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Studies on the Constituents of *Lespedeza homoloba* Nakai. II.[1] The Structure of Lespedeol B

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A new isoflavanone derivative named lespedeol B was isolated from the bark of *Lespedeza homoloba* Nakai. The structure was established as 2′,4′,5-trihydroxy-8-methyl-8-(4-methylpent-3-enyl)pyrano(3,2-*g*)isoflavanone by spectral and chemical data, in which lespedeol B was formed by the dehydrocyclization of lespedeol A.

In the previous paper,[1] we reported the structure of lespedeol A isolated from the bark of *Lespedeza homoloba* Nakai. In our further study on the constituents of this plant, a new isoflavanone derivative containing 2-methyl-2-(4-methylpent-3-enyl) chromene ring named lespedeol B was isolated from the benzene–dichloromethane (1:2) fraction, as pale yellow crystals; mp 165—166°, $C_{25}H_{26}O_6$ (M=422). Lespedeol B (I) showed a positive ferric chloride reaction and a negative mangesium–hydrochloric acid reaction. The infrared (IR) spectrum showed the presence of hydroxyl (3350 cm^{-1}) and carbonyl (1620 cm^{-1}) groups. The ultraviolet (UV) spectrum (Fig. 1) suggested the presence of chromene ring in I, which is supported by the appearance of each doublet at τ 4.50 and 3.21 (J=9.8 Hz), in the nuclear magnetic resonance (NMR) spectrum.

On the methylation with diazomethane, I gave dimethyllespedeol B (II), as a pale yellow oil. On the catalytic hydrogenation with 5% palladium on charcoal, II gave dimethyltetrahydrolespedeol B (III), $C_{27}H_{34}O_6$, as a pale yellow oil.

The UV spectrum of III (Fig. 1) is similar to that of lespedeol A.[1] On the NMR spectrum of II (Fig. 2), a multiplet at about τ 5.7 (3H) is assigned to the protons of 2 and 3 position in the isoflavanone nucleus. Therefore, lespedeol B is assumed to be an isoflavanone derivative.

Figure 2 shows two doublets at τ 4.69 and 3.34 (each 1H, J=9.6 Hz, chromene ring), three singlets at τ 8.58, 8.41 and 8.33 (each 3H, =C–CH_3, $\overset{-C}{-O}\!>$C–CH_3), a broad signal at τ 8.02 (2H, =C–CH_2–), and a broad signal at τ 4.95 (1H, =CH–), which are similar to those of 6-acetyl-5-hydroxy-2-methyl-2-(4-methylpent-3-enyl)chromene.[3] On the other hand, the NMR spectrum of III shows two broad triplets at τ 8.28 and 7.43 (each 2H, J=7.5 Hz) and a singlet at τ 8.70 (3H, CH_3–C–O–), and the signals of two protons of chromene ring and a proton attached to double bond disappear in this spectrum. These facts show the presence of 2-methyl-2-(4-methylpent-3-enyl)chromene as a partial structure.

The mass spectrum of I (Fig. 3) shows the following peaks; *m/e* (%), 339 (100), 321 (73.3), 217 (90.0), 203 (66.7), 136 (33.3), 123 (60.0). The base peak is found at *m/e* 339 due to the loss of a isohexenyl group from the molecular ion. The significant peak at *m/e* 321 shows the loss of water from *m/e* 339. The metastable peak is found at *m/e* 304. The characteristic peaks at *m/e* 203 and 136 show the fragmentation of retro Diels-Alder type from *m/e* 339.

1) Part I: A. Ueno, M. Ichikawa, T. Miyase, S. Fukushima, Y. Saiki and K. Morinaga, *Chem. Pharm. Bull.* (Tokyo), **21**, 1734 (1973).
2) Location: *2-2-1, Oshika, Shizuoka.*
3) W.M. Bandaranayake, L. Crombie and D.A. Whiting, *J. Chem. Soc.*, (*C*), **1971**, 804.

