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Studies on the Constituents of *Sophora flavescens* Aiton[1]

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New 2-alkylchromone derivatives were isolated from the epigeal part of *Sophora flavescens* Aiton. These compounds were obtained as a mixture of 2-*n*-heneicosyl-5,7-dihydroxy-6,8-dimethylchromone, 2-*n*-tricosyl-5,7-dihydroxy-6,8-dimethoxychromone and the homologous compounds, in which 2-alkyl groups are *n*-undecyl, *n*-tridecyl, *n*-pentadecyl, *n*-heptadecyl and *n*-tricosyl. Their structures were clarified by the spectral and chemical data, and the synthesis of 2-*n*-heptadecyl-5,7-dihydroxy-6,8-dimethylchromone.

Keywords——2-alkylchromones; isolation; synthesis; *Sophora flavescens*; 2-*n*-heptadecyl-5,7-dihydroxy-6,8-dimethylchromone; 2-*n*-heneicosyl; 2-*n*-tricosyl

The components of the roots of *Sophora flavescens* Aiton (Leguminosae) have appeared in the numerous studies, while those of the herb have been still unknown except luteolin-7-glucoside.[3] In the course of our studies on the constituents of the epigeal part of this plant, new 2-alkylchromone derivatives were isolated. From the portion soluble in ethyl acetate on the methanolic extract of the fresh stems and leaves, a compound (I) was obtained as a pale yellow crystalline powder, mp 122°, which showed a positive reaction by ferric chloride reagent and a negative magnesium–hydrochloric acid reaction. The infrared (IR) spectrum of I showed the presence of methylene groups (2900 cm^{-1}, strong) due to aliphatic long chain and a chelated carbonyl group (1660 cm^{-1}). The mass spectrum (MS) of I, gave mainly molecular Ion *m/e*: 500.3839 for $C_{32}H_{52}O_4$ and 528.4155 for $C_{34}H_{56}O_4$. On the other hand, this compound showed a single spot on thin layer chromatogram (TLC) of silica gel and could not be separated furthermore. Methylation of I with diazomethane gave monomethyl ether (II) and the methylation of I with dimethyl sulfate and potassium carbonate afforded dimethyl ether (III), which showed a negative ferric chloride reaction. The ultraviolet (UV) spectrum of I shown in Figure 1 was closely similar to that of leptorumol (5,7-di-

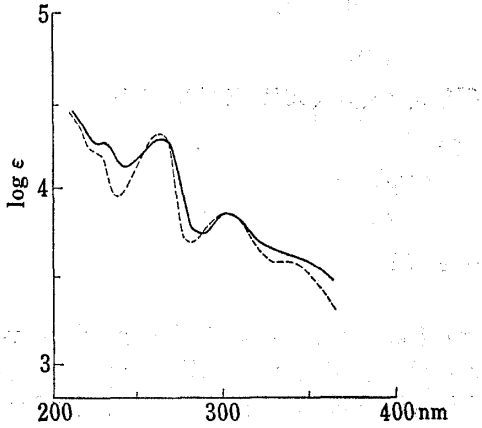

Fig. 1. UV Spectra of I (——) and Leptorumol (------) in EtOH

Table I. GLC of Methyl Ester of Acids obtained by Alkali Decomposition of I

t_R (min, sec)	Area ratio (%)	Identification
3′15″	0.5	Methyl *n*-tetradecanoate
6′30″	3.7	Methyl *n*-hexadecanoate
10′50″	3.2	Methyl *n*-octadecanoate
15′40″	1.5	Methyl *n*-eicosanoate
20′20″	30.9	Methyl *n*-docosanoate
25′00″	52.3	Methyl *n*-tetracosanoate
29′20″	3.1	Methyl *n*-hexacosanoate

1.5% OV-17 on Chromosorb W (2 m×3 mm i.d.) at 150—300° (3°/min); carrier gas, N_2 at 40 ml/min.

