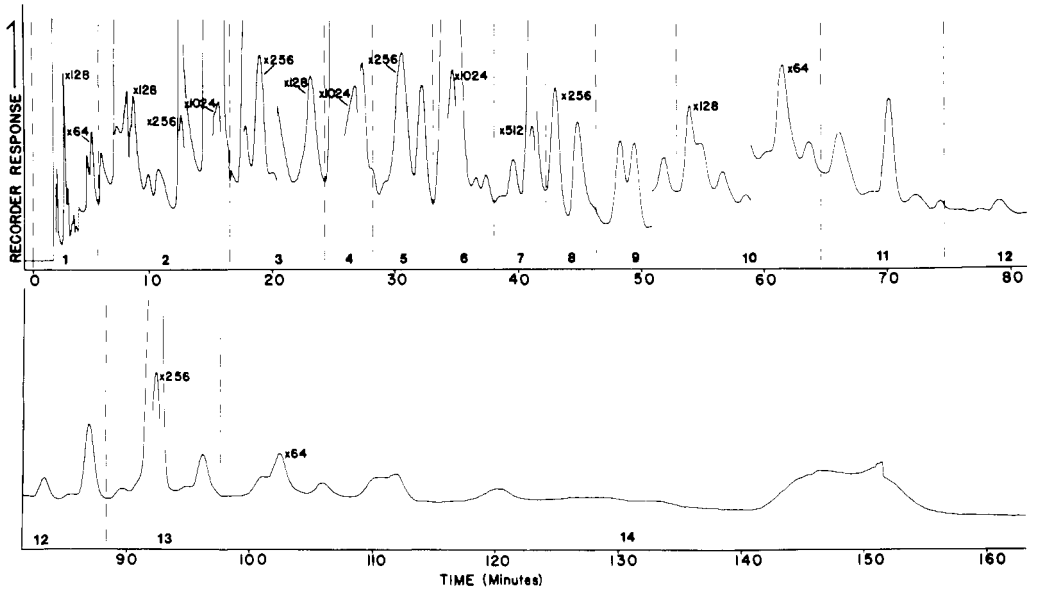

Figure 2. Chromatogram of ether extract of wild rice steam distillate on a 12 ft × 0.125 in. o.d. stainless steel packed silicone SE-30 column showing the 14 fractions collected for GC–MS analysis.

responsible for the stale flavor of stored rice (Yasumatsu et al., 1966a,b; Furuhasi and Ayano, 1971; Chikubu, 1970). In a study of feeding attractants for rats, Bullard and Holguin (1977) recently reported the identification of 73 volatile components in rice (*Oryza sativa L.*), 54 of which had never been previously identified in any unprocessed cereal grain. Rye volatiles have been studied by several workers (Neish, 1965; Hrklicka and Janicek, 1963; Collyer, 1964; Hampl et al., 1964) with von Sydow and Anjo (1969) identifying 92 components from rye crisp bread. Toasted oat flakes have received limited attention (Hrklicka and Janicek, 1963, 1964a,b).

The isolation and identification of some of the volatile constituents which contribute to the distinctive flavor of wild rice is described in this report.

EXPERIMENTAL SECTION

Sample Preparation. The Manomin strain wild rice used in this investigation was grown in Aitkin County, Minnesota, and was obtained from the Manomin Development Corp., Aitkin, Minn. The rice was fermented in piles 18 in. deep, watered daily at the rate of 1.5 gal/100 pounds, and turned on alternate days. Fermentation continued for 5 weeks before the seed was parched in a commercially operated semicylindrical, swept-surface barrel parcher heated by liquid propane (LP) gas-fired burners which imparted a toasted/smoky flavor to the wild rice. One-hundred-pound lots were continuously agitated by a paddle assembly and parched or dried at a bed temperature of 140–160 °F until the moisture was reduced to approximately 7% (Stuiber et al., 1972). Upon completion of the subsequent hulling and grading processes, the finished cereal was stored in vacuo in unlined 303 × 406 tinned steel cans at ambient temperature prior to analysis.