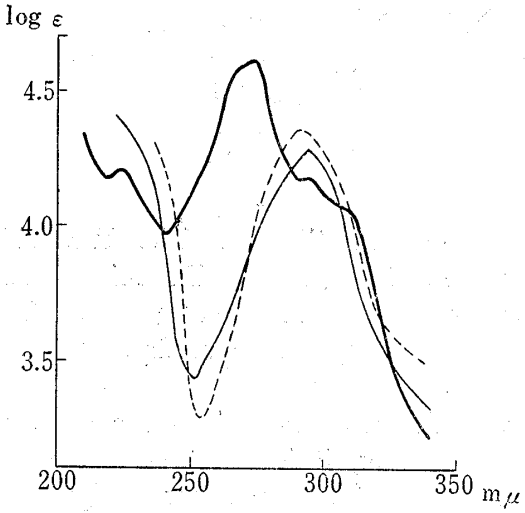


Fig. 1. The UV Spectra of I (———), III (– – –) and Lespedeol A (·····) in EtOH

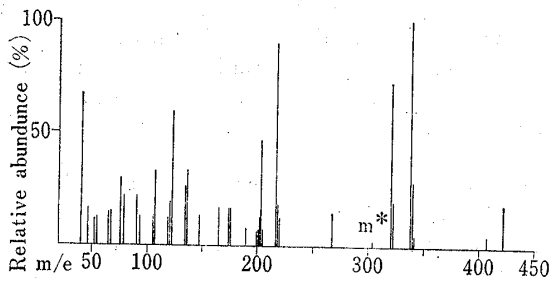
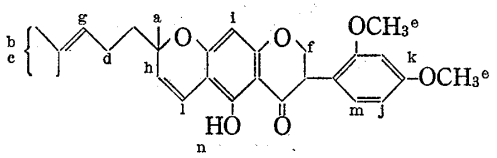


Fig. 3. The Mass Spectrum of Lespedeol B



a: 8.58 b: 8.41 c: 8.33 d: 8.02
e: 6.27, 6.24 f: 5.7 g: 4.95
h: 4.69 i: 4.20 j: 3.62 k: 3.60
l: 3.34 m: 3.07 n: −2.7

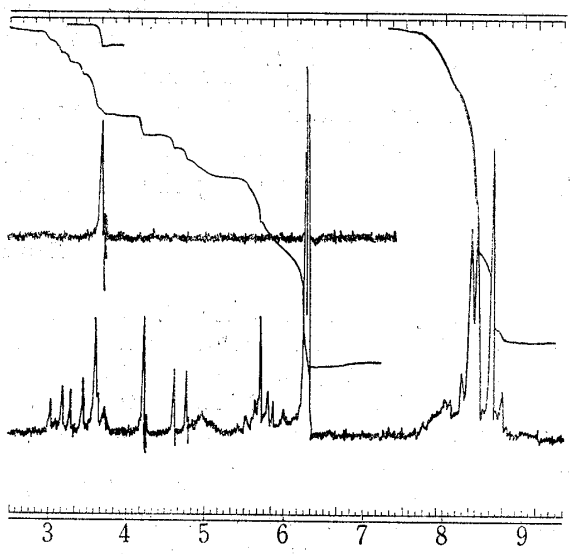


Fig. 2. The NMR Spectrum of II in CCl_4 (60 M; τ Values)

The peak of *m/e* 203 shows the presence of a chromene ring and two hydroxyl groups in the ring A, and the peak of *m/e* 136 shows the presence of two hydroxyl groups in the ring B of the isoflavanone nucleus.

On the NMR spectrum of II, the aromatic proton signals are similar to those of trimethyllespedeol A[1] and are assigned on the base of their chemical shift values and their *ortho* and *meta* coupling constants as shown in Table I.

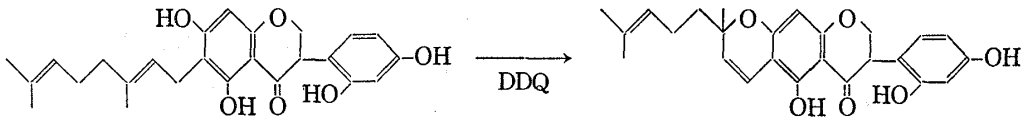
Table I. Aromatic Proton Signals of Lespedeol B, Its Derivatives and Lespedeol A in Their NMR Spectra

Compound	Lespedeol B	Dimethyl-lespedeol B	Tetrahydrodimethyl-lespedeol B	Lespedeol A
Solvent	C_5D_5N	CCl_4	CCl_4	C_5D_5N
position	Chemical shifts τ (J=Hz)			
6 or 8	3.79 (s)	4.20 (s)	4.20 (s)	3.70 (s)
3′	3.08 (d, J=2.1)	3.60 (d, J=2.3)	3.56 (d, J=2.2)	3.13 (d, J=2.3)
5′	3.27 (d–d, J=8.3, 2.1)	3.62 (d–d, J=8.6, 2.3)	3.61 (d–d, J=8.8, 2.2)	3.20 (d–d, J=8.3, 2.3)
6′	2.77 (d, J=8.3)	3.07 (d, J=8.6)	3.05 (d, J=8.8)	2.85 (d, J=8.3)

The Gibbs' test of II is positive, which shows that a substituent is absent at the *para* position of the chelated hydroxyl group. Therefore, the structure of lespedeol B is presumed to be I.