1) This work was presented at the 21st Annual Meeting of the Japanese Society of Pharmacognosy, Osaka, October, 1974; Abstract of Papers, 24 (1974).
2) Location: *2–2–1, Oshika, Shizuoka.*
3) T. Nakaoki, N. Morita, H. Mototsune, A. Hiraki, and T. Takeuchi, *Yakugaku Zasshi*, **75**, 172 (1955).

hydroxy-6,8-dimethylchromone[4]) and was revealed that I is a chromone derivative similar to leptorumol. The signals of the nuclear magnetic resonance (NMR) spectrum of I were assigned as follows: δ 13.15 (1H, s, C 5–OH), δ 5.95 (1H, s, O–C=CH–CO), δ 2.30 (3H, s, arom-CH_3), δ 2.24 (3H, s, arom-CH_3), δ 1.23 (mH, strong, $-(CH_2)_n-$), δ 0.87 (3H, $-CH_2-CH_3$), δ 2.55 (2H, t, arom-CH_2-CH_2-). On the NMR spectrum of III, the signal at δ 13.15 in I disappeared and new signals at δ 3.80 (3H, s, OCH_3) and δ 3.73 (3H, s, OCH_3) were found. On the IR spectra of I and III, an absorption at 1660 cm^{-1} in I due to chelated carbonyl group was found at 1658 cm^{-1} in III and was not shifted to high frequency by the O-methylation of the chelated hydroxyl group. This is characteristic of the derivatives[5]) of 5-hydroxychromone and 5-hydroxyflavone. The UV spectrum of II, λ_{max}^{EtOH} nm: 230, 246, 256, 263, 339, was also similar to that of 7-methylleotorumol, λ_{max}^{EtOH} nm: 225, 243, 263, 340. Therefore, the structure of I was presumed to be 2-alkyl-5,7-dihydroxy-6,8-dimethylchromone.

On the alkali decomposition of I, the phenolic product could not be obtained and the acidic product was methylated with diazomethane. As is shown in Table I, the analysis of the product by the gas liquid chromatography (GLC) shows that the product consists mainly of docosanoic acid (C_{22}) and tetracosanoic acid (C_{24}) containing each of small amount of straight saturated fatty acid from C_{14} to C_{26}. This result shows that I is a mixture of 2-*n*-heneicosyl-5,7-dihydroxy-6,8-dimethylchromone (IV) and 2-*n*-tricosyl-5,7-dihydroxy-6,8-dimethylchromone (V) containing of small amount of 2-*n*-tridecyl, 2-*n*-pentadecyl, 2-*n*-heptadecyl, 2-*n*-nonadecyl and 2-*n*-pentacosyl 5,7-dihydroxy-6,8-dimethylchromone.

In order to confirm the structure of I, the synthesis of 2-*n*-tricosyl-5,7-dimethoxy-6,8-dimethylchromone (VI), which is a main component of III, was tried, but Claisen condensation between methyl *n*-tetracosanoate and 2-hydroxy-4,6-dimethoxyacetophenone (VII) or 2-hydroxy-3,5-dimethyl-4,6-dimethoxyacetophenone (VIII) not occurred. As is shown in Chart 1, Claisen condensation of ethyl stearate with VII or VIII progressed smoothly in the

