Flavor Components in Vietnamese Green Tea and Lotus Tea†

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The aroma concentrates from Vietnamese green tea and lotus tea were prepared and analyzed. Characterization of the components were carried out using coupled gas chromatography and mass spectrometry and infrared spectrometry, besides gas chromatographic retention data.

Anethole and 1,4-dimethoxybenzene have been identified for the first time as the flavor constituents in green tea. Linalool, two linalooloxides (*cis* and *trans*, five membered), 3,7-dimethyl-1,5,7-octatriene-3-ol, 2,5 (or 2,6)-dimethylpyrazine and 1-ethyl-2-formylpyrrole were the predominant components in Vietnamese green tea.

1,4-Dimethoxybenzene has been identified as the main component in lotus tea. The compound was also isolated from both dried and fresh lotus pollen.

Tea is an important crop in Vietnam, next to rice and rubber. Tea plant grown in Vietnam belongs to the family of *Theaceae*, originated from Shan district. Tea scented with lotus pollen, called lotus tea, is purely Vietnamese special tea and it is the best liked and the most expensive in Vietnam.

This paper reports on the flavor analysis of 1) Bach-mao tea (the highest class of Vietnamese green tea manufactured by pan-firing process) and 2) lotus tea.

EXPERIMENTAL

Materials

1) Bach-mao tea was manufactured by pan-firing processing, so called "tra-xao" process, from summer leaves at Nam-Sanh tea company, located on Blao in South Vietnam.

2) Lotus tea was manufactured by private order. It was scented with natural lotus pollen (600 flowers were used for 1 kg of green tea).

3) Dry lotus pollen, which was separated from lotus tea.

4) Fresh lotus pollen. Lotus flower plucked at the ponds in suburb of Saigon on August 29, 1973, and stored at -20°C in our laboratory. Pollen were separated from flower just before distillation.

Preparation of aroma concentrate

Each 200 g of green tea was mechanically powdered

and immediately mixed with 500 ml of hot water (about 60°C) in the 2 liters flask of rotary evaporator connected with a condenser and two traps as shown in the previous paper.¹⁾ Distillation was conducted at 50°C (water bath temperature) under reduced pressure at 18 to 20 mmHg. When the material in the flask almost dried, another two portions (500 ml of hot water) were added and distillation was repeated. Thus, about 1.2 liters of distillate was obtained. The distillate in trap A (cooled by ice-salt mixture) was combined with the condensate in trap B (cooled by dryice-acetone mixture), saturated with sodium chloride and extracted with peroxide-free ether. After drying the mixture with sodium sulfate, the ether extract was concentrated by distilling off the ether at 38 to 40°C . The pale yellow oil which has an intense typical aroma of tea infusion was obtained.

The procedure was repeated on another 800 g of Bach-mao tea. Total yield of the aroma concentrate (I) was 2.6 g (0.26%). The distillation for lotus tea was carried out by the same procedure as above. From 800 g of lotus tea 2.58 g (0.3%) of the aroma concentrate (II) was obtained. This aroma concentrate contained some crystals.

Analysis of the aroma concentrate. Method of identification

Gas Chromatography (GC). Japan Electron Optics Laboratory Co., Ltd.—Model 810 Gas Chromatograph with a thermal conductivity detector (TCD) was utilized for separation. Experimental parameters are written in each figure of the chromatogram.

Infrared analysis (IR). IR spectra were recorded on a Japan Spectroscopic Co., Model IR-S, Infrared Spectrometer and analysed with either a film of the pure

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sample or with nujol.

Mass spectral analysis (MS). A Hitachi RMS-4 Mass Spectrometer, combined directly with gas chromatograph Hitachi K53 with a Bieman-type helium separator was used. Experimental parameters are written in each figure of the chromatogram recorded by total ion monitor.

Compounds were identified by comparing their spectra with those of authentic compounds or with published standard spectra, checking retention time agreements.

