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Chemical Studies on the Constituents of the Thymelaeaceous Plants. I. Structures of Two New Flavans from *Daphne odora* THUNB.

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Three new flavans, daphnodorin A(**1**), daphnodorin B(**2**) and daphnodorin C(**3**), as well as four known coumarins, daphnetin, daphnoretin, umbelliferone and daphneticin, were isolated from the root and the bark of *Daphne odora* THUNB. The plane structures of **1** and **2** were established by chemical and spectroscopic means.

Keywords——*Daphne odora*; Thymelaeaceae; flavan; daphnodorin A; daphnodorin B

The flowers of *Daphne odora* THUNB. have been used to treat sore throat, toothache and neuralgic pain, while its roots have been used to treat stomach ache, bruises and bites by venomous snakes, and its leaves to treat abcesses and neuralgic pain, in traditional Chinese medicine.[1] As constituents of this plant, luteolin, apigenin, daphnetin, daphnin (daphnetin-7-glucoside), daphnetin-8-glucoside and umbelliferone from the flowers, daphnetin and umbelliferone from the leaves, daphnetin from the branches, and daphneolone from the roots have been obtained.[2]

Recently, we investigated this plant in order to study its phenolics.[3] In the present work, we isolated three new flavans, daphnodorin A (**1**), daphnodorin B (**2**) and daphnodorin C(**3**), were isolated together with four known coumarins, daphnetin (**4**), daphnoretin (**5**), umbelliferone (**6**) and daphneticin (**7**), from the root and bark by repeated silica gel and Sephadex LH-20 column chromatography of the crude flavan fractions as described in the experimental section. This paper deals with the isolation of the above compounds, and the structure elucidation of two of the new compounds **1** and **2**.

Compound **1**, $C_{30}H_{22}O_9$, a yellow crystalline powder, mp 185—186 °C, $[\alpha]_{589}^{22}$: $-63.16°$ ($c=0.19$ dioxane), gave a greenish-brown coloration with Gibb's reagent and a dark brown coloration with $FeCl_3$ reagent. The ultraviolet (UV) spectrum of **1** showed the absorption maxima at 217 and 307 nm, and the infrared (IR) spectrum of **1** showed the absorption bands at 3250, 1615 and 1510 cm^{-1}. The proton nuclear magnetic resonance (^{1}H-NMR) spectrum of **1** (δ, DMSO-d_6) (Table I) showed two pairs of A_2X_2 signals, suggesting the presence of two 4-oxyphenyl groups at 7.39, 6.78 ($J_{AX}=8.8$ Hz) and 6.85, 6.60 ($J_{AX}=8.8$ Hz), the couplings were found through decoupling experiments. Further, the ^{1}H-NMR spectrum exhibited signals due to protons of a 2,4,6-trioxyphenyl(2,4,6-TOP) group at 5.74 (2H, s), signals due to protons at the C-6(or C-8), C-2, C-3 and C-4 positions of a 2,8(or 6)-disubstituted 5,7-dioxy-3,4-dihydrobenzopyran ring at 6.58 (1H, s), 4.86 (1H, br d, $J=9.4$ Hz), 2.64 (2H, m), 2.22 (1H, m) and 1.69 (1H, m), and signals due to six phenolic hydroxyl groups at 9.27—12.42. The carbon-13 nuclear magnetic resonance (^{13}C-NMR) spectrum of **1** (δ, DMSO-d_6) (Table II) showed signals arising from twenty seven sp^2 carbons [CO, $=\overset{|}{C}\text{–O}\times 9$, $=\overset{|}{C}\text{–}\times 6$, $=\text{CH–}\times 11$] and three sp^3 carbons [$\text{–CH}_2\text{–}\times 2$, –CH–O], all of which could be assigned to the partial