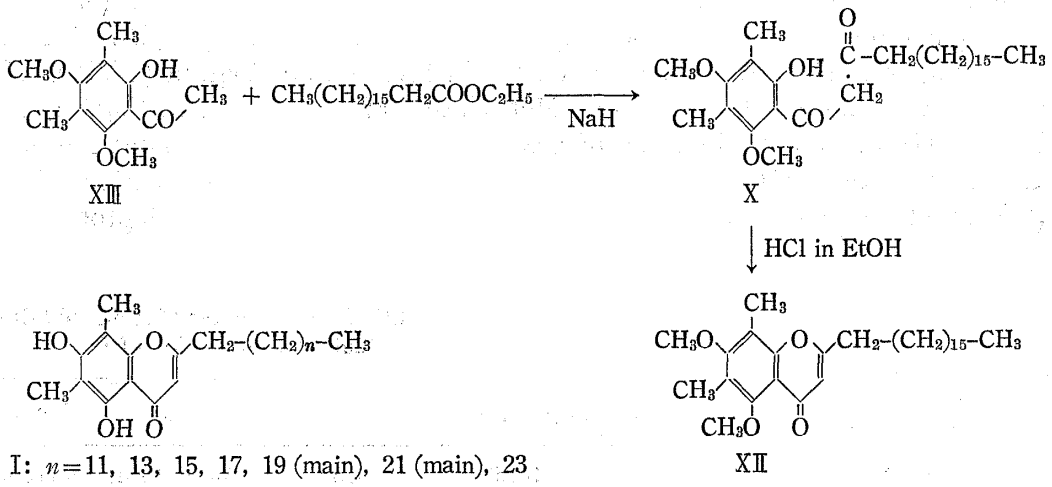

Chart 1. Synthesis and Structure of I

modified method of R. G. Cook and J. G. Down,[6]) to form diketone, ω-*n*-octadecanoyl-2-hydroxy-4,6-dimethoxyacetophenone (IX) or ω-*n*-octadecanoyl-2-hydroxy-3,5-dimethyl-4,6-dimethoxyacetophenone (X). Each of the diketone IX and X was cyclized by ethanolic hydrochloric acid to give 2-*n*-heptadecyl-5,7-dimethoxychromone (XI) and 2-*n*-heptadecyl-5,7-dimethoxy-6,8-dimethylchromone (XII). The property of XI was identical with that of litera-

4) S. Fukushima, T. Noro, Y. Saiki, A. Ueno, and Y. Akahori, *Yakugaku Zasshi*, **88**, 1135 (1968).
5) W. Baker, A.C.M. Finch, W.D. Ollis, and K.W. Robinson, *J. Chem. Soc.*, **1963**, 1477; A.J. East, W.D. Ollis, and R.E. Wheeler, *J. Chem. Soc.* (*C*), **1969**, 365.
6) R.G. Cooke and J.G. Down, *Aust. J. Chem.*, **24**, 1257 (1971); *idem*, *Tetrahedron Lett.*, **1970**, 1039.

ture,[6] and the IR, UV, mass and NMR spectra of XII showed almost identical with those of III. Consequently, the foregoing presumption on the structure of I was confirmed, and I is a mixture of IV, V and the homologoues as shown in Chart 1. A similar example has been reported by R. G. Cook and J. G. Down,[6] in which the mixture of 2-*n*-heptacosyl, 2-*n*-nonacosyl and 2-*n*-henetriacosyl-5,7-dihydroxy-6-methylchromone were isolated from *Dianella revoluta* (Liliaceae) and *Stypandra grandis* (Xanthorrhoeaceae). The presence of 2-akylchromone derivatives in *Sophora flavescens* is an interesting fact as the second example.

Experimental

All melting points are uncorrected. UV spectra were measured using a Hitachi recording spectrometer EPS-032 type. IR spectra were recorded for KBr tablet with Jasco IRA-2 spectrometer. NMR spectra were taken at 60 MHz with TMS as an internal standard using Hitachi R 24 high resolution NMR spectrometer. The signals were given on chemical shifts in δ and following abbreviation: s=singlet, d=doublet, t=triplet, m=multiplet. Mass spectra were determined at 80 eV with Hitachi RMU and high mass was taken by Nihondenshi JMS-01SG-2 mass spectrometer. GLC was carried out using Hitachi K-53 and the plates of TLC were prepared by Kiesel gel nach Stahl and column chromatography was performed on Wakogel C-200 and Silicic acid of Mallinckrot.