Isolation of Volatiles. Volatile flavor components were obtained by vacuum steam distillation in the apparatus shown in Figure 1. The sample flask was charged with 5 lb of wild rice and 6 L of saturated NaCl solution. Distillation was performed at 45 °C and 1–2 mmHg for a

Table I. Compounds Identified in Wild Rice (*Zizania aquatica*)

Compound	Retention index[a] Calcd	Retention index[a] Reference	Identification status
Alcohols			
Benzyl alcohol	11.88	12.20[d]	Positive
1-Propanol	3.96	3.71[d]	Positive
Aldehydes			
Benzaldehyde	8.45	8.49[b]	Positive
Cinnamaldehyde	13.62	14.00[e]	Positive
2-Pentenal	5.20	4.93[f]	Tentative
Furans			
2-Methyl-5-*n*-propylfuran	9.06		Tentative
2-Acetylfuran	8.26	8.70[d]	Positive
5-Methyl-2-acetylfuran	9.57	9.76[d]	Positive
2-Propionylfuran	9.16		Tentative
2-Furfural	7.86	7.96[b]	Positive
5-Methylfurfural	8.92	9.00[b]	Positive
Furfuryl alcohol	9.81	9.86[b]	Positive
Methyl 2-furoate	8.95	9.38[d]	Positive
2,5-Dimethyl-3(2*H*)-furanone	8.20	8.63[f]	Positive
Benzofuran	8.36	8.70[f]	Tentative
Dibenzofuran	15.59		Tentative
Ketones			
2-Cyclopentenone	7.11	7.30[d]	Positive
Cyclohexanone	6.17	6.76[e]	Tentative
2-Cyclohexenone	7.57	8.15[d]	Tentative
3-Methyl-2-cyclohexen-1-one	9.32	9.66[d]	Positive
Acetophenone	9.89	10.26[d]	Positive
Methylacetophenone	10.96	11.49[f]	Tentative
1-Phenyl-2-propanone	10.45	10.89[b]	Positive
Indan-1-one	13.08	12.95[b]	Positive
1,2,3,4-Tetrahydronaphthalen-1-one	16.91		Tentative
Lactones			
γ-Butyrolactone	9.20	9.77[e]	Positive
γ-Valerolactone	9.34	9.96[e]	Positive
2-Pyrone	10.75		Tentative
Phenols			
Phenol	13.20	13.38[c]	Positive
o-Cresol	13.45	13.08[c]	Positive
m-Cresol	13.92	13.98[c]	Positive
p-Cresol	14.14	14.11[c]	Positive
Dimethylphenol (2 isomers)	14.41/14.76		Tentative
3-Ethylphenol	14.89	15.05[c]	Positive
3-Ethyl-4-methylphenol	15.05		Tentative
3-Ethyl-5-methylphenol	15.21		Tentative
Vinylphenol	17.67	16.91[c]	Tentative
o-Methoxyphenol	12.12	12.20[d]	Positive
4-Ethyl-2-methoxyphenol	14.21	13.83[c]	Positive
2,6-Di-*tert*-butyl-*p*-cresol	12.58	12.65[e]	Positive
o,*o*′-Biphenol	14.89		Tentative
Pyrazines			
Pyrazine	5.67	5.77[d]	Positive
Acetylpyrazine	9.70	9.92[d]	Positive
Methylpyrazine	6.32	6.40[d]	Positive
Ethylpyrazine	6.73	7.06[d]	Positive
Vinylpyrazine	7.67	7.80[e]	Positive
2,3-Dimethylpyrazine	6.73	7.10[d]	Positive
2,5-Dimethylpyrazine	6.58	6.86[d]	Positive
2,6-Dimethylpyrazine	6.66	6.98[d]	Positive
Trimethylpyrazine	7.31	7.68[d]	Positive
2-Ethyl-3-methylpyrazine	7.23	7.57[d]	Positive
2-Ethyl-5-methylpyrazine	7.18	7.57[d]	Positive
2-Ethyl-6-methylpyrazine	7.15	7.53[d]	Positive
Tetramethylpyrazine	8.15	8.41[e]	Positive
2-Ethyl-3,6-dimethylpyrazine	7.90	8.10[d]	Positive
2-Ethyl-5,6-dimethylpyrazine	8.04	8.27[e]	Positive
2,5-Diethyl-3-methylpyrazine	8.38	8.70[d]	Positive
2-(2′-Furyl)-pyrazine	13.18	13.70[d]	Positive
2-(2′-Furyl)-5(6)-methylpyrazine	13.42		Tentative
6,7-Dihydro-5H-cyclopentapyrazine	9.74	10.25[d]	Tentative
2-Methyl-6,7-dihydro-5H-cyclopentapyrazine	10.41	10.73[d]	Positive
5-Methyl-6,7-dihydro-5H-cyclopentapyrazine	9.50	9.93[d]	Tentative
Methyl-6,7-dihydro-5*H*-cyclopentapyrazine (isomer)	9.83		Tentative
2-Ethyl-6,7-dihydro-5*H*-cyclopentapyrazine	11.21	11.42[d]	Positive
2,5-Dimethyl-6,7-dihydro-5*H*-cyclopentapyrazine	10.08	10.26[d]	Positive
3,5-Dimethyl-6,7-dihydro-5*H*-cyclopentapyrazine	10.16	10.93[d]	Positive
Pyridines			
Pyridine	6.05	6.00[b]	Positive