TABLE I. ^{1}H-NMR Data (in DMSO-d_6) for Daphnodorin A, B and Their Derivatives

Carbon	Chemical shifts (J)					
	1	**2**	**8**	**9**	**15**	**16**
2	4.86 d 9.4	4.57 d 7.4	4.85 d 7.9	4.99 dd 2.4, 7.4	4.53 d 7.5	4.75 d 6.0
3	2.22 m	3.73 m	2.18 m	2.09 m	3.82 m	3.97 m
	1.69 m		1.78 m	1.94 m		
4	2.64 m	2.74 dd 16.5, 5.5	2.69 m	2.55 m	2.55 dd 16.5, 5.1	2.60 dd 16.5, 5.1
	2.64 m	2.50 dd 16.5, 8.5	2.69 m	2.55 m	2.49 dd 16.5, 5.1	2.52 dd 16.5, 5.1
6	6.58 s	6.59 s	6.85 s	6.83 s	6.87 s	6.78 s
2′ and 6′	7.39 m$^{a)}$ 8.8$^{m)}$	7.39 m$^{c)}$ 8.8$^{m)}$	7.52 m$^{e)}$ 8.8$^{m)}$	7.52 m$^{g)}$ 8.6$^{m)}$	7.52 m$^{i)}$ 8.8$^{m)}$	7.58 m$^{k)}$ 8.8$^{m)}$
3′ and 5′	6.78 m$^{b)}$ 8.8$^{m)}$	6.78 m$^{d)}$ 8.8$^{m)}$	6.95 m$^{f)}$ 8.8$^{m)}$	6.92 m$^{h)}$ 8.6$^{m)}$	6.95 m$^{j)}$ 8.8$^{m)}$	6.94 m$^{l)}$ 8.8$^{m)}$
7″	5.74 s	5.70 s	5.94 d 2.3	5.99 s	5.88 d 2.3	5.93 s
9″	5.74 s	5.70 s	5.97 d 2.3	5.99 s	5.92 d 2.3	5.93 s
12″ and 16″	6.85 m$^{a)}$ 8.8$^{m)}$	6.86 m$^{c)}$ 8.8$^{m)}$	6.98 m$^{e)}$ 8.8$^{m)}$	7.17 m$^{g)}$ 8.6$^{m)}$	6.98 m$^{i)}$ 8.8$^{m)}$	6.97 m$^{k)}$ 8.8$^{m)}$
13″ and 15″	6.60 m$^{b)}$ 8.8$^{m)}$	6.58 m$^{d)}$ 8.8$^{m)}$	6.73 m$^{f)}$ 8.8$^{m)}$	6.84 m$^{h)}$ 8.6$^{m)}$	6.73 m$^{j)}$ 8.8$^{m)}$	6.78 m$^{l)}$ 8.8$^{m)}$
3-OH		5.03 d 5.2			4.85 d 5.2	5.05 d 4.3
5	12.42 br s	12.50 br s				
4′	11.52 br s	11.90 br s				
6″ -OH	10.51 s	10.49 s				
8″	9.73 s	9.73 s				
14″	9.60 s	9.61 s				
10″-OH	9.27 s	9.23 s	13.46 s		13.30 s	
5			3.86 s	3.82 s	3.86 s	3.81 s
4′ -OMe			3.79 s	3.77 s	3.78 s	3.78 s
8″			3.77 s	3.73 s	3.77 s	3.73 s
14″			3.75 s	3.70 s	3.75 s	3.64 s
6″-OMe			3.26 s	3.46 s	3.26 s	3.46 s
10″-OMe				3.46 s		3.46 s

The spectra of **8** and **15** were measured at 70 °C and the others were measured at room temperature. *a—l*) Assignments may be reversed. *m*) J_{AX}.

structures shown in Chart 1.

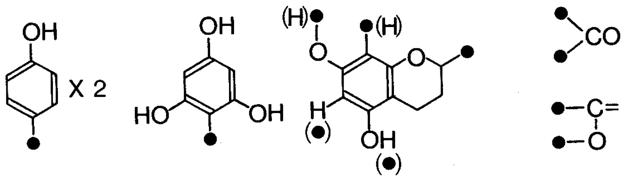

Chart 1

Table II. ^{13}C-NMR Data for **1**, **2** and **14**

Carbon	Chemical shifts (J_{CH})					
	1		**2**		**14**	
2	76.04 d	147.5	80.53 d	146.3	77.39 d	147.0
3	19.55 t	129.2	66.52 d	141.2	19.84 t	132.1
4	29.08 t	129.3	28.64 t	131.4	29.56 t	134.4
4a	104.50 s		103.43 s		105.37 s	
5	153.65 s		153.83 s		156.09 s	
6	89.30 d	163.5	89.69 d	163.5	86.72 d	162.7
7	148.25 s		147.59 s		148.37 s	
8	110.01 s		109.80 s		112.68 s	
8a	152.24 s		152.58 s		154.66 s	
1′	131.45 s		129.20 s		133.92 s	
2′, 6′	125.91 d	158.2	127.49 d	158.2	125.61 d	160.0
3′, 5′	114.65 d$^{a)}$	157.6	114.59 d$^{c)}$	157.6	113.89 d	159.0
4′	157.36 s$^{b)}$		157.58 s$^{d)}$		159.30 s	
2″	146.37 s		146.79 s		148.37 s	
3″	117.12 s		117.27 s		97.17 d	175.9
4″	194.56 s		194.57 s			
5″	105.74 s		105.95 s			
6″	165.23 s		165.53 s$^{e)}$			
7″	94.44 d	160.5	94.69 d$^{f)}$	160.5		
8″	165.87 s		165.97 s			
9″	94.44 d	160.5	94.59 d$^{f)}$	160.5		
10″	164.16 s		165.47 s$^{e)}$			
11″	121.22 s		121.39 s		124.03 s	
12″, 16″	126.48 d	159.2	126.75 d	159.2	127.40 d	157.9
13″, 15″	115.60 d$^{a)}$	158.6	115.80 d$^{c)}$	158.6	114.23 d	159.5
14″	156.12 s$^{b)}$		156.45 s$^{d)}$		159.30 s	
4′-OMe					55.33 q	
14″-OMe					55.33 q	
5-OMe					55.78 q	

a—f) Assignments may be reversed. **1** and **2** were measured in DMSO-d_6, and **14** was measured in CDCl$_3$ at room temperature.