Isolation of 2-*n*-Alkyl-5,7-dihydroxy-6,8-dimethylchromones (I)——24 kg of the fresh leaves and stems collected at Awagatake, Kakegawa-shi, Shizuoka in May, 1974, were steeped in MeOH for 18 days at room temperature. The methanolic extract was concentrated to remove MeOH and the residual liquid was extracted with EtOAc. After removal of the solvent, the EtOAc-extract was treated with EtOAc once more and 247 g of a portion soluble in EtOAc was obtained. This was chromatographed on a column of two layers (upper, cellite; under, silica gel) and the fraction eluted with *n*-hexane: benzene (1:3), was treated with acetone. The insoluble portion in acetone was recrystallized from $CHCl_3$ to give 228 mg of a pale yellow powder, mp 122°. Its ethanolic solution showed a blue green color by $FeCl_3$ and negative reaction by Mg–HCl. *Anal.* Found: C, 76.79; H, 10.58.[7] IR ν_{max}^{KBr} cm^{-1}: 2900 (C–H), 1660 (C=O). UV λ_{max}^{EtOH} nm (log ε)[7]: 230 (4.26), 263 (4.30), 303 (3.88), 330 (3.60). MS *m/e* (%): 500 (M^+, 100), 528 (M^+, 90), 247 (19), 233 (70), 220 (90), 191 (50), 181 (69.5), 153 (24), 151 (24), no peak above 10% from 500 to 247. High MS: (556.4506=$C_{36}H_{60}O_4$), 528.4155=$C_{34}H_{56}O_4$, 500.3839=$C_{32}H_{52}O_4$, (472.3568=$C_{30}H_{48}O_4$), (444.3200=$C_{28}H_{44}O_4$). NMR (in C_5D_5N+$CDCl_3$): δ 0.87 (3H, t), 2.55 (2H, t), 1.23 (*ca.* 46H, s), 2.30 and 2.24 (each 1H, d, 6,8-dimethyl), 5.95 (1H, s, H-3), 13.15 (1H, s, OH-5).

2-Alkyl-5-hydroxy-7-methoxy-6,8-dimethylchromones (II)——A mixture of I (40 mg) and an etherial solution of CH_2N_2 was left stand overnight, then the solvent was evaporated and the residue was recrystallized from MeOH to give 28 mg of pale yellow prisms, mp 53—55, which showed a red violet color by $FeCl_3$ in EtOH. *Anal.* Found: C, 77.15; H, 10.81. UV λ_{max}^{EtOH} nm (log ε as $C_{34}H_{56}O_4$)[7]: 230 (4.15), 246 (4.22), 256 (4.20), 263 (4.19), 339 (3.51).

2-*n*-Alkyl-5,7-dimethoxy-6,8-dimethylchromones (III)——A mixture of I (50 mg), Me_2SO_4 (2 ml), K_2CO_3 (4.5 g) and dry acetone (20 ml), was refluxed for 7 hr. The reaction mixture was filtered and concentrated. The residue was purified on preparative TLC and recrystallized from MeOH to give colorless plates, mp 63—64°. *Anal.* Found: C, 77.37; H, 10.38. UV λ_{max}^{EtOH} nm (log ε as $C_{35}H_{58}O_4$):[7] 229 (4.23), 236 (4.25), 256 (3.98), 310 (3.58). IR ν_{max}^{KBr} cm^{-1}: 1658 (C=O). MS *m/e* (%): 528 (50), 556 (100), 261 (16), 243 (17), 231 (20), 218 (11), 165 (10). NMR (in $CDCl_3$): 0.87 (3H, t), 1.23 (*ca.* 46H, s), 2.55 (2H, t), 3.73, 3.80 (each 3H, s, 5,7-OCH_3), 2.30, 2.24 (each 3H, s, 6,8-CH_3), 5.95 (1H, s, H-3).