The phenolic derivatives having a isopentenyl or geranyl group at the *ortho* position to the hydroxyl group give generally chromene derivatives by the oxidation with DDQ.[4] The

4) G. Cardillo, R. Cricchio and L. Merlini, *Tetrahedron*, **24**, 4825 (1968).



Lespedeol A Lespedeol B (I)

Chart 1

reaction using lespedeol A proceeded in the equation as shown in Chart 1. The IR and NMR spectra of the product are in fair agreement with those of lespedeol B. The mixed melting point of lespedeol B and this product did not show depression of the melting point. Therefore, the structure of lespedeol B is confirmed to be 2′,4′,5-trihydroxy-8-methyl-8-(4-methylpent-3-enyl)pyrano(3,2-*g*)isoflavanone.

Though, two chalcones[5] and a xanthone[6] were hitherto reported as the derivatives containing a 2-methyl-2-(4-methylpent-3-enyl)chromene ring, it is the first case that an isoflavanone derivative having the same partial structure is found in natural origin.

Experimental

All melting points are uncorrected. UV spectra were measured using a Hitachi recording spectrometer EPS-032 type. IR spectra were determined on KBr disks or NaCl plates (liquid) using a Hitachi infrared spectrometer EPI-G 21 type. NMR spectra were taken at 60 MHz with TMS as an internal standard using a JNM-C-60H high resolution NMR spectrometer. The chemical shifts were given in τ values. Abbreviation; s=singlet, d=doublet, t=triplet, m=multiplet, br.=broad. Mass spectrum was measured using a Hitachi RMS mass spectrometer.

Isolation of Lespedeol B (I)——The dried bark of *Lespedeza homoloba* Nakai was extracted with MeOH and the extract was separated to each fractions by the method reported in previous paper.[1] The *n*-hexane–benzene (3:1) fraction was chromatographed on a silica gel column. The fraction eluted with benzene–dichloromethane (1:2) gave about 500 mg of I as pale yellow crystals, mp 165—166° by the recrystallization from *n*-hexane–benzene. *Anal.* Calcd. for $C_{25}H_{26}O_6$: C, 71.07; H, 6.20. Found: C, 71.11; H, 6.48. IR ν_{max}^{KBr} cm^{-1}: 3350, 1620, 1260. UV λ_{max}^{EtOH} mμ (log ε): 275 (4.62) (Fig. 1). NMR (C_5D_5N, τ): 8.62, 8.43, 8.33 (each 3H, s), 5.3 (3H, m), 4.50, 3.21 (each 1H, d, J=9.8 Hz), 3.79 (s), 3.31 (d), 3.08 (d-d), 2.77 (d), −3.21 (s, chelated OH).

Dimethyllespedeol B (II)——To an etherial solution of I (150 mg) containing a small amount of MeOH was added an etherial solution of CH_2N_2 under cooling. The reaction mixture was stirred at room temperature for two days and then concentrated. The residue was chromatographed on a silica gel column using *n*-hexane–benzene (1:3) to give 55.7 mg of II as a pale yellow viscous oil which was difficult to obtain as a benzene free state, but it showed a single spot of *Rf* 0.53 on a thin-layer chromatogram of silica gel (TLC) using *n*-hexane–benzene (1:5). Gibbs' test of II showed a blue color (+). IR ν_{max}^{film} cm^{-1}: 3200, 1630, 1520.

Dimethyltetrahydrolespedeol B (III)——II (45 mg) was hydrogenated with 5% Pd-C (50 mg) as a catalyst in EtOH (20 ml) at room temperature. The catalyst was filtered off and the filtrate was concentrated. The residue was purified by the thin-layer chromatography using silica gel with benzene to give 30 mg of III as a colorless oil. *Anal.* Calcd. for $C_{27}H_{34}O_6$: C, 71.31; H, 7.54. Found: C, 71.61; H, 7.55. IR ν_{max}^{film} cm^{-1}: 3400, 1640, 1580. NMR (CCl_4, τ): 8.70 (3H, s), 5.7 (3H, m), 7.43 (2H, br. t, J=7.5 Hz).

Formation of Lespedeol B from Lespedeol A——A solution of lespedeol A (212 mg) and DDQ (113 mg) in dry benzene (40 ml) was refluxed for 5 hr. The reaction mixture was filtered and the filtrate was concentrated. The residue was recrystallized from *n*-hexane–benzene to give I (64 mg) as pale yellow crystals, mp 165—166°. Mixed mp 165—166° with natural lespedeol B. The IR, NMR and *Rf* of TLC were in fair agreement with those of the natural product.

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5) G. Cardillo, L. Merlini and R. Mondelli, *Tetrahedron*, **23**, 497 (1967).
6) W.D. Ollis, M.V.J. Ramsay, I.O. Sutherl and S. Mongkolsuk, *Tetrahedron*, **21**, 1453 (1965).