The methylation of **1** with diazomethane–ethyl ether gave a pentamethyl ether (**8**), yellow needles, mp 201—203 °C, and a hexamethyl ether (**9**), a pale yellow viscid substance. The ^{1}H-NMR spectrum of **8** (δ, DMSO-d_6, 70 °C)$^{4)}$ (Table I) exhibited a signal due to a chelated hydroxyl group at 13.46, the signals due to five methoxyl groups including an upfield shifted signal (3.26) and one pair of coupled signals due to protons of the 2,4,6-TOP group at 5.97 and 5.94 (each 1H, d, $J=2.3$ Hz), in addition to the signals due to two 4-oxyphenyl groups and the signals due to a 5,7-dioxybenzopyran skeleton as in **1**. The upfield shifted methoxyl signal at 3.26 was assignable to the methoxyl group located at the 2,4,6-TOP moiety since the irradiation of this methoxyl signal resulted in a 19.8% nuclear Overhauser effect (NOE) enhancement of the benzene proton signal at 5.94, but no NOE enhancement of the benzene proton signal at 5.97. The electron impact mass spectrum (EI-MS) of **8** showed the M^+ ion peak at *m/z* 596 and fragment peaks at *m/z* 462 and 134, which were characterized as the retro-Diels–Alder fragment peaks of the flavan skeleton[5] in addition to a fragment peak assignable to a 2-hydroxy-4,6-dimethoxybenzoyl group at *m/z* 181. The ^{1}H-NMR spectrum of **9** was similar to that of **8** except that a signal due to two methoxyl groups was observed at 3.46 instead of a signal due to a chelated hydroxyl group at 13.46 and a signal due to a methoxyl group at 3.26. Furthermore, in the ^{1}H-NMR spectrum of **8** the signals due to protons of the 2,4,6-TOP group appeared as an AB quartet at 5.97 and 5.94, while those proton signals were observed as a singlet at 5.99 in the spectrum of **9**. In the EI-MS of **9**, the M^+ ion peak at *m/z* 610 and the fragment peaks at *m/z* 476 and 195 were higher than those of **8** by 14 mass units. Consequently, from the ^{1}H-NMR and mass spectra, it became clear that the chelated hydroxyl group in **8** was one of the hydroxyl group on the 2,4,6-trihydroxyphenyl group, and that the carbonyl group was linked to the 2,4,6-trihydroxyphenyl group.

The acetylation of **1** with acetic anhydride and sodium acetate afforded a hexaacetate (**10**), pale yellow powder, $C_{42}H_{34}O_{15}$ (*m/z* 778). The ^{1}H-NMR spectrum of **10** showed six acetyl group signals including two upfield shifted signals (1.99 and 1.68) and nonequivalent signals due to the protons of 2,4,6-TOP at 6.69 and 6.39 (each 1H, d, $J=2.3$ Hz) in addition to signals due to two 4-oxyphenyl groups and a 2,8(or 6)-disubstituted 5,7-dioxybenzopyran moiety.

From the ^{1}H-NMR spectra of **1**, **8**, **9** and **10**, it was presumed that **1** is an 8(or 6)-monosubstituted 5,7,4′-trioxyflavan derivative bearing a 4-hydroxyphenyl group and 2,4,6-trihydroxyphenyl group, and that the 2,4,6-trihydroxyphenyl group is in a magnetically shielded position. Compound **8** was decomposed upon heating in acetic acid with H_2SO_4 to give 4-methoxybenzoic acid (**11**), 3,5-dimethoxyphenol (**12**), 2-hydroxy-4,6-dimethoxybenzoic acid (**13**) and compound **14**, colorless fine needles, mp 131—133 °C. The EI-MS of **14** showed the M^+ ion peak at *m/z* 416 as well as fragment peaks at *m/z* 282 and 134, which were afforded by retro-Diels–Alder fragmentation as in the case of **8**. However, the fragment peak at *m/z* 181 due to 2-hydroxy-4,6-dimethoxybenzoyl group was absent. The UV spectrum of **14** showed absorption maxima at 221, 247, 253, 306 sh, 319 and 334 sh nm, and the IR spectrum showed the presence of an aromatic ring. The ^{1}H-NMR spectrum of **14** showed two pairs of A_2X_2 signals due to two 4-oxyphenyl groups at 7.68, 6.93 and 7.38, 6.93 (each 2H, $J_{AX}=$ 8.8 Hz), signals assignable to the protons at C-2, C-3 and C-4 of the C-ring on the flavan skeleton at 5.06 (1H, dd, $J=10.3$, 2.1 Hz), 2.85 (1H, m), 2.73 (1H, m), 2.26 (1H, m) and 2.07 (1H, m), and three methoxyl signals at 3.85 (3H, s) and 3.81 (6H, s). Furthermore, two signals having long-range coupling with $J=0.9$ Hz were observed at 6.89 and 6.66. However, the signals due to 2-hydroxy-4,6-dimethoxybenzoyl protons seen in the spectrum of **8** were absent. The ^{13}C-NMR spectrum of **14** (Table II) showed signals due to twenty sp^2 carbons [$=\overset{|}{C}\text{–O}\times 6$, $=\overset{|}{C}\text{–}\times 4$, $=\text{CH–}\times 10$] and six sp^3 carbons [$\text{–CH}_2\text{–}\times 2$, $\text{–}\overset{|}{C}\text{H–O–}$, $\text{O–CH}_3\times 3$]. From the above spectra, it was concluded that **14** is a furanoflavan derivative bearing a 4-methoxyphenyl group and that the structure is a **14a**, **14b** or **14c**.