Alkali Decomposition of I and Identification of Fatty Acid——A solution of I (20 mg) in EtOH (9 ml) and 20% aq. KOH was refluxed under N_2 atomospher for 15 hr. After cooling, the reaction mixture was acidified with HCl and extracted with ether. The ethereal extract was washed with aq. $NaHCO_3$ and the aqueous layer was acidified with HCl to separate colorless crystals, which were methylated using an etherial solution of CH_2N_2. The product was analyzed on GLC and identified as shown in Table I by the mixed GLC with authenthic samples of methylate of fatty acid.

Synthesis of 2-*n*-Heptadecyl-5,7-dimethoxy-6,8-dimethylchromone (XII)——To a stirred suspension of NaH (105 mg) in dry ether and ethyl stearate (2.7 g), was added under heating at 50° 197 mg of 2-hydroxy-3,5-dimethyl-4,6-dimethoxyacetophenone (VIII) prepared by the method of the previous report.[4] The mixture was heated at 110° under stirring for 2 hr and then poured into ice cooled 1 N HCl. The resulting precipitate was collected, washed with H_2O and dried to give 286.6 mg (70%) of crude ω-*n*-octadecanoyl-2-hydroxy-3,5-dimethyl-4,6-dimethoxyacetophenone (X). This product was dissolved in EtOH: conc.-HCl

7) *Anal.* Calcd. for 0.5 M $C_{32}H_{52}O_4$+0.5 M $C_{34}H_{56}O_4$; C, 76.99; H, 10.58, This was used tentatively as molecular weight of I for the calculation of the UV spectrum and the following derivatives are in the same proportion, respectively.

(4 : 1) and the solution was refluxed on a boiling water bath for 5 min. After cooling the mixture, the separated solid was collected, washed with H_2O, dried and recrystallized from EtOH to give 160 mg (38%) of XII as colorless needles, mp 67—68°. *Anal.* Calcd. for $C_{30}H_{48}O \cdot 1/6\ H_2O_4$: C, 75.80; H, 10.18. Found: C, 75.74; H, 10.10. IR ν_{max}^{KBr} nm^{-1}: 2920 (C–H), 1658 (C=O). UV λ_{max}^{EtOH} nm (log ε): 226 (4.22), 229 (4.21), 256 (4.01), 280 (3.65), 310 (3.64). MS *m/e* (%): 472 (M^+, 100), 261 (5), 243 (7.4), 231 (8.3), 218 (4.7), 165 (3.3), 147 (3.9). NMR (in $CDCl_3$): δ 0.86 (3H, t), 1.24 (30H, s), 2.26, 2.32 (each 3H, s, 6,8-CH_3), 2.56 (2H, t), 3.76, 3.82 (each 3H, s, 5,7-OCH_3), 6.01 (1H, s, H-3).

2-*n*-Heptadecyl-5,7-dimethoxychromone (XI)——This compound 176.8 mg (23.4%) was obtained by the cyclization of 384.6 mg of *ω-n*-octadecanoyl-2-hydroxy-4,6-dimethoxyacetophenone, colorless plates, mp 94—96° (EtOH), which was prepared from ethyl stearate (2.7 g) and 2-hydroxy-4,6-dimethoxyacetophenone (VII, 332.9 mg) by the same method in the case of XII. XI was crystallized from EtOH to colorless plates, mp 108—110°. IR ν_{max}^{KBr} cm^{-1}: 2920 (C–H), 1662 (C=O). UV λ_{max}^{EtOH} nm (log ε): 230 (4.57), 245 (4.53), 253 (4.52), 284 (4.16). MS *m/e* (%): 444 (M^+, 100), 233 (34), 203 (17), 181 (10). NMR (in $CDCl_3$): δ 0.86 (3H, t), 1.25 (30H, s), 2.53 (2H, t), 3.77, 3.83 (each 3H, s, 5,7-OCH_3), 5.95 (1H, s, H-3).

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