ISOFLAVAN DERIVATIVES FROM *GLYCYRRHIZA GLABRA* (LICORICE)

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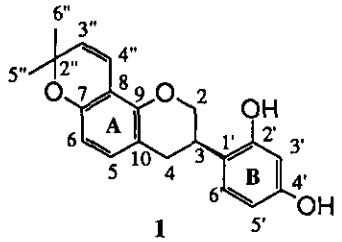
Abstract—A new isoflavan was isolated from the root of *Glycyrrhiza glabra* (licorice; Fabaceae), and its structure was elucidated as 8-prenyl-phaseollinisoflavan on the basis of spectroscopic evidence. Five known isoflavans have also been isolated, and identified with glabridin, 4'-*O*-methylglabridin, hispaglabridins A, B and what was previously elucidated as "3'-methoxyglabridin". The structure of the latter compound was revised to 3'-hydroxy-4'-*O*-methylglabridin by an irradiation experiment and long range ^{13}C-^{1}H COSY spectral analysis.

The dried roots of several *Glycyrrhiza* species (Fabaceae) have found wide medicinal use historically as the crude drug licorice throughout the world, and are characterized by the presence of the sweetening saponin glycyrrhizin. The main constituent glycyrrhizin has long been conceived to be by far the most important principle underwriting the pharmacological efficacy of licorice. However, there have been an increasing number of reports referring to either flavonoids or isoflavonoids as biologically active, *i.e.*, antimicrobial,[1-7] anti-oxidant,[5,6] enzyme-inhibitory[8-12] and anti-viral[13] principles of licorice. These findings imply that the phenolics cannot be disregarded not only from the pharmacological viewpoint but also as the possible chemical leads for developing medicinal drugs. We have engaged for years in chemical studies on *G. uralensis*,[5] *G. inflata*[14-16] and *G. pallidiflora*,[17] and have isolated a number of non-glycosidal phenolics. In a continuation of this research program, we investigated the roots of *G. glabra* (synonymous with *G. glabra* var. *glandulifera*) which is the principal source of licorice in the Occident. This paper describes properties and structural assignments of a new isoflavan constituent 8-prenyl-phaseollinisoflavan and structural revision of 3'-methoxyglabridin, a known isoflavan previously isolated from the Spanish variety of *G. glabra*, to 3'-hydroxy-4-*O*-methylglabridin.

The preliminary microchemical survey of *G. glabra* roots by thin-layer chromatography (tlc) indicated that a number of phenolic constituents occur in a less polar part of the extract. Thus, the CH_2Cl_2 extract



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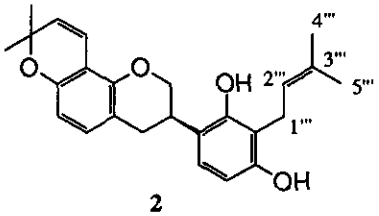

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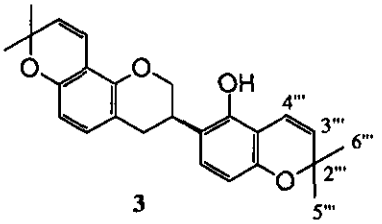

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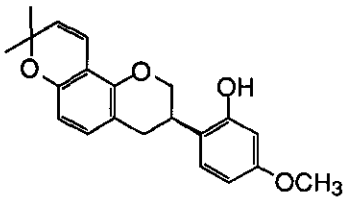

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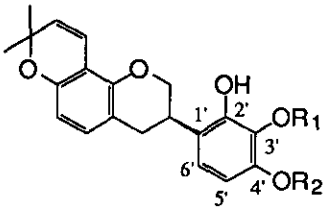
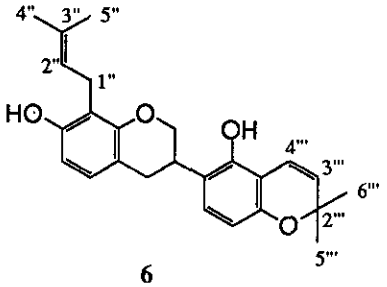