In the case of isobergapten, a coumarin having a benzofuran moiety, the ^{1}H-NMR spectrum shows a long-range coupling ($J=1.0$ Hz) between C_6-H and β-H of the furan ring,[6] and irradiation of the C_5-OCH_3 signal (3.96) resulted in a 17.3% NOE enhancement of the C_6-H signal (6.88). In the case of bergapten, a long-range coupling ($J=1.0$ Hz) between C_8-H and β-H of the furan ring was observed, and the irradiation of C_5-OCH_3 (4.27) resulted in a 6.5% NOE enhancement of β-H of the furan ring (7.02). In the ^{1}H-NMR of **14a** long-range coupling ($J=0.9$ Hz) between C_6-H (or C_8-H) of the flavan skeleton (6.66) and β-H of the furan ring was observed, and irradiation of the C_5-OCH_3 signal (3.85) resulted in an 18.5%

NOE enhancement of C_6-H (or C_8-H) (6.66), but no NOE enhancement of β-H of the furan ring (6.89). From the above results, the structure of **14** was estimated to be **14a**; accordingly, the plane structure of **1** was represented as formula **1**.

Compound **2**, $C_{30}H_{22}O_{10}$, a yellow crystalline powder, mp 215—217 °C (dec.), $[\alpha]^{22}$ $-107.62°$ ($c=0.22$ dioxane) gave a greenish-brown coloration with Gibb's reagent and a

