al. (1976) to be highly variable. Cereals C and D fortified with sodium iron pyrophosphate (SIP) were products marketed by another manufacturer. The RBV for D was slightly higher than that for C. Infant cereals E, J, and K containing SIP were produced by the same manufacturer, but since E was bought much earlier than J and K, the source of SIP could have been different. Interestingly the RBV of iron from these cereals varied from 23 to 63%, indicating that the SIP used was probably not of the same quality. The bioavailability of iron from infant cereals F and G (manufacturer 5) was comparable to that of iron from cereal E (manufacturer 3). All these cereals fortified with SIP were poor sources of iron. However, all forms of SIP were not of poor quality. The SIP in product J was probably the best and that in K was of intermediate quality.

The differences in the bioavailability of iron from ferric orthophosphate and from SIP could be due to variation in the chemical composition of the specimens, the shape and size of the particles, or their solubility in dilute hydrochloric acid or gastric or intestinal juices (British Ministry of Health, 1968; Fritz et al., 1975; Harrison et al., 1976; Coccodrilli et al., 1976). The relationship between these parameters and the bioavailability of iron from iron phosphates needs further investigation. As in the case of iron powders (Shah et al., 1977), there is a need to develop an in vitro chemical test which can distinguish between an acceptable and a poor quality ferric orthophosphate or sodium iron pyrophosphate.

Processing has been reported to increase the bioavailability of iron from ferric pyrophosphate added to liquid infant formula products (Theuer et al., 1971) and of iron from ferric orthophosphate added to weight control dietaries (Hodson, 1970). However, the processing involved in the manufacture of cereal products does not involve conditions which would be conducive to promoting the reduction of ferric ion to the ferrous state (Steele, 1976). Iron source additives and vitamins are usually added at a stage beyond which the product is subject to minimum heat treatment. This is confirmed by the RBV of SIP in breakfast cereals N and O. In the case of N the effects of processing were present but the RBV of iron from SIP was not affected. Thus it was evident that the particular specimen of SIP used in these cereals was of a very good quality.

The bioavailability of iron added to soy-based infant formulas L and M was higher than all the cereal products assayed except the experimental cereals N and O. Most probably this improvement in availability was the result of processing, as reported by Theuer et al. (1971), in the case of ferric pyrophosphate.

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# Volatile Constituents of *Castanopsis* Flower

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The volatile constituents of *Castanopsis* flower (*C. caspidata* Schottky var. Sieboldii Nakai), which had not been studied prior to this report, have been investigated by gas chromatography–mass spectrometry. Fifty-six compounds were identified in the oil which was obtained from this flower by the distillation–extraction method (Likens and Nickerson apparatus). The compounds reported here were 11 monoterpenes, 4 sesquiterpenes, 21 aliphatic compounds, 12 aromatic compounds, and 7 heterocyclic compounds. Major compounds of this oil were salicylaldehyde, *o*-aminoacetophenone, methyl salicylate, linalool, and nonanal.

*Castanopsis caspidata* Schottky var. *Sieboldi Nakai* (*Sudajii* in Japanese) grows wild over central and southern Japan and is commonly found in the forest. Some grow to heights of 20 m. The flower (spike form) blooms from May to June, and the plant bears nuts which are edible in autumn. This plant is a member of Fagaceae family which includes *Castanea* (chestnut tree). The flowers of *Castanopsis* and *Castanea* have similar characteristic odors, and recently those odors have received much attention as they are used to give an animal note to fragrances.

