New Sterols and Triterpenoids of *Ficus pumila* Fruit

Junichi KITAJIMA,* Kaoru KIMIZUKA, and Yasuko TANAKA

Showa College of Pharmaceutical Sciences, Higashi Tamagawa Gakuen 3, Machida, Tokyo 194–8543, Japan.
Received March 18, 1998; accepted June 4, 1998

From the methanolic extract of *Ficus pumila* L. fruit (Moraceae), two new sterols, (24*S*)-stigmast-5-ene-3β,24-diol, (24*S*)-24-hydroxystigmast-4-en-3-one, two new cycloartane-type triterpenoids, (24*RS*)-3β-acetoxycycloart-25-en-24-ol, (23*Z*)-3β-acetoxycycloart-23-en-25-ol and a new euphane-type triterpenoid, (23*Z*)-3β-acetoxyeupha-7,23-dien-25-ol were obtained along with known sterols, triterpenoids and a furocoumarin (psoralene).

The structures of the new compounds were determined by spectral methods.

Key words *Ficus pumila*; euphane-type triterpenoid; cycloartane-type triterpenoid; sterol; Moraceae; fruit

The fruit of *Ficus pumila* L. (Moraceae; "ohitabi" in Japanese) has been used in Chinese folk medicine as an antitumor, antiinflammatory and tonic medicament. To date, however, no report has been published regarding the ingredients of this fruit. In this paper, we describe the separation and characterization of eighteen constituents from this fruit.

The methanolic extract of the cut fresh fruit was treated as described in Experimental, and eight fractions with psoralene (**18**) were obtained from the ether-soluble portion. From fr. 5, β-sitosterol (**1**), cholesterol (**2**), campesterol (**3**) and stigmasterol (**4**) were isolated together with a new triterpenoid **14** by recrystallization and HPLC using octadecyl silica (ODS) column. From fr. 2, lupenyl acetate (**7**) and β-amyrin acetate (**8**) were isolated, and from fr. 4, lupeol (**9**), β-amyrin (**10**), α-amyrin (**11**), taraxerol (**12**) and a new triterpenoid **13** were obtained by repeated silica gel column chromatography and HPLC using ODS column. From frs. 6 and 7, two new sterols, **5** and **6** were isolated by silica gel column chromatography and HPLC using ODS column together with triterpenoids **15**, **16** and **17**.

Sterol **5** ($C_{29}H_{50}O_2$, mp 178—180 °C, $[\alpha]_D^{23}$ −54.5°) showed the $[M+Na]^+$ ion peak at *m/z* 453 in the positive FAB-MS and $[M]^+$ ion peak at *m/z* 430 in the electron ionization (EI)-MS. The ^{1}H- (Table 1) and ^{13}C- (Table 2) and ^{13}C–^{1}H shift correlated spectroscopy (COSY) NMR spectral data revealed the presence of two *tert*-methyls, three *sec*-methyls, one *prim*-methyl, eleven methylenes, seven methines (one of them was oxygenated), three quaternary carbons (one of them was oxygenated), and one trisubstituted double bond. Accordingly, **5** was considered to be a steroid diol. As the ^{13}C-NMR spectrum of **5** showed good similarity with that of **4** except the signals of C-22 to C-29, with a downfield shift of C-24 (δ 76.02), **5** was suggested to be a 24-hydroxyl derivative of **4**. This was supported by the mass fragments which were believed to be caused by cleavage between C-22 and C-23 (*m/z* 314 $[M-C_7H_{16}O]^+$), and C-24 and C-25 (*m/z* 387 $[M-C_3H_7]^+$) in its EI-MS. Finally, **5** was identified as (24*S*)-stigmast-5-ene-3β,24-diol as reported by Catalan *et al.*[1] by the comparison of its ^{1}H-NMR data.

Sterol **6** ($C_{29}H_{48}O_2$, amorphous powder, $[\alpha]_D^{23}$ +29.1°) showed the $[M+H]^+$ ion peak at *m/z* 429 in the positive FAB-MS. The NMR spectral data (Tables 1, 2) showed that **6** was also C_{29}-sterol having an enone structure. As the ^{13}C-NMR spectrum of **6** showed good similarity with that of stigmast-4-en-3-one (**19**)[2] except the signals of C-22 to C-29,

Table 1. ^{1}H-NMR Spectral Data for **5** and **6** [δ Values (ppm) and *J* Values (Hz), 500 MHz]

	5	6
H-3	3.52 (1H, m)	
H-4		5.73 (1H, s)
H-6	5.35 (1H, d, *J*=3.0 Hz)	
H_3-18	0.68 (3H, s)	0.72 (3H, s)
H_3-19	1.01 (3H, s)	1.18 (3H, s)
H_3-21	0.94 (3H, d, *J*=6.5 Hz)	0.94 (3H, d, *J*=6.5 Hz)
H_3-26	0.888 (3H, d, *J*=7.0 Hz)	0.889 (3H, d, *J*=7.0 Hz)
H_3-27	0.892 (3H, d, *J*=7.0 Hz)	0.892 (3H, d, *J*=7.0 Hz)
H_3-29	0.85 (3H, t, *J*=7.5 Hz)	0.85 (3H, t, *J*=7.5 Hz)

Solvent: $CDCl_3$. δ in ppm from TMS [coupling constants (*J*) in Hz are given in parentheses].