5a R_1=Me R_2=H
5b R_1=H R_2=Me



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that accounts for about 5% in weight of licorice was chosen as the subject for chemical investigation. The extract was chromatographed on a silica gel column on elution with $CHCl_3$-acetone increasing the amount of acetone stepwise. From less polar fractions eluted with $CHCl_3$ and 3 % $CHCl_3$-acetone, six compounds (**1-6**) were isolated. These are all isoflavan derivatives, four (comps. **1-4**) of which were readily identified with previously known compounds glabridin (**1**),[18] hispaglabridins A (**2**),[2] B (**3**)[2] and 4'-*O*-methylglabridin (**4**).[2] Compound (**5**) was found to be identical with what has been elucidated as 3'-methoxyglabridin (**5a**),[2] but it was found that its structure should be revised to **5b** as mentioned later. Table I presents results of analysis of ^{13}C resonances of these isoflavans, which were not available previously. Assignments were made based on both ^{13}C-^1H COSY and long range ^{13}C-^1H COSY spectral analysis. These data were of particular help in structural elucidation of compound (**6**), a new compound isolated from this licorice.

Spectroscopic properties and melting point of compound (**5**) were in good agreement with those reported for "3'-methoxyglabridin (**5a**)".[2] The substitution pattern of this compound in the B-ring was deduced on the basis of a shift reagent experiment in its uv spectrum. The methoxyl group was assigned to C-3' since the addition of $AlCl_3$ in the uv spectrum produced no bathochromic shift as is usually observed with catechols.[2] We cast doubt on its structure since this compound was susceptible to molecular oxygen implying the presence of catechol moiety in the molecule. There was another strong evidence urging revision of the structure (**5a**) for this compound. It was reported that the methoxyls of *ortho*-disubstituted anisoles in the ^{13}C resonances appear downfield at *ca.* 60 ppm whereas those of *ortho*-monosubstituted or nonsubstituted anisoles usually appear at *ca.* 55 ppm.[19] The observed chemical shift of a methoxyl group of 4'-*O*-methylglabridin was 55.1 ppm

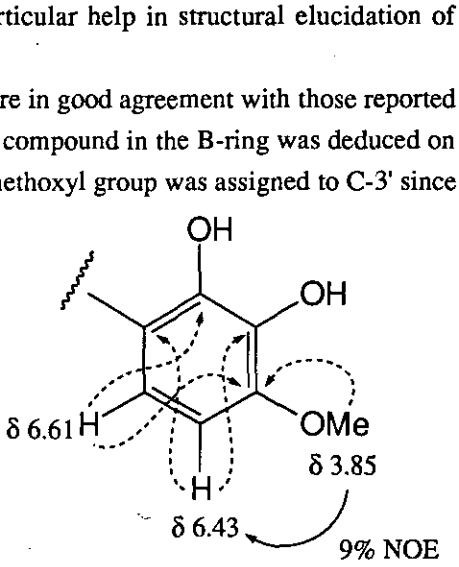


Figure 1 ^3J Interaction Observed in the Long-Range CH COSY of **5b** (-----) and NOE

Table I. ^{13}C-Nmr data for Isoflavan Derivatives Isolated from *G. glabra* root.

C	(1)	(2)	(3)	(4)	(5b)	(6)
C-2	70.0	70.2	70.1	69.9	69.9	70.1
C-3	31.7	31.8	31.8	31.5	31.9	31.7
C-4	30.6	30.6	30.6	30.4	30.4	31.0
C-5	129.2	129.2	129.2	129.2	129.2	127.6
C-6	108.7	108.6	108.6	108.6	108.6	108.0
C-7	151.9	151.7	151.8	151.4	151.8	153.5
C-8	109.9	109.9	109.9	109.9	109.9	114.4
C-9	149.8	149.8	149.8	149.6	149.8	152.4
C-10	114.3	114.6	114.6	114.7	114.5	114.7
C-1'	120.0	120.4	121.1	120.0	121.0	121.4
C-2'	154.4	152.9	151.5	154.6	142.3	151.7
C-3'	103.1	113.3	109.6	101.9	132.3	109.6
C-4'	155.2	153.7	150.4	159.0	145.8	150.2
C-5'	108.0	107.7	107.4	105.8	102.8	107.4
C-6'	128.4	125.1	127.0	128.0	117.6	126.9
A-ring chromene						
C-2"	75.6	75.7	75.6	75.8	75.6	-
C-3"	128.9	128.9	128.9	128.9	128.8	-
C-4"	116.9	117.0	117.0	116.9	117.0	-
C-5"	27.6*	27.5*	27.5*	27.3*	27.5*	-
C-6"	27.8*	27.7*	27.8*	27.5*	27.7*	-
B-ring chromene						
C-2"'	-	-	76.0	-	-	75.9
C-3"'	-	-	129.2	-	-	129.2
C-4"'	-	-	116.7	-	-	116.6
C-5"'	-	-	27.7*	-	-	27.7*
C-6"'	-	-	27.8*	-	-	27.7*
A-ring prenyl						
C-1"	-	-	-	-	-	22.4
C-2"	-	-	-	-	-	122.2
C-3"	-	-	-	-	-	134.2
C-4"	-	-	-	-	-	25.8
C-5"	-	-	-	-	-	17.9
B-ring prenyl						
C-1"'	-	22.8	-	-	-	-
C-2"'	-	121.3	-	-	-	-
C-3"'	-	136.2	-	-	-	-
C-4"'	-	25.8	-	-	-	-
C-5"'	-	17.9	-	-	-	-
OMe	-	-	-	55.1	56.1	-