In this study, the volatile constituents of *Castanopsis*

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Table I. Compounds Identified in the Oil of *Castanopsis* Flower and Their Kovats Indices

| peak no. CW 20M | compound | peak area % | Kovats indices: Carbowax 20M $I_u$ | Carbowax 20M $I_k$ | OV-101 $I_u$ | OV-101 $I_k$ | peak no. OV-101 |
|---|---|---|---|---|---|---|---|
| 1 | octane | 0.16 | 800 | 800 | 800 | 800 | 7 |
| 2 | tetrahydrofuran | 0.21 | 866 | 867 | 609 | 610 | 2 |
| 3 | solvent (methylene chloride) | | | | | | 1 |
| 4 | pentanal | *a* | 950 | 951 | 675 | 678 | 3 |
| 5 | hexanal | 0.14 | 1056 | 1053 | 778 | 780 | 6 |
| 6 | *trans*-2-hexenal | | 1095 | 1092 | 829 | 832 | 9 |
| 7 | myrcene | | 1140 | 1137 | 986 | 987 | 18 |
| 8 | pyridine | | 1147 | 1143 | 695 | 696 | 4 |
| 9 | heptanal | | 1154 | 1151 | 889 | 883 | 13 |
| 10 | *d*-limonene | 0.12 | 1180 | 1181 | 1031 | 1030 | 20[c] |
| 11 | 3-ocimene (tentative) | 0.12 | 1219 | | 1030 | | 20[c] |
| 12 | octanal | 0.12 | 1247 | 1248 | 984 | 985 | 17 |
| 13 | alloocimene (tentative) | 0.10 | 1254 | | | | b |
| 14 | 1-hexanol | 0.35 | 1289 | 1290 | 858 | 858 | 12 |
| 15 | *cis*-3-hexen-1-ol | 4.5 | 1322 | 1322 | 849 | 847 | 10 |
| 16 | *trans*-2-hexen-1-ol | | 1341 | 1340 | 856 | 856 | 11[c] |
| 17 | nonanal | 3.6 | 1350 | 1348 | 1088 | 1087 | 27 |
| 18 | *trans*-linalool oxide | 0.13 | 1383 | 1383 | 1067 | 1069 | 24 |
| 19 | furfural | 1.2 | 1393 | 1394 | 815 | 815 | 8 |
| 20 | *cis*-linalool oxide | | 1409 | 1410 | 1081 | 1082 | 26 |
| 21 | 2-acetylfuran | | 1434 | 1434 | 893 | 893 | 14 |
| 22 | decanal | | 1446 | 1448 | 1192 | 1188 | 35 |
| 23 | benzaldehyde | | 1456 | 1456 | 942 | 946 | 16 |
| 24 | linalool | 4.6 | 1478 | 1479 | 1095 | 1092 | 28 |
| 25 | 1-octanol | 0.18 | 1487 | 1490 | 1065 | 1061 | 23 |
| 26 | 5-methylfurfural | | 1491 | 1495 | 941 | 942 | 15 |
| 27 | unknown | 0.15 | 1534 | | 1169 | | 32 |
| 28 | *a*-bergamotene | 0.13 | 1552 | 1554 | 1224 | 1226 | 37 |
| 29 | phenylacetaldehyde | 0.33 | 1564 | 1563 | 1022 | 1024 | 19 |