Table I (*Continued*)

Compound	Retention index[a] Calcd	Retention index[a] Reference	Identification status
2-Methylpyridine	5.67	5.78[e]	Positive
Methylpyridine (isomer)	5.35		Tentative
4-Ethylpyridine	5.80	7.75[b]	Tentative
Ethylpyridine (isomer)	6.74		Tentative
2,3-Dimethylpyridine	6.92	7.02[b]	Positive
Dimethylpyridine (isomer)	6.68		Tentative
2,3,6-Trimethylpyridine	7.39		Tentative
2,4,6-Trimethylpyridine	7.30	7.39[b]	Positive
Dimethylethylpyridine (3 isomers)	7.67/8.23/9.09		Tentative
4-Formylpyridine	7.02		Tentative
4-Methyl-2-pyridone	9.34		Tentative
1-Ethyl-2-pyridone	9.50		Tentative
Quinoline	14.58		Tentative
Pyrroles			
Pyrrole	8.63	8.73[e]	Positive
1-Acetylpyrrole	8.74	9.03[d]	Positive
Pyrrole-2-carboxaldehyde	13.38	13.50[d]	Positive
5-Methylpyrrole-2-carboxaldehyde	14.00	14.40[d]	Tentative
Methylindole	17.37		Tentative
Oxindole	10.65		Tentative
Miscellaneous			
Acenaphthene	14.24		Tentative
Benzimidazole	12.41		Tentative
Benzonitrile	9.17	9.70[d]	Tentative
Benzothiazole	12.42	12.48[b]	Positive
Benzyl cyanide	12.27	12.25[b]	Positive
Biphenyl	13.33	13.46[e]	Positive
Biphenylmethane	14.15		Tentative
Chloroform	3.79	3.66[d]	Positive
Dibutyl phthalate	20.46	20.47[b]	Tentative
Diethyl phthalate	16.45	16.80[d]	Positive
Dimethyl phthalate	15.62	16.32[f]	Tentative
1,2-Dimethoxy-4-methylbenzene	11.32	11.70[e]	Positive
Ethylcyclopentene	6.95		Tentative
Ethylnaphthalene	12.86	13.08[e]	Positive
1-Methylnaphthalene	11.88	11.92[c]	Positive
2-Methylnaphthalene	11.65	11.64[b]	Positive
Methylpyrazole	10.69		Tentative
Phenyl isocyanate	10.41		Tentative
3-*n*-Propyl-1-cyclopentene	7.69		Tentative
Styrene	6.15	6.32[d]	Positive
p-Toluidine	6.17		Tentative
Tolunitrile (two isomers)	10.16/12.26	12.80[e]	Positive

[a] Retention index (I_E) according to H. van den Dool and P. D. Kratz, *J. Chromatogr.* **11**, 463 (1963). [b] I_E value obtained on a 500 ft × 0.03 in. i.d. Carbowax 20M capillary column. [c] I_E value reported by Hruza et al., *J. Agric. Food Chem.* **22**, 123 (1974). [d] I_E value reported by Kinlin et al., *J. Agric. Food Chem.* **20**, 1021 (1972). [e] I_E value reported by Walradt et al., *J. Agric. Food Chem.* **19**, 972 (1971). [f] International Flavors and Fragrances, Inc., Union Beach, N.J., unpublished data.

period of 4.5 h. The aqueous distillate from the cold traps was combined and extracted with diethyl ether for 24 h in a continuous liquid–liquid extractor. The ether was removed by fractional distillation with final ether removal under a slow stream of nitrogen.

Aroma Evaluation of Volatile Distillate Components. Wild rice volatiles were analyzed on a Varian Model 1740 gas chromatograph (GLC) equipped with a flame ionization detector (FID), alkali flame ionization detector (AFID), and heated column effluent splitter assembly and exit port to permit an evaluation of the aroma of chromatographically resolved components by two to four persons and to give an indication of the presence of nitrogen-containing molecules. The GLC was equipped with a 500 ft × 0.03 in. i.d. stainless steel capillary column coated with Carbowax 20M which was operated at a flow rate of 20 mL of N_2/min and the temperature was programmed from 50 to 190 °C at 2 °C/min.