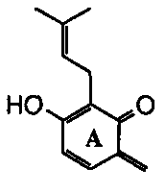
Spectra were measured in $CDCl_3$ with TMS as internal standard. Assignments were confirmed by ^{13}C-^{1}H COSY and long range ^{13}C-^{1}H COSY spectra.

*: Figures in the same column may be interchanged.

agreeing with the above rule, while that for compound (**5**) was 56.1 ppm apparently disagreeing with this rule if compound (**5**) has the structure (**5a**). It is not unlikely that *ortho*-substituted catechols fail to form the metal complex due to the presence of a relatively bulky substituent such as a methoxyl group and

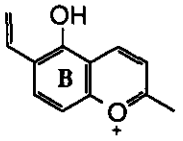
result in showing no bathochromic shift in the uv spectrum. This matter was finally settled by an irradiation experiment in which a 9% NOE on a proton signal at δ 6.43 was observed on irradiation of the methoxyl (Figure 1). This finding completely denied the location of a methoxyl at C-3', and unequivocally assigned it to C-4'. The ^{13}C-^{1}H COSY and long range ^{13}C-^{1}H COSY spectral analyses were also supportive of the 2',3'-dihydroxy-4'-methoxy substitution pattern for the B-ring (Table I and Figure 1). Thus, the structure of "3'-methoxyglabridin" was revised to **5b**, and it should be renamed 3'-hydroxy-4'-*O*-methylglabridin.

Compound (**6**) was a new isoflavan obtained as optically active colorless plates, mp 151-152°, $[\alpha]_D$ -7.6°, for which the name of 8-prenyl-phaseollinisoflavan was proposed. Its ^{1}H-nmr spectrum revealed the presence of proton signals assignable to one γ,γ-dimethylallyl [δ 5.27 (m), 3.40 (d, *J*=7.2 Hz), 1.80, 1.73 (3H each, s)], one chromene ring [δ 6.63 (d, *J*=9.9 Hz), 5.59 (d, *J*=9.9 Hz), 1.42, 1.40 (3H each, s)] and two sets of AB couplings [δ 6.80 (d, *J*=8.2 Hz), 6.76 (d, *J*=8.4 Hz), 6.39 (d, *J*=8.2 Hz), 6.25 (d, *J*=8.4 Hz)]. The spectral patterns of both ^{1}H- and ^{13}C-nmr were similar to those of hispaglabridin B, indicating that 8-prenyl-phaseollinisoflavan is an isomer of hispaglabridin B (**3**). The analysis of mass fragmentation ions and ^{13}C-^{1}H long range COSY cross peaks was of particular help to distinguish 8-prenyl-phaseollinisoflavan from hispaglabridin B (**3**). The prominent ion peaks at 190 and 187 arising from retro Diels-Alder fragmentation of the C-ring were assigned to **7** ($C_{12}H_{14}O_2$) and **8** ($C_{12}H_{11}O_2$), respectively,



m/z 190

7



m/z 187

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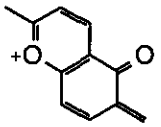

m/z 173

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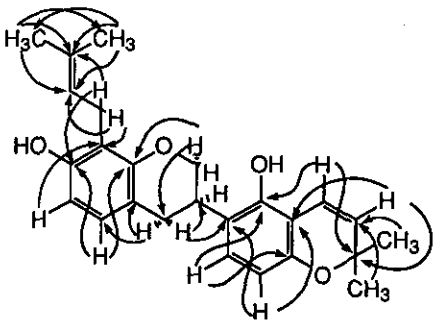