| 30[c] | furfuryl alcohol | 0.47 | 1571 | 1572 | 857 | 858 | 11[c] |
| 30[c] | acetophenone | | 1571 | 1575 | 1041 | 1044 | 21 |
| 31 | 1-nonanol | 0.55 | 1587 | 1586 | 1163 | 1161 | 31 |
| 32 | salicylaldehyde | 29.7 | 1597 | 1598 | 1031 | 1029 | 20[c] |
| 33 | *a*-terpineol | 0.92 | 1617 | 1621 | 1184 | 1185 | 34 |
| 34 | *β*-farnesene (tentative) | | 1669 | | 1432 | | 45 |
| 35 | methyl salicylate | 4.1 | 1692 | 1694 | 1180 | 1181 | 33 |
| 36 | *a*-farnesene (tentative) | 0.33 | 1695 | | 1495 | | 46 |
| 37 | *o*-hydroxyacetophenone | 0.16 | 1709 | 1714 | 1140 | 1144 | 30 |
| 38 | nerol | 0.22 | 1715 | 1717 | 1218 | 1220 | 36 |
| 39 | ethyl salicylate | 0.19 | 1727 | 1730 | 1254 | 1257 | 39 |
| 40 | unknown | 0.24 | 1745 | | 1361 | | 44 |
| 41 | guiacol | | 1754 | 1760 | 1070 | 1071 | 25 |
| 42 | geraniol | 1.3 | 1759 | 1763 | 1244 | 1243 | 38 |
| 43 | benzyl alcohol | 0.43 | 1772 | 1774 | 1031 | 1033 | 20[c] |
| 44 | phenylethyl alcohol | 1.8 | 1807 | 1807 | 1105 | 1104 | 29 |
| 45 | unknown | 0.23 | 1880 | | | | *b* |
| 46 | nonadecane | 0.43 | 1900 | 1900 | 1900 | 1900 | 52 |
| 47 | 1,8-menthadien-9-ol (tentative) | | 1927 | | | | *b* |
| 48 | nerolidol | 0.76 | 1951 | 1954 | 1551 | 1550 | 48 |
| 49 | eicosane | 0.36 | 2000 | 2000 | 2000 | 2000 | 55 |
| 50 | eugenol | | 2039 | 2040 | 1347 | 1347 | 43 |
| 51 | nonanoic acid | 3.3 | 2049 | 2047 | 1302 | 1303 | 41 |
| 52 | unknown | 0.38 | 2066 | | 1308 | | 42 |
| 53 | *o*-aminoacetophenone | 8.0 | 2085 | 2091 | 1285 | 1288 | 40 |
| 54 | heneicosane | 15.2 | 2100 | 2100 | 2100 | 2100 | 57 |
| 55 | methyl palmitate | 1.5 | 2143 | 2149 | 1918 | 1917 | 53 |
| 56 | unknown | 1.6 | 2189 | | 1938 | | 54 |
| 57 | docosane | 0.21 | 2200 | 2200 | 2200 | 2200 | 59 |
| 58 | unknown | 2.4 | 2252 | | 1709 | | 50 |
| 59 | 4-methoxyquinoline (tentative) | 0.54 | 2294 | | 1506 | | 47 |
| 60 | tricosane | 4.0 | 2300 | 2300 | 2300 | 2300 | 60 |
| 61 | tetracosane | 0.22 | 2400 | 2400 | 2400 | 2400 | 61 |
| 62 | pentacosane | 2.8 | 2500 | 2500 | 2500 | 2500 | 62 |