RESULTS AND DISCUSSION

1) *Bach-Mao tea*

Figure 1 shows the gas chromatogram of aroma concentrate (I) on Golay column (Carbowax 20 M) recorded by total ion monitor in GC-MS analysis. Identified compounds

are listed in Table I.

Gas chromatography was also run on packed column and recorded by TCD as well as total ion monitor in GC-MS analysis. The chromatogram recorded by TCD is shown in Fig. 2. Identified components and their compositions are listed in Table II.

Anethole and 1,4-dimethoxybenzene have never been found as the components of the flavor up to date. The two compounds seemed to be specific to Vietnamese tea (variety: Shan).

2) *Lotus tea*

Aroma concentrate (II), from lotus tea was separated into four traps by gas chromatography using column Carbowax 6000 as shown in Fig. 3-1.

FIG. 1. GC-MS Chromatogram of Bach-Mao Tea Aroma Concentrate.

Column: Golay column, Carbowax 20 M, 0.5 mm (*i.d.*) $\times$ 45 m. Injection temp.: 200°C, Helium flow rate: 3 ml/min. Column temp.: 40°C $\xrightarrow{1^\circ/\text{min}}$ 60°C $\xrightarrow{2^\circ/\text{min}}$ 170°C (hold 11 min) $\xrightarrow{2^\circ/\text{min}}$ 190°C. Detector: Total ion monitor. Helium separator temp.: 200°C, Ionization source temp.: 200°C, Electron energy: 70 eV.

TABLE I. IDENTIFIED COMPONENTS IN BACH-MAO TEA VOLATILES
(Reference to Fig. 1)

Peak No.	Identified comp.	Mass spectra ^{a)} <i>m/e</i> (%)
5	2,5 or 2,6-Dimethyl pyrazine	42 (100), 108 (65), 39 (28), 40 (18)
6	2-Methyl-5-ethyl pyrazine	121 (100), 43 (72), 40 (70), 122 (61), 39 (42)
7	<i>cis</i> -Linalooloxide (five membered)	59 (100), 68 (35), 146 (29), 67 (26), 93 (26)
8	<i>trans</i> -Linalooloxide (five-membered)	59 (100), 94 (40), 68 (36), 135 (33), 93 (28)
9	2-Acetylfuran	95 (100), 110 (36), 39 (30), 43 (25), 96 (6)
11	Linalool	71 (100), 93 (73), 41 (71), 55 (57), 69 (47)
13	1-Ethyl-2-formyl pyrrole and 3,7-Dimethyl-1,5,7-octatriene-3-ol	123 (100), 94 (82), 122 (45), 108 (42), 106 (27) 71 (100), 82 (60), 67 (41), 41 (35), 43 (33)
14	1-Ethyl-2-acetyl pyrrole	122 (100), 94 (58), 137 (55), 66 (28), 80 (10)
16	Linalooloxides (<i>cis</i> and <i>trans</i> six membered)	59 (100), 93 (83), 121 (31), 131 (31)
17	1,4-Dimethoxybenzene	123 (100), 138 (66), 95 (41), 52 (21)
21	Anethole	149 (100), 147 (46), 133 (31), 117 (27), 121 (19)
22	Geranyl acetate	69 (100), 41 (88), 43 (79), 57 (50)
23	Ionol (BHT)	57 (100), 41 (61), 69 (50), 145 (33)
24	β -Ionone	177 (100), 43 (59), 41 (44), 91 (37)
25	2-Acetyl pyrrole	94 (100), 109 (69), 66 (59), 39 (34)

^{a)} Electron energy: 70 eV. Scanning speed: 6 sec.

FIG. 2. Gas Chromatogram of Bach-Mao Tea Aroma Concentrate.