Identification of Volatile Components. Fractions from repeated injections of the volatile distillate were collected from a 12 ft × 0.125 in. o.d. stainless steel column packed with 10% SE-30 on 100–200 mesh Chromosorb G. The carrier gas flow rate was maintained at 25 mL of N_2/min, and the column was temperature programmed from 75 to 250 °C at 2 °C/min. The column effluent was split 1:10 between the FID and a heated exit port. For each separation, the effluent of the exit port was arbitrarily divided into 14 fractions which were trapped in $^1/_{16}$ in. o.d. glass capillary tubes cooled in dry ice. Corresponding fractions from several separations were combined prior to analysis by gas chromatography–mass spectrometry (GC–MS). The GC–MS equipment was a DuPont Model 21-491 double-focusing instrument interfaced to a Varian Model 2700 gas chromatograph via a single-stage jet separator. The GLC was equipped with an AFID and the mass spectrometer was equipped with a total ion monitor (TIM). The responses from these two detectors were recorded on a Varian Model A-25 dual-channel recorder. Mass spectra were obtained, whenever there was an indication of a peak on either the AFID or the TIM, on a DuPont Type 5-124 recording oscillograph. The Carbowax 20M capillary column and conditions described above were used for these analyses with helium as the carrier gas.

Gas chromatographic retention indices (I_E values) were

Table II. Aroma Profile Descriptors from a GC Separation of a Wild Rice Steam Distillate Extract and Corresponding Retention Indices (I_E) on a Carbowax-20M Capillary Column

Retention index	Aroma descriptor	Retention index	Aroma descriptor
2.62	Breadcrust, buttery	7.37	Raw potato-strong
3.24	Toasted-pleasant	7.67	Licorice
3.66	Roasted-pleasant	7.70	Cooked potatoes
4.16	Planty, vegetable	7.91	Musty, green
4.24	Green plant	8.02	Earthy, potato
4.66	Unpleasant, aldehyde	8.07	Aldehyde
4.84	Brothy	8.20	Alkyl pyrazine
4.95	Buttery	8.28	Burned
5.03	Paint-like	8.43	Crackers
5.14	Pyridine-like	8.58	Milkweed
5.26	Unpleasant, N-heterocyclic	8.77	Muskmelon, cucumber
5.81	Pyrazine, roasted	8.83	Pleasant, sweet
5.98	Alkyl pyrazine	8.98	Aldehyde-like
6.13	Sulfury, earthy	9.14	Burned, milkweed
6.21	Earthy, pyrazine	9.31	Musty, moldy-strong
6.42	Pyrazine (acetyl-)	9.49	Crackers-pleasant
6.58	Alkyl pyrazine	9.58	Brothy
6.71	N-Heterocyclic	9.70	Floral
7.02	Chili pepper	9.81	Green plant
7.23	Alkyl pyrazine	9.86	Brothy-pleasant
9.95	Musty-heavy	12.66	Alfalfa hay
10.06	Horsey	12.78	Sweet, candy
10.13	Unpleasant	12.89	Chocolate
10.28	Baked, sweet	13.06	Smoky-strong
10.35	Benzothiazole-like	13.19	White glue
10.45	Wild rice, smoky	13.30	Spicy, cinnamon drops
10.59	Phenolic	13.47	Medicinal
10.71	Barley, steamrolled	13.86	Celery
10.77	Grainy	13.91	Horse urine
10.83	Mousey	14.13	Camphoric
10.90	Cinnamon, musty	14.46	Peachy
11.02	Phenolic	14.75	Smoky, medicinal
11.06	Crackers	14.89	Smoky-pleasant
11.23	Horsey, planty	15.16	Spicy-pleasant
11.38	Pleasant	15.41	Resinous
11.46	Musty grain	15.73	Crushed celery
11.58	Lactone-like	15.84	Burned
11.65	Scorched wood	16.00	Brothy-heavy
11.72	Guaiacol-like	16.10	Eugenol-like
11.82	Spicy, whiskey	16.34	Vegetable, cloves
12.19	Floral, honey	16.43	Tanned leather
12.34	Stale cigarettes	16.57	Musty, dirty
12.56	Smoky	16.81	Green plant-sharp
16.90	Naphthalene-like		
17.34	Naphthalene-like		
17.89	Smoky, wild rice		
17.99	Planty-rich		
18.20	Phenolic, medicinal		
18.31	Planty-rich		
18.36	Mothballs, smoky		
18.46	Hot cedar wood		
18.92	Planty		
19.22	Smoky		
19.43	Wood smoke, campfire		
20.38	Smoky		
21.29	Sweet, guaiacol-like		
22.61	Sweet, cough drops		