Figure 2 ^{2}J and ^{3}J Interaction Observed in the Long-Range CH COSY of 8-Prenyl-phaseollinisoflavan

indicating that the chromene occurs on the B-ring whereas the prenyl unit is substituted on the A-ring. The peak at 173 which was commonly observed as a base peak (**9**) in the mass spectra of glabridin (**1**), hispaglabridins A (**2**), B (**3**), 4'-*O*-methylglabridin (**4**), and 3'-hydroxy-4'-*O*-methylglabridin (**5b**) was missing. Since the chemical shifts of the B-ring carbons of 8-prenyl-phaseollinisoflavan were identical with those of hispaglabridin B (**3**) (Table I) but apparently differed from reported values for the B-ring carbon signals of both kanzonols O and J where the chromene ring is cyclized with 2'-OH,[20] the structure of 8-prenyl-phaseollinisoflavan was assigned as formula (**6**). The long range ^{13}C-^{1}H and ^{13}C-^{1}H COSY spectral analysis further substantiated its structure as indicated in Figure 2. The stereochemistry of naturally occurring isoflavans at C-3 has been based on either ord (optical rotatory dispersion) or cd (circular dichromism) analysis. The cd spectrum of 8-prenyl-phaseollinisoflavan (**6**) showed a negative Cotton effect at 275 nm. Glabridin (**1**), hispaglabridin A (**2**), 4'-*O*-methylglabridin (**4**) and 3'-hydroxy-4'-*O*-methylglabridin (**5b**) were reported to exhibit a positive Cotton effect in the 270-290 nm region and

were assigned as 3*R*.[1,2,18] However, hispaglabridin B (**3**) showed a negative Cotton effect in the same region although its structure was elucidated by the chemical correlation with glabridin (**1**) thus indicating that it has the same stereochemistry as other isoflavans. Similar examples with compounds having the chromene in the B-ring were also reported by Nomura *et al.*[20,21] This disparity might be expained as follows; the presence of a chromene ring modified the direction of the electron transition moment, and thus the chiral exciton coupling between A- and B-ring chromophore was influenced to produce the reversed Davydov-split. At this moment the absolute configuration at C-3 of 8-prenyl-phaseollinisoflavan could not be determined unequivocally. However, it should be noted that 8-prenyl-phaseollinisoflavan (**6**) showed the opposite Cotton effect to that of phaseollinisoflavan, an isoflavan previously obtained from the root of Spanish variety of *G. glabra*.[2]

EXPERIMENTAL

All melting points were measured on a Yanagimoto melting point apparatus and are uncorrected. Spectral data were obtained using the following apparatus: proton and carbon-13 nuclear magnetic resonance (^{1}H- and ^{13}C-nmr) spectra with a JEOL JNM GSX-400 (^{1}H, 400 MHz; ^{13}C, 100 MHz) spectrometer with tetramethylsilane (TMS) as an internal standard; mass spectra (ms) with a JEOL JMS-SX-102A mass spectrometer; infrared (ir) spectra with a JASCO FT/IR-8000 infrared spectrometer; ultraviolet (uv) spectra with a Shimadzu UV-240 spectrometer; optical rotations with a JASCO DIP-370 polarimeter; cd spectra with a JASCO J-720 spectropolarimeter. Column chromatography was carried out with the following materials: Wakogel C-200 or C-300 (eluted with benzene-acetone, chloroform-acetone or hexane-ethyl acetate), Sephadex LH-20 (Pharmacia, eluted with MeOH or MeOH-$CHCl_3$) and RP-8 reversed-phase silica gel (Merck, column size: 3.2 x 33 cm; eluted with MeOH-H_2O). Thin-layer chromatography (tlc) was conducted on a 0.25 mm pre-coated silica gel plate ($60GF_{254}$, Merck), and spots were detected by inspection under short (254 nm) or long (360 nm) wavelength uv lights, or by the colors developed with 10% H_2SO_4 spraying followed by heating on a hot plate.