[a] Area % less than 0.1. [b] Not found in OV-101 chromatogram. [c] More than two compounds in one peak.

flower were isolated and identified by means of gas chromatography–mass spectrometry.

## EXPERIMENTAL SECTION

Taxonomic identification of this plant was made by professor Masao Arai, Tokyo Agricultural University, Tokyo, Japan.

*Castanopsis* flowers were collected at Akabane, Tokyo, during May, 1978. The spikes (500 g), about 10 cm in length, were subjected to simultaneous distillation and extraction (SDE) (Schultz et al., 1977), a modification of the method of Liken and Nickerson (1964). The extracting solvent was methylene chloride and steam distillation–extraction ($CH_2Cl_2/H_2O$ = 200 mL/1500 mL) was con-

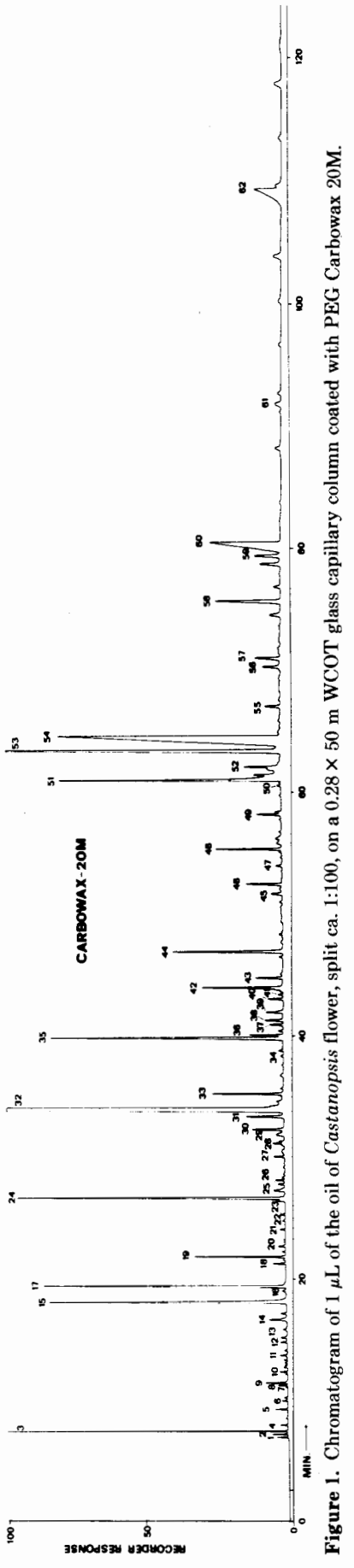


**Figure 1.** Chromatogram of 1 μL of the oil of *Castanopsis* flower, split ca. 1:100, on a 0.28 × 50 m WCOT glass capillary column coated with PEG Carbowax 20M.

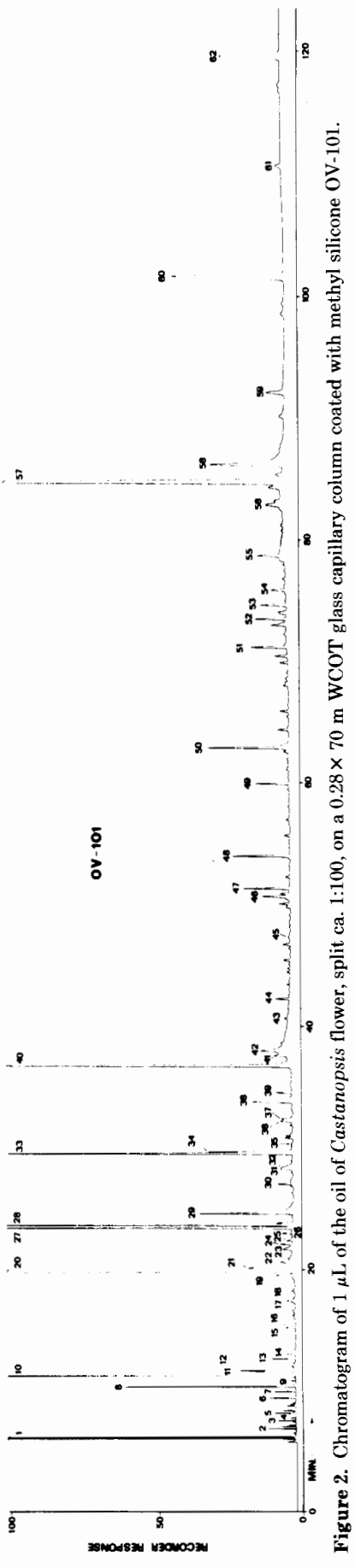


**Figure 2.** Chromatogram of 1 μL of the oil of *Castanopsis* flower, split ca. 1:100, on a 0.28 × 70 m WCOT glass capillary column coated with methyl silicone OV-101.

tinued 6 h. The extract was dried over anhydrous sodium sulfate for 12 h and the solvent was removed using a rotary flash evaporator. Approximately 0.16 g (0.023% relative to the quantity of spikes used) of volatile oil including residue of solvent (gas chromatographic peak area percent of solvent is 2.9) was obtained.