determined by the method of van den Dool and Kratz (1963), relative to aliphatic ethyl esters for all analytical work to assist in data correlation and confirmation of identifications. To obtain retention indices from GC–MS data, I_E values were extrapolated for each of the components using 2-methylnaphthalene (I_E = 11.64), occurring in each of the fractions, as a secondary reference point. Reference retention indices were obtained from either chemically pure reference chemicals or literature reports of indices obtained in a similar manner (Walradt et al., 1971; Kinlin et al., 1972; Hruza et al., 1974).

RESULTS AND DISCUSSION

A typical chromatogram of the preparative SE-30 separations and the positions of the 14 fractions collected for GC–MS analysis is shown in Figure 2. Fractionation of the distillate was performed in order to simplify the analysis and to aid in resolving components which co-chromatograph on a single column. The compounds identified from the wild rice volatile fractions are listed in Table I. The use of the AFID in conjunction with the TIM assisted in confirming the presence of nitrogen-containing components in mixed spectra and, due to its greater sensitivity, aided in detecting trace nitrogenous components present below detection levels of the TIM.

Positive identifications were based upon the agreement of mass spectral fragmentation patterns and I_E values of authentic compounds or published data in addition to the appropriate detector response. Tentative identifications were based upon mass spectral fragmentation and detector

response only because I_E values were unavailable in the laboratory.

The volatile isolate obtained by vacuum distillation exhibited the smoky, toasted aroma characteristic of the original wild rice. These aroma characteristics were quite apparent in this sample because of the particular parching process utilized. Some current wild rice processes result in less-severe thermal exposure, thereby producing a light toasted or tea-like character. The descriptors from the aromagram of the total isolate are listed in Table II with their corresponding I_E values. Many of the descriptors can be associated with the thermal process, including the smoky character contributed by the phenols and the toasted and roasted aromas associated with certain pyrazines. The alkyl pyridines identified probably contribute to the tea-like and green aromas of wild rice, as they have been encountered in other foods such as black tea (Vitzthum et al., 1975), roasted filberts (Kinlin et al., 1972), and roasted peanuts (Walradt et al., 1971). Although correlation of aroma profile with component identification was hampered by the complexity of the total sample from which the profile was obtained, certain assignments could be made. The pleasant, sweet note at I_E 8.83 was associated with 5-methylfurfural identified at I_E 8.92. Acetophenone was responsible for the strong floral aroma at I_E 9.70. Phenol was identified as a major component of the sample and contributed to the "smoky" note observed at I_E 13.06 on the aromagram. Cinnamaldehyde contributed the "cinnamon drop" aroma at I_E 13.30. "Smoky", "phenolic", and "medicinal" aromas were the predominant notes observed after I_E 11.00. A number of "musty", "dirty", "earthy", and "horsey" aromas were noted but could not be attributed to specific chemical components identified in this study.

The compounds identified in wild rice sharply contrast those identified in unprocessed domestic white rice (Bullard and Holguin, 1977). The latter is characterized by a predominance of alkylbenzenes, aldehydes, and 2-alkanones whereas the former achieves a portion of its uniqúe sensory quality through the presence of the pyrazines, furans, and phenolic constituents developed during thermal processing. Similarly, the classes of compounds identified from wheat (McWilliams and Mackey, 1969) differ from those found in baked white bread (Mulders, 1973a, b; Mulders and Dhont, 1972; Mulders et al., 1972; Mulders et al., 1973). Roasted barley (Shimizu et al., 1967, 1970a, b; Wang et al., 1968, 1969; Collins, 1971) and rye crisp bread (von Sydow and Anjou, 1969) are also characterized by an abundance of flavor chemicals attributed to thermal processes that are not found in the unprocessed kernel.

The true nature of wild rice flavor probably involves contributions from many of the 112 components identified in this study plus numerous other components yet to be identified. The various sensory impacts which processed wild rice elicit are the result of a blend of these compounds whose concentrations are determined by both the available precursor levels and the reaction or processing conditions to which they are exposed.