Plant Material Commercially available licorice roots derived from *Glycyrrhiza glabra* were obtained from central Asian countries through Maruzen Pharmaceuticals Co., Ltd., Onomichi, Japan.

Extraction and Isolation The crushed *G. glabra* root (6 kg) was extracted with CH_2Cl_2 at room temperature for 48 h, and the extract was evaporated to dryness under reduced pressure to yield a reddish brown powder (310 g). A portion of the residue (150 g) was dissolved in acetone (*ca.* 1 *l*) and adsorbed on silica gel powder (150 g). The adsorbed material was transferred to a silica gel column (600 g; column size: 10 x 15 cm) packed in $CHCl_3$. The column was eluted with a gradient solvent system of $CHCl_3$-acetone (C-A), increasing the amount of acetone stepwise to give a number of fractions, which were then combined into eight fractions (fr. I~fr. VIII) on the basis of their tlc patterns: (i) with 100% $CHCl_3$, fr. I (30.3 g), fr. II (12.0 g); (ii) with 97:3 C-A, fr.III (51.0 g); (iii) with 95:5 C-A, fr. IV (17.7 g); (iv) with 93:7~91:9 C-A, fr. V (5.6 g); (v) with 88:12 C-A, fr. VI (11.7 g); (vi) with 84:16~80:20 C-A, fr. VII (17.1 g); (vii) with 67:33~0:100 C-A, fr. VIII (21.0 g). Fractions of 2.2 *l* each were collected. Fr. I was further chromatographed over silica gel (600 g; column size: 8 x 24 cm) and the column was eluted with the following solvent system: benzene, 800 m*l*; benzene-acetone (B-A) (99:1), 1.6 *l*; B-A

(98:2), 1.6 *l*; B-A (97:3), 1.6 *l*; B-A (95:5), 1.6 *l*; B-A (93:7), 1.6 *l*; B-A (90:10), 1.6 *l*; acetone, 1.2 *l*. Twenty-six fractions (400 m*l* each/fr.) were collected, and combined based on tlc patterns as follows: fr. A (0.7 g); fr. B (0.7 g); fr. C (3.3 g); fr. D (4.1 g); fr. E (4.0 g); fr. F (6.7 g); fr. G (3.9 g); fr. H (3.7 g); fr. I (5.1 g); fr. J. Fr. C was subjected to silica gel chromatography on elution with $CHCl_3$-benzene (3:1). Thirty fractions were collected, and fr. 5-7, fr. 9-12 and fr. 15-29 were combined. The latter two combined fractions were rechromatographed successively over Sephadex LH-20 and RP-8 reversed-phase silica gel to afford pure hispaglabridins A (**2**; 408 mg) and B (**3**; 487 mg). Fr. E was separated by elution with hexane-ethyl acetate (100:0, 97:3, 94:6, 91:9, 88:12; 84:16, 80:20; 200～400m*l* each) into twenty-four fractions (100～200 m*l*/fr.). Fractions 13-15 were combined and rechromatographed on a column of reversed-phase RP-8 silica gel to give 8-prenyl-phaseollinisoflavan (**6**; 54 mg). Combined fractions 16-22 of the same column were subjected successively to Sephadex LH-20 and reversed-phase RP-8 silica gel column chromatography to afford 3'-hydroxy-4'-*O*-methylglabridin (**5b**; 601 mg). Fr. F was separated by elution with hexane-ethyl acetate (100:0, 97:3, 94:6, 91:9, 88:12; 84:16, 80:20; 300～600 each) into twenty fractions (200～300 m*l*/1 fr.). Fractions 8-9 were combined and purified by a column of reversed-phase RP-8 silica gel to give 4'-*O*-methylglabridin (**4**; 531 mg). Combined fractions 16-18 of the same column were subjected to reversed-phase RP-8 silica gel column chromatography to afford 3'-hydroxy-4'-*O*-methylglabridin (**5b**; 340 mg). Fr. III was chromatographed over silica gel (600 g; column size: 8 x 24 cm) and the column was eluted with the following solvent system: benzene, 1 *l*; benzene-acetone (B-A) (99:1), 1 *l*; B-A (97:3), 1 *l*; B-A (95:5), 1 *l*; B-A (94:6), 1 *l*; B-A (93:7), 1 *l*; B-A (92:8), 2 *l*; B-A (91:9), 1 *l*; B-A (90:10), 1 *l*; B-A (88:12), 1 *l*; acetone, 3 *l*. Twenty-four fractions (500 m*l* each/fr.) were collected. Fractions 11-13 were combined and recrystallized from benzene to give glabridin (**1**; 4.62 g).