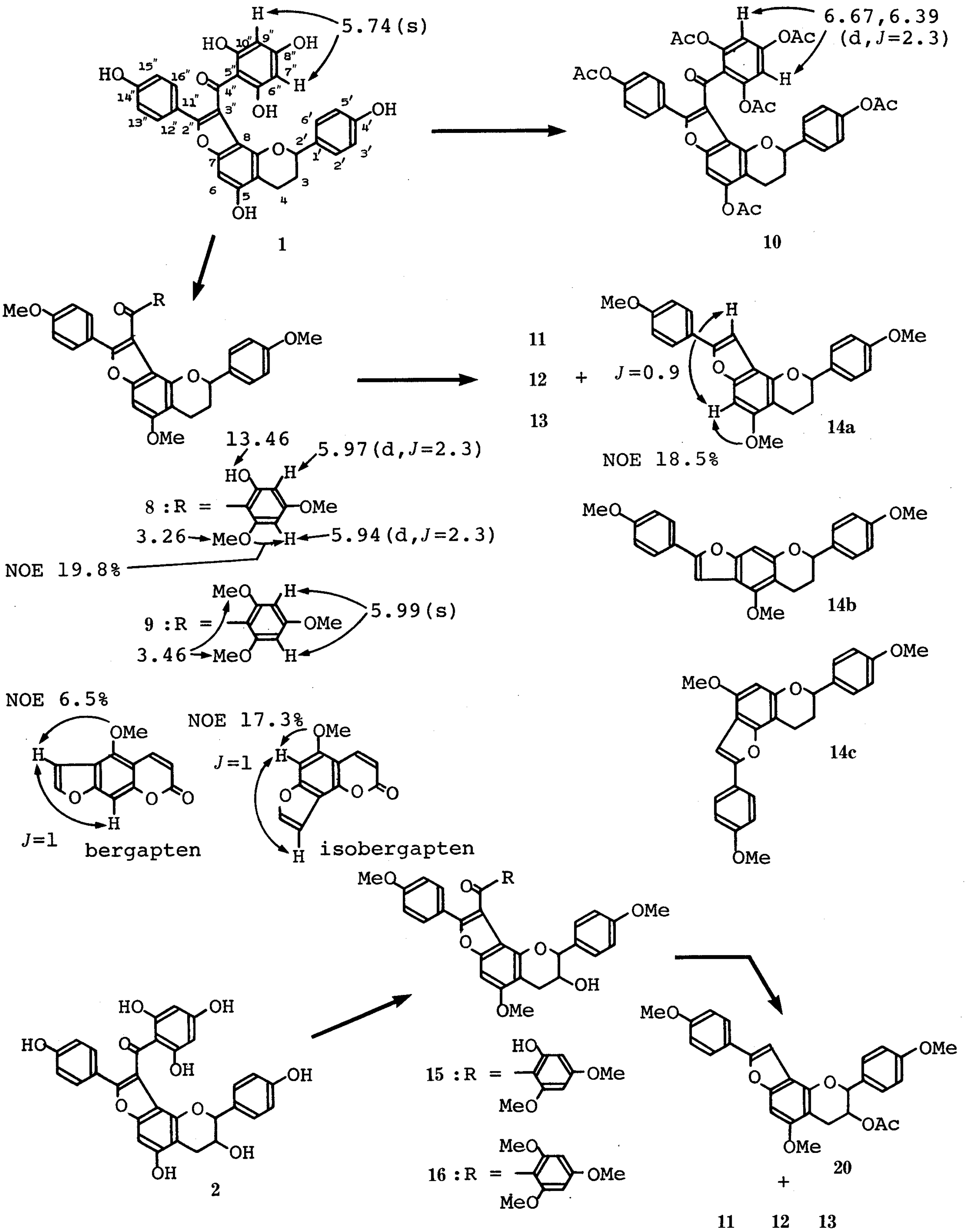

Chart 2

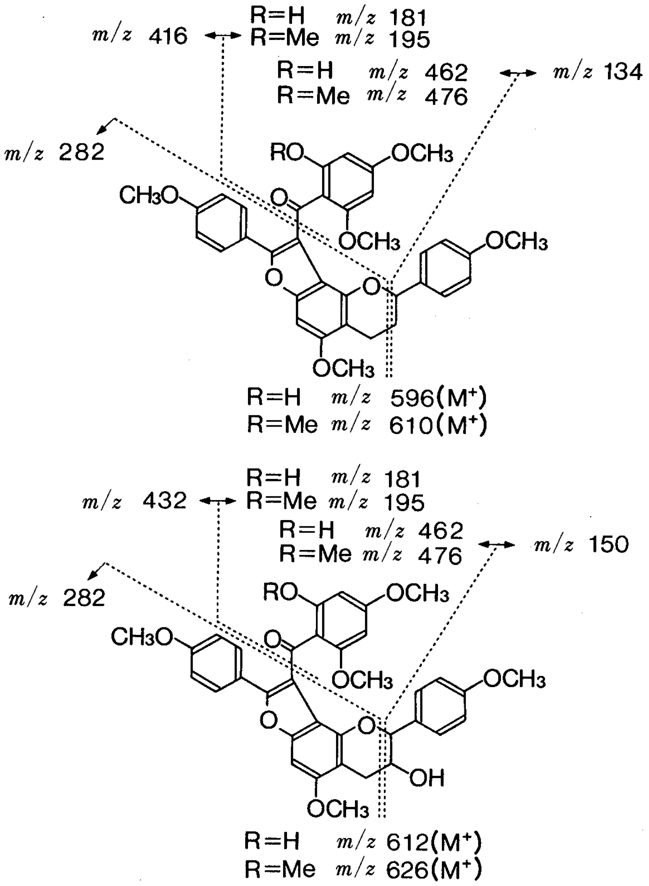

Chart 3

dark brown coloration with $FeCl_3$ reagent. The UV spectrum of **2** showed absorption maxima at 215 and 301 nm and the IR spectrum of **2** showed absorption bands at 3250, 1625, 1600 and 1500 cm^{-1}. The ^1H-NMR spectrum of **2** was very similar to that of **1** except that signals due to a 3-hydroxy-3,4-dihydrobenzopyran ring at 4.57 (1H, d, $J=7.4$ Hz), 3.73 (1H, m), 5.03 (1H, d, $J=5.2$ Hz), 2.74 (1H, dd, $J=16.5$, 5.5 Hz) and 2.50 (1H, dd, $J=16.5$, 8.5 Hz) were observed instead of the signals due to the 3,4-dihydrobenzopyran ring. The ^{13}C-NMR spectra of **1** and **2** (Table II) were nearly superimposable except that the latter showed a signal due to methine carbon at 66.52 (d, $J_{CH}=141.2$ Hz) instead of a signal due to methylene carbon at 19.55 (t, $J_{CH}=129.2$ Hz) in the former. The methylation of **2** with diazomethane–ether gave a pentamethyl ether (**15**) (M^+ 612) and a hexamethyl ether (**16**) (M^+ 626), which afforded a diacetate (**17**) and a monoacetate (**18**) with acetic anhydride and sodium acetate, respectively. The acetylation of **2** with acetic anhydride and sodium acetate afforded a heptaacetate (**19**) (M^+ 836). The ^1H-NMR spectra of **15** and **16** (Table I) were very similar to those of **8** and **9** except for the signals due to C_3-H of the dihydrobenzopyran ring. These results indicated that **2** is a 3-hydroxy derivative of **1**. This presumption was supported by the mass spectrum (MS) of **15**, which showed the same fragment peaks at *m/z* 462, 282 and 181 as that of **8**, while its M^+ ion (*m/z* 612) and the fragment peaks at *m/z* 432 and 150 were higher than those of **8** by 16 mass units (corresponding to a hydroxyl group). Compound **15** was decomposed upon heating in acetic acid with H_2SO_4 to afford **11**, **12**, **13** and **20**, a colorless viscid oil, $[\alpha]_D^{22}$ $-40.0°$ ($c=0.30$ dioxane), M^+ 474. The ^1H-NMR spectrum of **20** was similar to that of **14** except that signals due to –CH(OAc)– appeared at 5.43 (1H, m) and 1.99 (3H, s) instead of the signals due to $-CH_2-$. Thus, **20** was decided to be the structure bearing the acetoxyl group at the C_3 position of **14**.

Based on these findings, the plane structure of **2** can be represented as formula **2**. The structure of daphnodorin C (**3**) and the absolute configurations of **1** and **2** will be reported in a subsequent paper.

