NOTE**Benzofuran Compositions of Unsaponifiabiles of
Styrax japonica Sieb. Et Zucc**

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Three fluorescent compounds under UV lamp were obtained from the unsaponifiabiles of immature fruit oil of *Styrax japonica* Sieb et Zucc. and their compositions were determined by GLC. The fluorescent compounds was found to be 5-(3-hydroxypropyl)-2-(3,4-methylenedioxyphenyl) benzofuran : (demethoxyegonol), egonol, and 5-(3-hydroxypropyl)-2-(3,4-dimethoxyphenyl) benzofuran : (demethoxyhomoegonol).

1 Introduction

Egonol first isolated in 1915 by Okada¹⁾ from the seed oil of *Styrax japonica* Sieb. et Zucc., was formulated by Kawai as 5-(3-hydroxypropyl)-7-methoxy-2-(3',4'-methylenedioxyphenyl) benzofuran (2). Egonol has also been isolated from other *Styrax* species (*S. formosanus*, *S. obassia* and *S. americana*) and its spectroscopic data were reported²⁾. From the glycosidic fraction obtained from the seeds of *S. officinalis*, Segal *et al.*³⁾ isolated egonol and veratryl analogue, which has been indexed as homoegonol (4)⁴⁾. Egonol and demethoxyegonol (1) were obtained from the unsaponifiable constituent of the seed oil of *Styrax obassia* Sieb. et Zucc.⁵⁾. In the course of chemical investigation of constituents of *Styrax* family,¹¹⁾ we isolated egonol and two minor constituents anew from the unsaponifiabiles of immature fruit oil of *Styrax japonica*. We report here the isolation and identification of egonol and two other minor constituents and their compositions which are determined by GLC.

2 Experimental**2.1 General Procedures**

IR was taken in KBr on a Nihonbunko A-3 infrared spectrometer. NMR spectra were measured with a Varian XL-100 spectrometer in CDCl₃ which contained tetramethylsilane as internal reference. MS spectra were measured with a Hitachi RMU-6 M mass spectrometer using an ionization voltage of 70 eV. UV spectra were recorded on a Hitachi EP-3 T spectrometer. All

melting points were determined on a micro mp apparatus (Yazawa BY-2) and indicated as uncorrected values.

2.2 Extraction of *S. japonica* Sieb. et Zucc

The immature fresh fruits (2170 g) of *Styrax japonica* Sieb. et Zucc., which were harvested on september 1978 in Koganei-shi, Tokyo and dried in an electric oven at 60°C, were crushed and extracted with ether. The ether extracts (68.9 g) were given by concentration of the filtrate. The *n*-hexane solubles were refluxed with acetone (350 mL) for 1h. Then the filtrate was concentrated *in vacuo*, and the acetone solubles (59.4 g) were given. The acetone solubles were saponified by refluxing with 10 % KOH-EtOH solution (350 mL) and silica gel (C-200, Wako, 300 g) was added to the cooled reaction mixture. Unsaponifiabiles (5.8 g) were extracted with three 1000 mL ether portions from silica gel powder which was adsorbed with the reaction mixture.

2.3 TLC

A part of unsaponifiabiles was fractionated on 20 × 20 cm plates coated with a 0.5 mm layer of Wako gel B-5 (Wako Pure Chemical Industry Ltd., Osaka). The sample (700 mg) was applied uniformly along a line of 1.0 cm from one base edge of the plate (20 × 20 cm) and developed with C₆H₆-EtOAc; (10 : 1 vol/vol) for 1h.

The plate was observed under UV lamp and three fluorescent compounds were collected from the plates followed by extracting with ether. The ether extracts containing three fluorescent compounds were analyzed by subsequent GLC. The

unsaponifiables were also fractionated on silica gel column chromatography with C_6H_6 and C_6H_6 -ether (100 : 1).

2.4 GLC

The amount of demethoxyegonol, egonol and homoeegonol were analyzed with a Shimadzu GC-4 CM gas chromatography equipped with a flame ionization detector. The GLC was fitted with a 2 m glass column, 3 mm inside diameter packed with 1.5 % SE-30 on Gas Chrom-Z 80 mesh. The column was operated generally at 259°C with nitrogen at 20 mL/min as carrier gas. Detector temperature was 280°C.

3 Results and Discussion

Unsaponifiables were extracted with ether from silica gel powder adsorbed with the reaction mixture. Authentic samples of demethoxyegonol (1), and egonol (2) were isolated from the unsaponifiables of *Styrax obassia* Sieb et Zucc⁵⁾.

Three fluorescent compounds under UV lamp were determined from unsaponifiables by TLC using C_6H_6 -EtOAc (10 : 1 vol/vol) as developing solvents. The eluates were determined by GLC (shown in Table-2).

The isolated compound gave a solid one (R_f = 0.14) which was recrystallized from MeOH and identified as demethoxyegonol, mp 112~115°C (lit. 118~119°C)⁵⁾, (lit., 124~125°C)⁸⁾, UV : λ_{max}^{EtOH} 214.5, 320 and 330 nm. (ϵ , 37200, 33700 and 27500).