Glabridin (1) Colorless granules from benzene, mp 233-235°C; lit.,[18] 154-155°.[22] $[\alpha]_D^{25}$ +9.5° ($CHCl_3$, *c*=0.315). ^{1}H-Nmr (400 MHz, $CDCl_3$) δ: 7.68 (1H, br s, OH), 7.48 (1H, br s, OH), 6.91 (1H, d, *J*=8.2 Hz, 6'-H), 6.81 (1H, d, *J*=8.2 Hz, 5-H), 6.64 (1H, d, *J*=9.9 Hz, 4"-H), 6.46 (1H, d, *J*=2.2 Hz, 3'-H), 6.39 (1H, dd, *J*=8.2 Hz, 2.2 Hz, 5'-H), 6.33 (1H, d, *J*=8.2 Hz, 6-H), 5.55 (1H, d, *J*=9.9 Hz, 3"-H), 4.39 (1H, ddd, *J*=1.8 Hz, 3.2 Hz, 10.3 Hz, 2eq-H), 4.01 (1H, dd, *J*=10.3 Hz, 10.3 Hz, 2ax-H), 3.52 (1H, m, 3ax-H), 2.99 (1H, dd, *J*=11.0 Hz, 15.7 Hz, 4ax-H), 2.83 (1H, ddd, *J*=1.8 Hz, 5.3 Hz, 15.7 Hz, 4eq-H), 1.42, 1.40 (3H each, s, 5"- and 6"-CH_3). ^{13}C-Nmr (100 MHz, $CDCl_3$) δ: see Table I. EI-ms *m/z* (rel. int., %): 324 (M^+, 31), 309 (M^+-CH_3, 100), 187 (14), 173 (52), 136 (8). *Anal.* Calcd for $C_{20}H_{20}O_4$: C, 74.06; H, 6.21. Found: C, 74.24; H, 6.27.

Hispaglabridin A (2) Colorless needles from cyclohexane, mp 132-133°C; lit.,[2] 132-133°. $[\alpha]_D^{25}$ -8.0° ($CHCl_3$, *c*=0.174). ^{1}H-Nmr (400 MHz, $CDCl_3$) δ: 6.80 (1H, d, *J*=8.1 Hz, 6'-H), 6.79 (1H, d, *J*=8.4 Hz, 5-H), 6.64 (1H, d, *J*=10.0 Hz, 4"-H), 6.36 (1H, d, *J*=8.1 Hz, 5'-H), 6.33 (1H, d, J=8.4 Hz, 6-H), 5.54 (1H, d, J=10.0 Hz, 3"-H), 5.24 (1H, m, 2"'-H), 5.23 (2H, br s, OH), 4.36 (1H, ddd, *J*=1.5 Hz, 3.4 Hz, 10.2 Hz, 2eq-H), 3.97 (1H, dd, *J*=10.2 Hz, 10.2 Hz, 2ax-H), 3.47 (1H, m, 3ax-H), 3.43 (2H, d, *J*=7.2 Hz, 1"'-H_2), 2.93 (1H, dd, *J*=10.8 Hz, 15.7 Hz, 4ax-H), 2.82 (1H, ddd, *J*=1.5 Hz, 5.2 Hz, 15.7 Hz, 4eq-H), 1.83 (3H, s, 5"'-CH_3), 1.77 (3H, s, 4"'-CH_3), 1.42, 1.40 (3H each, s, 5"- and 6"-H). ^{13}C-Nmr (100 MHz, $CDCl_3$) δ: see Table I. EI-ms *m/z*: 392 (M^+, 32), 377 (M^+-CH_3, 100), 189 (20), 187 (25), 174 (14), 173 (67). *Anal.*

Calcd for $C_{25}H_{28}O_4$: C, 76.50; H, 7.19. Found: C, 76.66; H, 7.13.