Experimental

All melting points were measured on a Büchi melting point apparatus and are uncorrected. The UV spectra were recorded with a Shimadzu UV-200s spectrometer, the IR spectra with a Hitachi EPI-G2 spectrometer, and the ORD spectra with a JASCO ORD/UV-5 spectrometer. The ^{1}H-NMR spectra were taken with a Varian XL-300 (300 MHz) spectrometer with tetramethylsilane (TMS) as an internal standard, and ^{13}C-NMR spectra with a Varian XL-300 (75 MHz) spectrometer with TMS as an internal standard. Column chromatography was carried out on Merck Silica gel 60 (70—230 mesh) and Sephadex LH-20. Thin layer chromatography (TLC) was performed on Merck precoated plates (Silica gel 60 F_{254}; 0.25 mm) and the developed spots were detected under a UV lamp (253.6 and 365 nm).

Isolation of the Compounds——The dried and crushed roots of *Daphne odora* (2.8 kg) were extracted 3 times by refluxing with EtOAc (15 l) for 5 h (for each extraction). The EtOAc extract (406 g) was chromatographed on silica gel (1.8 kg) with a mixture of hexane–EtOAc, and the eluate was collected in fractions (400 ml/fraction (F)): F 1—59 (3 : 1), F 60—145 (2 : 1), F 146—415 (1 : 1) and F 416—618 (1 : 2) (figures in parentheses show the solvent ratio in v/v). F 77—145 afforded daphnetin (**4**) (253 mg). F 160—263 (5.6 g) was rechromatographed on Sephadex LH-20 with MeOH to give daphnoretin (**5**) (270 mg), daphnodorin C (**3**) (5.2 g) and daphnodorin A (**1**) (5.38 g). F 322—415 was rechromatographed on Sephadex LH-20 with MeOH to afford daphnodorin B (**2**) (1.7 g). On the other hand, the dried and crushed bark of this plant (4.4 kg) was treated in the same way as described above to afford the EtOAc extract (741 g). The EtOAc extract was chromatographed on silica gel (2.0 kg) with a mixture of hexane–EtOAc, and the eluate was collected in fractions (400 ml/F): F 1—60 (3 : 1), F 61—189 (2 : 1), F 190—485 (1 : 1). F 43—189 was rechromatographed on silica gel to give umbelliferone (**6**) (500 mg) and **4** (4.4 g). F 245—319 gave **5** (1.05 g) and the mother liquors were rechromatographed on Sephadex LH-20 with MeOH to yield **3** (29 g) and **1** (10.7 g). F 321—436 was rechromatographed on Sephadex LH-20 with MeOH to give daphneticin (**7**) (60 mg) and **2** (7.3 g).

Daphnodorin A (1)——Recrystallized from $CHCl_3$–MeOH to give a yellow crystalline powder, mp 185—186 °C (dec.), Gibb's test: greenish-brown, $FeCl_3$ test: dark brown. UV $\lambda_{\max}^{\text{dioxane}}$ nm (log ε): 217 (4.58), 307 (4.58); UV $\lambda_{\max}^{\text{EtOH}}$ nm (log ε): 209 (4.61), 305 (4.53). IR $\nu_{\max}^{\text{Nujol}}$ cm^{-1}: 3250, 1615, 1510. ORD ($c=0.19$, dioxane) $[\alpha]^{22}$ (nm): $-63.16°$ (589), $-84.21°$ (550), $-105.26°$ (500), $-168.42°$ (450), $-252.63°$ (400), $-926.36°$ (350), $-1347.37°$ t (338), $-842.11°$ (330). The ^{1}H-NMR and ^{13}C-NMR data are summarized in Tables I and II, respectively.

Methylation of 1 with CH_2N_2(Formation of 8 and 9)——A solution of **1** (2.6 g) in MeOH (10 ml) was methylated with CH_2N_2–Et_2O in the usual way. The product was purified by column chromatography on silica gel, using hexane–EtOAc (3 : 1) to afford **8** (1.39 g) and **9** (0.14 g). **8**: yellow needles, mp 201—203 °C (dec.). UV $\lambda_{\max}^{\text{dioxane}}$ nm (log ε): 230 (4.77), 282 (4.29). IR $\nu_{\max}^{\text{Nujol}}$ cm^{-1}: 2950, 1615, 1580, 1505. ORD ($c=0.34$, dioxane) $[\alpha]^{24}$ (nm): $-41.18°$ (589), $-58.82°$ (550), $-73.53°$ (500), $-123.53°$ (450), $-147.06°$ (420). MS *m/z* (rel. int.): 596 (32, M$^+$), 462 (5), 416 (13), 282 (59), 181 (20), 134 (43). *Anal.* Calcd for $C_{35}H_{32}O_9 \cdot H_2O$: C, 68.39; H, 5.58. Found: C, 68.56; H, 5.83. The ^{1}H-NMR data are summarized in Table I. **9**: yellow viscid oil. UV $\lambda_{\max}^{\text{dioxane}}$ nm (log ε): 220 (4.66), 283 (4.36). IR $\nu_{\max}^{\text{CHCl}_3}$ cm^{-1}: 1605, 1505. ORD ($c=0.63$, dioxane) $[\alpha]^{23}$ (nm): $-114.29°$ (589), $-133.33°$ (550), $-171.43°$ (500), $-247.62°$ (450), $-361.90°$ (400). MS *m/z* (rel. int.): 610 (96, M$^+$), 476 (49), 416 (8), 282 (14), 195 (100), 134 (66). The ^{1}H-NMR data are summarized in Table I.