The oil was subjected to gas chromatographic analysis in wall-coated open-tubular glass capillary columns (WCOT) coupled to a splitter of our own design. Identifications were based on mass spectra obtained on a gas chromatograph–mass spectrometer interfaced with a glass capillary column (24.5 cm × 1.0 mm i.d.) and confirmed by retention data (Kovats, 1958; Ettre, 1964) on this high-resolution system. Two types of WCOT glass capillary column were used in these investigations: 50 m × 0.28 mm i.d. coated with Carbowax 20M and 70 m × 0.28 m i.d. coated with OV-101. The columns were prepared in our own laboratory using the heated inlet tube modification of the Golay technique (Jennings et al., 1974). All columns possessed over 4000 theoretical plates per meter at $K = 3$. A Hewlett-Packard Model 5710A gas chromatograph equipped with a flame ionization detector (FID) modified for capillary analyses was used for Carbowax 20M column analysis. Column temperature was programmed from 80 to 200 °C at 2 °C/min. Columns were operated with nitrogen carrier gas at an average linear velocity of 13 cm/s based on a methane peak at 120 °C. This was equivalent to a flow rate of ca. 0.68 $cm^3$/min. A Varian Model 2800 gas chromatograph equipped with FID modified for capillary analysis was used for OV-101 column analysis. Column temperature was programmed from 80 to 240 °C at 2 °C/min. Columns were operated with nitrogen carrier gas at an average linear velocity of 17 cm/s based on a methane peak at 120 °C. This was equivalent to a flow rate of ca. 0.90 $cm^3$/min.

A Hewlett-Packard Model 3385-A reporting integrator was used to determine the peak areas reported in Table I.

A Hitachi Model RMU-6M combination mass spectrometer–gas chromatograph equipped with a Hitachi Model 002B Datalyzer was used for mass spectral identification of the gas chromatographic components under the following conditions: ion source, 200 °C; ion acceleration, 3.1 kV; electron energy, 70 eV.

## RESULTS AND DISCUSSION

Table I shows the compounds identified in the oil of *Castanopsis* (*C. caspidata* Schottky var. *Sieboldii Nakai*) flower. Peak numbers on the left side show the elution order on the Carbowax 20M column (Figure 1); peak numbers on the right side show the elution order on the OV-101 column (Figure 2). Those peak areas which had value of less than 0.1% are not listed. $I_u$ designates retention indices of unknowns. $I_k$ represents the retention indices of authentic samples. For some compounds, formulae were deduced from mass spectral data, but known compounds were not available. We listed those compounds as "tentatively" identified.

More volatile compounds are separated more efficiently by Carbowax 20M than by OV-101; for example, peaks (on Carbowax 20M) 10, 11, 16, 30, 32, and 43 were not separated by the OV-101 chromatogram. On the other hand, peaks 5, 22, 51, 56, and 58 on OV-101 (Figure 2) were not found on Carbowax 20M chromatogram (these all remained unidentified and were not listed up in Table I). Furfuryl alcohol and acetophenone were not separated by Carbowax 20M but separated by OV-101.

Many aliphatic alcohols, aldehydes, and acids were identified: *cis*-3-hexen-1-ol, nonanal, 1-nonanol, and nonanoic acid. Each of the compounds possess a strong and characteristic odor. The odor of this flower is characterized by these compounds. The quantity of terpenes was less than other odor giving compounds. Acetophenone and its derivatives (*o*-aminoacetophenone and *o*-hydroxyacetophenone) were found in this oil. *o*-Aminoacetophenone had been found in Bear (Palamand and Grigsby, 1974). These compounds have never been, however, found in essental oils previously. The major component of this oil was salicylaldehyde (area %, 29.7) and has been found in the essential oil (ter Heide, 1972). This has a benzaldehyde like odor. Salicylaldehyde and *o*-aminoacetophenone contribute the somewhat floral odor of this flower. Many hydrocarbons, which appear in the high-temperature range on the gas chromatograms, were also identified. These hydrocarbons may derive from wax components of this flower. Some constituents which give a burnt odor may be artifacts produced by heating during distillation (furfural derivatives).

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