The integrated NMR spectrum included signals indicating no methoxyl group, the presence of one methylenedioxy group (s, δ , 5.98), the seven aromatic protons (m, δ , 6.76~7.45), two benzylic protons (m, δ , 2.69~2.87), two protons adjacent to hydroxyl function (t, δ , 3.68, J = 6.6 Hz). The remaining three protons were attributed to a hydroxyl group and two aliphatic protons (δ , 1.82~2.08). The value of chemical shift was in close agreement with that one reported^{5),8)}. The IR spectrum provided the ab-

Table-1 Total lipid compositis of *Styrax japonica* Sieb. et Zucc.

Dried immature fruits (% of fresh wt)	32.7
Ether extracts (% of dry wt)	9.7
<i>n</i> -Hexane solubles (% of ether extract)	89.4
Acetone solubles (% of <i>n</i> -hexane solubles)	96.4
Unsaponifiables (% of acetone solubles)	9.8

sorption of 3380 (OH), 2950 (aliphatic CH₂), 1600 (unsaturated ring), 1480 (substituted furan ring), 1230 (furan ring) and 930 cm⁻¹ (methylenedioxy). In the MS spectrum, the peak at m/e 296 (M^+) (100%) (base peak) and other fragments were observed at m/e 251 (53 %) and m/e 252 (81 %).

Acetylation with pyridine-Ac₂O (24h, room temp.) yielded the monoacetate mp 101~102°C.

The above data agreed with the authentic demethoxyegonol. Our isolated egonol (2) gave a solid compound (R_f = 0.10) which was recrystallized from MeOH and identified as authentic egonol with the mp, NMR, UV, and MS spectra⁵⁾.

Acetate of 2 showed the mp 104 ~105°C (lit. 103~105°C)⁵⁾. These above data also support the authentic egonol structure. The isolated compound (3) gave a solid one (R_f = 0.07), which was recrystallized from MeOH identified as demethoxy homoeegonol; mp 112.2~111.7°C (lit. 108~110°C)⁸⁾, UV : λ_{max}^{EtOH} 214.5, 316 and 330 nm. (ϵ , 34500, 36000, and 30000). The integrated

Table-2 Compositions of benzofuranoids and their R_f values in unsaponifiables.

Compounds	1	2	3
RR_t values ^{a)}	1.00	1.54	1.17
per cent composition (%)	18.0	71.5	10.5
R_f values ^{b)}			
C_6H_6 -EtOAc (10 : 1)	0.14	0.10	0.07
C_6H_6 -EtOAc (2 : 1)	0.29	0.24	0.21
C_6H_6 -Ether (10 : 1)	0.19	0.14	0.10

a) RR_t = relative retention time on LGC; RR_t for demethoxyegonol (1) is taken.

b) R_f values of TLC at room temperature. Demethoxyegonol (1); Egonol (2); Demethoxy-homoeegonol (3)

NMR spectrum included signals indicating a methoxyl group, the presence of two methoxyl group (s, δ , 3.93 and s, 3.98), seven aromatic protons (δ , 6.83~7.50), two benzylic protons (δ , 2.70~2.90), and two protons adjacent to hydroxyl function (t, δ , 3.70, $J=6.6$ Hz).

The remaining three protons were attributed to a hydroxyl group and two aliphatic protons (δ , 1.77~2.01). The IR spectrum showed the absorption of 3350 (OH), 2950 (aliphatic CH_2), 1603 (unsaturated ring), 1470 (substituted furan ring) and 1230 cm^{-1} (furan ring).

In the MS spectrum, the peak at m/e 312 (M^+) (100 %) (base peak) and other fragment; m/e 267 (30 %) and m/e 268 (45 %) agreed with the structure (3).

Acetylation of the compound with pyridine-acetic anhydride (24h, room temp.) yielded the monoacetate, mp 109.8~111.0°C. These above data were in close agreement with that reported⁸⁾.

It was very noticeable that demethoxyhomoeconol (3) was first isolated from natural sources. The compound (3) was only synthesized before⁹⁾.

By comparison of plants belong to the same family, we also found that the seeds of *S. officinalis* contained egonol and homoeconol, while immature fruits of *S. japonica* contained egonol, demethoxyegonol and demethoxyhomoeconol^{10), 11)}.

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エゴ (*Styrax japonica* Sieb. Et Zucc) 不けん化物におけるベンゾフラン化合物の組成

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エゴ (*Styrax japonica* Sieb. Et Zucc) 未熟種子油の不けん化物成分中のベンゾフラン化合物を検索した。紫外線ランプ下で強い蛍光を示した三種類のベンゾフラン化合物: 5-(3-hydroxypropyl)-2-(3',4'-methylenedioxy phenyl) benzofuran [デメトキシエゴノール], エゴノール, 5-(3-hydroxypropyl)-2-(3',4'-dimethoxyphenyl) benzofuran [デメトキシホモエゴノール] を単離, 同定し, それらの含有組成を GLC より決定した。また, デメトキシホモエゴノールが天然物から単離されたのは, これが初めてであることもわかった。

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★IFSCC. 新役員を選出

英国の P.J. Rothwell 氏が, 西独の L. Motischke 氏の後を引き継いで, 国際化粧品化学者協会連合(IFSCC)の会長に選出された。その他の役員については省略。

★スイス, 初の大豆を収穫

スイスは初の商業ベースで生産した大豆を単収 41 ブッシェル/エーカー (3,034 kg/1.06 ha) で収穫した。1988 年の作付は試験的なものであるが, 1989 年には, このスイスで開発された品種の大豆作付面積はさらに拡大する見込み。

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