Acetylation of 1 with Ac_2O and AcONa——A solution of **1** (200 mg) in a mixture of Ac_2O (5 ml) and AcONa (200 mg) was heated in 3 h. After the reaction mixture had been treated in the usual way, the products were purified by column chromatography on silica gel with hexane–EtOAc (2 : 1) to afford a pale yellow viscid oil (**10**) (120 mg). **10**: UV $\lambda_{\max}^{\text{dioxane}}$ nm (log ε): 216 (4.72), 287 (4.46). IR $\nu_{\max}^{\text{Nujol}}$ cm^{-1}: 1760, 1655, 1605 1500. ORD ($c=0.49$, dioxane) $[\alpha]^{23}$ (nm): $-24.5°$ (589), $-30.6°$ (550), $-42.9°$ (500), $-55.1°$ (450), $-6.1°$ (400). MS *m/z*: 778 (M$^+$). ^{1}H-NMR ($CDCl_3$) δ: 7.85, 7.14 (each 2H, m, $J_{AX}=8.8$ Hz), 7.17, 7.00 (each 2H, m, $J_{AX}=8.8$ Hz), 6.94 (1H, s), 6.69, 6.39 (each 1H, d, $J=2.3$ Hz), 4.73 (1H, dd, $J=2.3$, 10.0 Hz), 2.66 (2H, m), 2.32, 2.20 (each 1H, m), 2.33, 2.32, 2.31, 2.27, 1.99, 1.68 (each 3H, s).

Cleavage of 8 with H_2SO_4/AcOH; Formation of 11, 12, 13 and 14——**8** (520 mg) was dissolved in AcOH (20 ml) and 5 drops of H_2SO_4 were added. After stirring, the mixture was heated on a boiling water bath for 5 min, then diluted with ice water (100 ml) and extracted with EtOAc. The EtOAc solution was treated with 5% $NaHCO_3$, washed with water, dried and concentrated *in vacuo*. The residue was purified by preparative layer chromatography (PLC) (hexane–EtOAc = 2 : 1) to afford **11** (82 mg), **12** (45 mg), **13** (32 mg) and **14** (60 mg). The ^{1}H-NMR spectra of **11**, **12** and **13** were identical with those of authentic samples of 4-methoxybenzoic acid, 3,5-dimethoxyphenol and 2-hydroxy-4,6-dimethoxybenzoic acid, respectively. **14**: colorless fine needles, mp 131—133 °C. UV $\lambda_{\max}^{\text{dioxane}}$ nm (log ε): 334 (4.29), 319 (4.47), 306 sh (4.40), 256 (4.00), 247 (4.01), 221 (4.38). IR $\nu_{\max}^{\text{CHCl}_3}$ cm^{-1}: 3400, 1610, 1505, 1460. Optically inactive (700—360 nm), MS *m/z* (rel. int.): 416 (28, M$^+$), 282 (100), 267 (61), 134 (35). ^{1}H-NMR ($CDCl_3$) δ: 7.68, 6.93 (each 2H, m, $J_{AX}=8.8$ Hz), 7.38, 6.93 (each 2H, m, $J_{AX}=8.8$ Hz), 6.89, 6.66 (each 1H, d, $J=0.9$ Hz), 3.85

(3H, s), 3.81 (6H, s), 5.06 (1H, dd, J=10.3, 2.1 Hz) 2.85, 2.73 (each 1H, m), 2.26, 2.07 (each 1H, m). ^{13}C-NMR data of **14** are summarized in Table II.

Daphnodorin B (2)——Recrystallized from $CHCl_3$–MeOH to give a yellow crystalline powder, mp 215—217 °C (dec.). Gibb's test: greenish-brown, $FeCl_3$ test: dark brown. UV $\lambda_{max}^{dioxane}$ nm ($\log\varepsilon$): 215 (4.57), 301 (4.57). IR ν_{max}^{Nujol} cm^{-1}: 3250 (OH), 1625 (CO), 1600, 1500. ORD (c=0.22, dioxane) $[\alpha]^{22}$ (nm): $-107.62°$ (589), $-143.50°$ (550), $-193.31°$ (500), $-269.06°$ (450), $-287.00°$ (400), $-1721.97°$ (350). The ^{1}H-NMR and ^{13}C-NMR data are summarized in Tables I and II, respectively.