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Volatile Constituents of Cinnamon (*Cinnamomum zeylanicum*) Oils

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Analysis of *Cinnamomum zeylanicum* leaf, stem bark, and root bark oils indicated 72 compounds, of which 32 have not been reported before in cinnamon oils. All three oils had a similar array of compounds but in varying proportions. Of the new compounds reported there were 11 monoterpenes, 4 sesquiterpenes, 2 aliphatic, and 15 aromatic compounds.

Cinnamomum zeylanicum, the cinnamon of commerce, provides various types of oils depending on which part of the plant is utilized. Oils derived from the leaf, stem bark, and root bark have commercial usage. The most complete studies available on the volatile constituents of cinnamon oils are those by Angmor et al. (1972) and Wijesekera et al. (1974). A total of 41 compounds have been identified with the major component of cinnamon leaf oil being eugenol (about 70% of total volatiles) and cinnamaldehyde the major component of stem bark oil (about 75% of total).

In this paper we examined cinnamon oils in more detail by subfractionation of each oil and GC analysis of the fractions on a SCOT column and report the presence of a substantial number of additional compounds.

EXPERIMENTAL SECTION

Isolation of Volatiles. Samples of commercial cinnamon leaf and bark oils were obtained from Bush Boake and Allen Ltd, London. Oil was also obtained by steam distillation of cinnamon leaf, stem bark, and root bark (30-g lots) in a specially designed all-glass apparatus (Senanayake et al., 1978). The volatiles were trapped in a layer of pentane–ether (0.5 mL), dried over anhydrous sodium sulfate, and concentrated over a gentle stream of nitrogen. In order to facilitate the separation and identification of the minor constituents, the major compounds of leaf oil (eugenol) and stem bark oil (cinnamaldehyde) were removed. Cinnamon root bark oil was analyzed without fractionation.

To remove eugenol, leaf oil (2 mL) was dissolved in diethyl either (5 mL) and shaken with 10% potassium hydroxide solution (3 × 5 mL). The ether layer was removed, washed with distilled water (5 mL), and dried over anhydrous sodium sulfate. Excess ether was evaporated with nitrogen gas. The noneugenol fraction was separated into hydrocarbon and oxygenated fractions by column chromatography, as described by Stahl and Jork (1969) with minor modifications. A sample was loaded onto a column of deactivated silica gel (12.0 × 1.0 cm) and eluted with *n*-pentane to remove the hydrocarbons, followed by diethyl ether to remove oxygenated compounds (Hedin et al., 1975).

To remove cinnamaldehyde, stem bark oil (1 mL) was shaken with *n*-pentane (1 mL). Two layers formed, with cinnamaldehyde the bottom layer. The top layer of pentane was relatively free of cinnamaldehyde, but contained the other constituents of stem bark oil. Chromatograms of the bottom layer showed mainly cinnamaldehyde and traces of other major peaks. The pentane layer was treated as described for the noneugenol fraction of cinnamon leaf oil, to separate the hydrocarbon and oxygenated fractions.

Gas Chromatography. Oil samples (1 μL) were analyzed by flame ionization gas chromatography using a high-performance glass SCOT column (70 m × 0.5 mm i.d.) with Carbowax 20M (S.G.E., Melbourne) fitted with a pre-column stream splitter (30:1). The operating conditions were: high-purity nitrogen, 3 mL/min; hydrogen, 25 mL/min; and air, 300 mL/min; injector temperature, 200 °C; detector temperature, 235 °C. The relative abundance of each compound in the oils was calculated by a digital integrator. Substantial preliminary analyses of the oils were made on 3 m × 3.2 mm o.d. stainless steel columns packed with either 10% Carbowax 20M, 15% LAC-2R-446 or 10% SE-30 on 80–100 mesh Chromosorb W.

For preparative analyses, a glass column (3 m × 6.4 mm o.d.) packed with 20% Carbowax 20M on 60–80 mesh Gas Chrom Q was used. The emergence of each compound from the column was monitored in a trial run, and in a subsequent run fractions were collected for infrared analysis according to the method of Edwards and Fagerson (1965), where the fractions were trapped in 3.8-cm long hypodermic needles cooled by dry ice. The trapped fractions were transferred to an ultra-microcavity cell (type D, 0.5 mm path, Research and Industrial Instruments, London) in carbon tetrachloride with a syringe. All spectra were recorded with a Hilger and Watt Infrascan IR

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