Column: 3 mm (*i.d.*) $\times$ 300 cm stainless steel. Column packing: 23% Carbowax 6000 on Shimalite W 60/80. Injection temp.: 180°C, Helium flow rate: 30 ml/min. Column temp.: 40°C $\xrightarrow{2^\circ/\text{min}}$ 180°C. Detector: T.C.D.

TABLE II. IDENTIFIED COMPONENTS AND THEIR COMPOSITIONS
IN BACH-MAO TEA VOLATILES
(Reference to Fig. 2)

Mass spectra of peaks 23, 24 and 25 were not clean and could not be characterized.

Peak No.	Composition (Peak area %)	Identified compounds
5	0.27	Limonene
6	2.71	Pentanol
8	1.57	<i>cis</i> -2-Penten-1-ol
9	10.91	2,5 or 2,6-Dimethyl pyrazine
10	0.55	<i>cis</i> -3-Hexenol
11	5.58	2-Methyl-5-ethyl pyrazine
12	5.87	<i>trans</i> -Linalooloxide (five membered)
13	6.52	<i>cis</i> -Linalooloxide (five membered)
14	2.58	2-Acetylfuran
15	20.68	Linalool
17	5.28	<i>trans,trans</i> -3,5-Octadiene-2-one and 5-methyl furfural
18	10.96	3,7-Dimethyl-1,5,7-octatriene-3-ol
19	13.58	1-Ethyl-2-formyl pyrrole
20	3.21	2-Acetyl pyrrole
21	1.28	Anethole
22	4.18	α -Terpineol

FIG. 3-1. Preparative Gas Chromatogram of Aroma Concentrate from Lotus Tea.

Column: 3 mm (*i.d.*) $\times$ 300 cm stainless steel. Column packing: 23% Carbowax 6000 on Shimalite W 60/80. Column temp.: 75°C $\xrightarrow{2^\circ/\text{min}}$ 180°C. Detector: T.C.D.

FIG. 3-2. GC-MS Chromatogram of Trap 2.

Column: Gelay Column, Carbowax 20 M, 0.5 mm (*i.d.*) $\times$ 45 m. Injection temp.: 200°C, Helium flow rate: 3 ml/min. Column temp.: 40°C $\xrightarrow{1^\circ/\text{min}}$ 60°C $\xrightarrow{2^\circ/\text{min}}$ 170°C.

Detector: Total ion monitor.

FIG. 4. IR and Mass Spectra of Trap 2 from Lotus Tea Aroma Concentrate.

FIG. 5. GC-MS Chromatogram of Aroma Concentrate from Lotus Pollen.

(A), dried pollen, (B), fresh pollen.

Column: 3 mm (i.d.) $\times$ 300 cm stainless steel, Column packing: 23% Carbowax 20 M on Shimalite W 60/80. Injection temp.: 200°C, Helium flow rate: 30 min/ml. Column temp.: 75°C $\xrightarrow{2^\circ/\text{min}}$ 180°C. Detector: total ion monitor.

Trap No. 2 was obtained as crystalline form. Rechromatogram of trap No. 2 after dissolving with ether, on column Carbowax 20M, showed only one peak (peak A) besides solvent peak. GC retention data, IR and MS of peak A were entirely identical with those of authentic 1,4-dimethoxybenzene as shown in

Fig. 4. Natural occurrence of 1,4-dimethoxybenzene has been only reported as a minor component of essential oil from flower of lilac (*Syringa vulgaris* L.).²⁾ The components in the other three traps were the same as those in the aroma concentrate from Bach-Mao tea.

3) Isolation of 1,4-dimethoxybenzene from both dry and fresh lotus pollen

Since 1,4-dimethoxybenzene was identified as the main aroma constituent in lotus tea, aroma concentrates were prepared from both dry and fresh lotus pollen and analyzed by the same procedure as mentioned above. As the result, 1,4-dimethoxybenzene was found in amount to 93% of aroma concentrate prepared from dry pollen, and 30.4% of that from fresh pollen. The chromatograms are shown in Fig. 5.

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