Methylation of 2 with CH_2N_2 (Formation of 15 and 16)——A solution of **2** (1.56 g) in MeOH (10 ml) was methylated with CH_2N_2–Et_2O in the usual way. The product was purified by column chromatography on silica gel using hexane–EtOAc (3 : 1) to afford **15** (190 mg) and **16** (355 mg). **15**: pale yellow viscid oil. UV $\lambda_{max}^{dioxane}$ nm ($\log\varepsilon$): 218 (4.58), 303 (4.55). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1615, 1580, 1505, 1460. MS m/z (rel. int.): 612 (8, M^+), 462 (5), 432 (22), 282 (36), 181 (16), 150 (38). ORD (c=0.42, dioxane) $[\alpha]^{23}$ (nm): $-104.8°$ (589), $-133°$ (550), $-171.4°$ (500). The ^{1}H-NMR data are summarized in Table I. **16**: pale yellow viscid oil. UV $\lambda_{max}^{dioxane}$ nm ($\log\varepsilon$): 218 (4.37), 288 (4.03). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1600, 1580, 1500, 1460. MS m/z (rel. int.): 626 (M^+, 12), 476 (8), 282 (5), 195 (38), 150 (51). ORD (c= 0.42, dioxane) $[\alpha]^{23}$ (nm): $-171.4°$ (589), $-209.5°$ (550), $-276.2°$ (500), $-381.0°$ (450). ^{1}H-NMR data are summarized in Table I.

Acetylation of 15 and 16 with Ac_2O and AcONa——A solution of **15** (100 mg) in a mixture of Ac_2O (5 ml) and AcONa (100 mg) was heated for 3 h at reflux temperature. The reaction mixture was treated in the same way as for **1** to afford **17** (90 mg). **16** (100 mg) was treated in the same way as for **15** to give **18** (85 mg). **17**: pale yellow viscid oil. ^{1}H-NMR ($CDCl_3$) δ: 7.83, 6.90 (each 2H, m, J_{AX}=8.9 Hz), 7.03, 6.76 (each 2H, m, J_{AX}=8.6 Hz), 6.64 (1H, s), 6.03, 5.96 (each 1H, d, J=2.3 Hz), 5.25 (1H, m), 4.91 (1H, br d, J=4.2 Hz), 3.83, 3.82, 3.78, 3.48 (each 3H, s), 2.75 (1H, dd, J=17.1, 4.3 Hz), 2.67 (1H, dd, J=17.1, 4.8 Hz), 1.94, 1.87 (each 3H, s). **18**: pale yellow viscid oil. ^{1}H-NMR ($CDCl_3$) δ: 7.85, 6.88 (each 2H, m, J_{AX}=8.9 Hz), 7.02, 6.75 (each 2H, m, J_{AX}=8.9 Hz), 6.64 (1H, s), 5.81 (2H, s), 5.26 (1H, m), 4.92 (1H, br d, J=4.2 Hz), 3.83, 3.82, 3.77, 3.68, 3.45 (each 3H, s), 2.76 (1H, dd, J=17.1, 4.3 Hz), 2.62 (1H, dd, J= 17.1, 4.6 Hz), 1.96 (3H, s).

Acetylation of 2 with Ac_2O and AcONa——A solution of **2** (150 mg) in a mixture of Ac_2O (5 ml) and AcONa (150 mg) was heated for 3 h at reflux temperature. The reaction mixture was treated in the same way as for **1** to afford an acetate (**19**) (100 mg). **19**: colorless viscid oil, M^+ 836. UV $\lambda_{max}^{dioxane}$ nm ($\log\varepsilon$): 216 (4.79), 287 (4.56). ^{1}H-NMR ($CDCl_3$) δ: 7.90, 7.15 (each 2H, m, J_{AX}=8.8 Hz), 7.16, 6.99 (each 2H, m, J_{AX}=8.8 Hz), 7.02 (1H, s), 6.72, 6.52 (each 1H, d, J=2.1 Hz), 4.85 (1H, d, J=5.9 Hz), 5.21 (1H, m), 2.82 (1H, dd, J=16.5, 5.0 Hz), 2.66 (1H, dd, J=16.5, 6.2 Hz), 2.31 (6H, s), 2.30, 2.25, 1.94, 1.90, 1.75 (each 3H, s).

Cleavage of 15 with H_2SO_4/AcOH (Formation of 11, 12, 13 and 20)——**15** (180 mg) was dissolved in AcOH (10 ml) and 4 drops of H_2SO_4 were added. The mixture was treated in the same way as for **8** to afford **11** (43 mg), **12** (18 mg), **13** (15 mg) and **20** (12 mg). **20**: colorless viscid oil. UV $\lambda_{max}^{dioxane}$ nm ($\log\varepsilon$): 330 (4.24), 314 (4.56), 300 sh (4.30), 251 (3.91), 242 (3.91), 221 sh (4.31). ORD (c=0.30, MeOH) $[\alpha]^{23}$ (nm): $-40.0°$ (589), $-60.0°$ (550), $-73.3°$ (500), $-140.0°$ (450), $-246.7°$ (400), $-880.0°$ (350). MS m/z (rel. int.): 474 (82, M^+), 282 (100), 268 (60). ^{1}H-NMR ($CDCl_3$) δ: 7.71, 6.95 (each 2H, m, J_{AX}=8.8 Hz), 7.31, 6.87 (each 2H, m, J_{AX}=8.8 Hz), 6.93 (1H, d, J=0.7 Hz), 6.70 (1H, d, J=0.7 Hz), 3.85 (6H, s), 3.80 (3H, s), 5.43 (1H, m), 5.23 (1H, d, J=5.9 Hz), 2.93 (1H, dd, J=17.0, 5.3 Hz), 2.81 (1H, dd, J=17.0, 5.8 Hz), 1.99 (3H, s).

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