Hispaglabridin B (3) Colorless plates from MeOH-H_2O, mp 84-86°C; lit.,[2] amorphous. $[\alpha]_D^{25}$ -32.4° ($CHCl_3$, *c*=0.161). ^{1}H-Nmr (400 MHz, $CDCl_3$) δ: 6.82 (1H, d, *J*=8.2 Hz, 5-H), 6.78 (1H, d, *J*=8.6 Hz, 6'-H), 6.66 (1H, d, *J*=9.8 Hz, 4'''-H), 6.65 (1H, d, *J*=9.8 Hz, 4"-H), 6.37 (1H, d, J=8.2 Hz, 6-H), 6.29 (1H, d, *J*=8.6 Hz, 5'-H), 5.64 (1H, br s, 2'-OH), 5.60 (1H, d, *J*=9.8 Hz, 3"-H), 5.56 (1H, d, *J*=9.8 Hz, 3'''-H), 4.33 (1H, ddd, *J*=1.8 Hz, 3.5 Hz, 10.1 Hz, 2eq-H), 4.02 (1H, dd, *J*=10.1 Hz, 10.1 Hz, 2ax-H), 3.50 (1H, m, 3ax-H), 2.95 (1H, dd, *J*=11.0 Hz, 15.6 Hz, 4ax-H), 2.80 (1H, ddd, *J*=1.8 Hz, 5.2 Hz, 15.6 Hz, 4eq-H), 1.43, 1.41 (6H each, s, 5"-, 5'''-, 6"- and 6'''-CH_3). ^{13}C-Nmr (100 MHz, $CDCl_3$) δ: see Table I. EI-ms *m/z* (rel. int., %): 390 (M^+, 27), 375 (M^+-CH_3, 100), 201 (7), 189 (16), 187 (52), 180 (31), 174 (9), 173 (55). *Anal.* Calcd for $C_{25}H_{26}O_4$: C, 76.90; H, 6.71. Found: C, 76.73; H, 6.65.

4'-*O*-Methylglabridin (4) Colorless needles from cyclohexane, mp 125-126°C; lit.,[2] 120-121°. $[\alpha]_D^{25}$ +7.0° ($CHCl_3$, *c*=0.187). ^{1}H-Nmr (400 MHz, $CDCl_3$) δ: 7.00 (1H, d, *J*=8.5 Hz, 6'-H), 6.82 (1H, d, *J*=8.2 Hz, 5-H), 6.65 (1H, d, *J*=9.9 Hz, 4"-H), 6.46 (1H, dd, J=8.5 Hz, 2.4 Hz, 5'-H), 6.36 (1H, d, J=8.2 Hz, 6-H), 6.34 (1H, d, J=2.4 Hz, 3'-H), 5.55 (1H, d, J=9.9 Hz, 3"-H), 5.37 (1H, br s, 2'-OH), 4.37 (1H, ddd, *J*=1.8 Hz, 3.2 Hz, 10.2 Hz, 2eq-H), 4.02 (1H, dd, *J*=10.2 Hz, 10.2 Hz, 2ax-H), 3.74 (3H, s, 4'-OCH_3), 3.49 (1H, m, 3ax-H), 2.98 (1H, dd, *J*=11.0 Hz, 15.6 Hz, 4ax-H), 2.85 (1H, ddd, *J*=1.8 Hz, 5.3 Hz, 15.6 Hz, 4eq-H), 1.43, 1.41 (3H each, s, 5"- and 6"-CH_3). ^{13}C-Nmr (100 MHz, $CDCl_3$) δ: see Table I. EI-ms *m/z* (rel. int., %): 338 (M^+, 26), 323 (M^+-CH_3, 100), 189 (8), 187 (21), 174 (10), 173 (74), 150 (15). *Anal.* Calcd for $C_{21}H_{22}O_4$: C, 74.54; H, 6.55. Found: C, 74.33; H, 6.50.

3'-Hydroxy-4'-*O*-methylglabridin (5b) Colorless needles from cyclohexane, mp 104-105°C; lit.,[2] 104-105°. $[\alpha]_D^{25}$ +10.9° ($CHCl_3$, *c*=0.183). Uv λ_{max}^{MeOH} nm (log ε): 279 (3.99), 288 (3.90), 308sh (3.37). Ir ν_{max}^{KBr} cm^{-1} : 3461, 2977, 2932, 2845, 1632, 1584, 1510, 1480, 1373, 1329, 1289, 1237, 1215, 1156, 1090. ^{1}H-Nmr (400 MHz, $CDCl_3$) δ: 6.82 (1H, d, *J*=8.2 Hz, 5-H), 6.65 (1H, d, *J*=9.9 Hz, 4"-H), 6.61 (1H, d, *J*=8.5 Hz, 6'-H), 6.43 (1H, d, *J*=8.5 Hz, 5'-H), 6.36 (1H, d, *J*=8.2 Hz, 6-H), 5.55 (1H, d, *J*=9.9 Hz, 3"-H), 5.86, 5.82 (1H each, br s, OH), 4.39 (1H, ddd, *J*=1.7 Hz, 3.4 Hz, 10.4 Hz, 2eq-H), 4.05 (1H, dd, *J*=10.4 Hz, 10.4 Hz, 2ax-H), 3.85 (3H, s, 4'-OCH_3), 3.55 (1H, m, 3ax-H), 3.00 (1H, dd, *J*=10.7 Hz, 15.9 Hz, 4ax-H), 2.86 (1H, ddd, *J*=1.7 Hz, 5.2 Hz, 15.9 Hz, 4eq-H), 1.42, 1.40 (3H each, s, 5"- and 6"-CH_3). ^{13}C-Nmr (100 MHz, $CDCl_3$) δ: see Table I. EI-ms *m/z* (rel. int., %): 354 (M^+, 27), 339 (M^+-CH_3, 100), 189 (7), 187 (14), 174 (8), 173 (50), 153 (9). *Anal.* Calcd for $C_{21}H_{22}O_5$: C, 71.17;H, 6.26. Found: C, 71.30; H, 6.29.

8-Prenyl-phaseollinisoflavan (6) Colorless plates from MeOH-H_2O, mp 151-152°C. $[\alpha]_D^{25}$ -7.6° ($CHCl_3$, *c*=2.047).Uv λ_{max}^{MeOH} nm (log ε): 278 (4.07), 285sh (4.05), 310 (3.54). Ir ν_{max}^{KBr} cm^{-1} : 3422, 3355, 2980, 2963, 2940, 2913, 2867, 1630, 1605, 1485, 1451, 1433, 1358, 1298, 1238, 1208, 1188, 1169, 1088. ^{1}H-Nmr (400 MHz, $CDCl_3$) δ: 6.80 (1H, d, *J*=8.2 Hz, 5-H), 6.76 (1H, d, *J*=8.4 Hz, 6'-H), 6.63 (1H, d, *J*=9.9 Hz, 4"-H), 6.39 (1H, d, *J*=8.2 Hz, 6-H), 6.25 (1H, d, *J*=8.4 Hz, 5'-H), 5.59 (1H, d, *J*=9.9 Hz, 3"-H), 5.27 (1H, m, 2'''-H), 5.37 (2H, br s, OH), 4.32 (1H, ddd, *J*=2.1 Hz, 3.2 Hz, 10.3 Hz, 2eq-H), 4.01 (1H, dd, *J*=10.3 Hz, 10.3 Hz, 2ax-H), 3.48 (1H, m, 3ax-H), 3.40 (2H, d, *J*=7.2 Hz, 1'''-H_2), 2.95 (1H, dd, *J*=11.0 Hz, 15.7 Hz, 4ax-H), 2.83 (1H, ddd, *J*=2.1 Hz, 5.1 Hz, 15.7 Hz, 4eq-H), 1.80 (3H, s, 5'''-CH_3), 1.73 (3H, s, 4'''-CH_3), 1.42, 1.40 (3H each, s, 5"- and 6"-CH_3). ^{13}C-Nmr (100 MHz, $CDCl_3$) δ: see Table

I. EI-ms *m/z* (rel. int., %): 392 (M^+, 73), 377 (M^+-CH_3, 62), 202 (16), 191 (35), 190 (40), 187 (100), 176 (30), 162 (25), 149 (26). Cd (c=0.000019, MeOH): $[\theta]_{290}$ 0, $[\theta]_{275}$ -3,450, $[\theta]_{260}$ -1,910, $[\theta]_{250}$ -990. *Anal.* Calcd for $C_{25}H_{28}O_4$: C, 76.50; H, 7.19. Found: C, 76.77; H, 7.13.

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22. The melting point of glabridin in the original literature (ref. 18) might have been described erroneously since the mixed melting point of compound (**1**) and the authentic sample, preserved at Faculty of Pharmaceutical Sciences, University of Tokyo, did not depress.

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