Table 2. ^{13}C-NMR Spectral Data for **5** and **6** [δ Values (ppm), 125 MHz]

	5	(4)	6	(19)
C-1	37.29	(37.27)	35.70	(35.68)
C-2	31.67	(31.62)	33.99	(33.89)
C-3	71.78	(71.68)	199.68	(198.89)
C-4	42.35	(42.29)	123.76	(123.64))
C-5	140.73	(140.67)	171.68	(171.01)
C-6	121.63	(121.58)	32.95	(32.86)
C-7	31.92	(31.86)	32.04	(32.07)
C-8	31.92	(31.86)	35.62	(35.73)
C-9	50.14	(50.14)	53.79	(53.84)
C-10	36.50	(36.49)	38.60	(38.53)
C-11	21.10	(21.10)	21.03	(21.03)
C-12	39.76	(39.76)	39.61	(39.48)
C-13	42.35	(42.39)	42.42	(42.35)
C-14	56.77	(56.72)	55.87	(55.94)
C-15	24.32	(24.26)	24.18	(24.12)
C-16	28.26	(28.21)	28.23	(28.10)
C-17	55.94	(56.03)	55.90	(56.08)
C-18	11.89	(11.98)	11.95	(11.98)
C-19	19.39	(19.34)	17.83	(17.38)
C-20	36.40	(36.10)	36.39	(36.10)
C-21	18.81	(18.76)	18.72	(18.72)
C-22	29.14	(33.91)	29.05	(34.01)
C-23	31.92	(26.11)	31.80	(25.99)
C-24	76.02	(45.80)	75.99	(45.80)
C-25	33.97	(29.18)	33.93	(29.11)
C-26	16.86	(19.78)	16.84	(19.81)
C-27	16.71	(19.14)	16.68	(19.18)
C-28	28.46	(23.04)	28.44	(23.10)
C-29	7.75	(11.84)	7.73	(11.14)

Solvent: $CDCl_3$.

* To whom correspondence should be addressed.

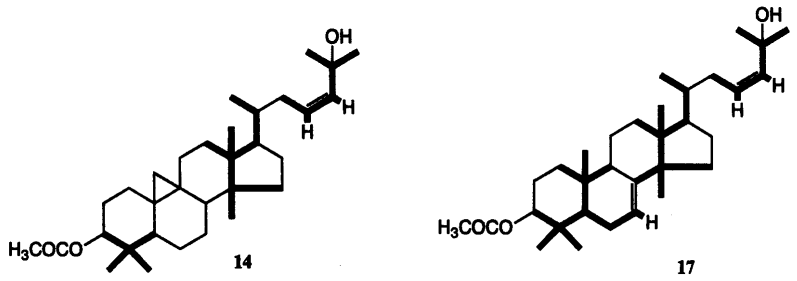

Fig. 1. Partial Structures of **14** and **17** Solved by HMBC Spectra

Table 3. ^{1}H-NMR Spectral Data for **13**, **14** and **17** [δ Values (ppm) and J Values (Hz), 500 MHz]

	13	14	17
H-3	4.57 (1H, dd, J=11.0, 5.5 Hz)	4.57 (1H, dd, J=11.0, 5.5 Hz))	4.52 (1H, dd, J=11.5, 4.0 Hz)
H-7			5.25 (1H, dd, J=6.5, 3.0 Hz)
H-9			2.23 (1H, dddd, J=6.5, 6.5, 6.0, 3.0 Hz)
H-17			1.49 (1H, m)
H_3-18	0.959, 0.962 (3H, s)	0.97 (3H, s)	0.83 (3H, s)
H_2-19	0.34 (1H, d, J=4.0 Hz)	0.34 (1H, d, J=4.0 Hz)	
	0.57 (1H, d, J=4.0 Hz)	0.58 (1H, d, J=4.0 Hz)	
H_3-19			0.77 (3H, s)
H_3-21	0.87 (3H, d, J=6.5 Hz)	0.86(3H, d, J=6.5 Hz)	0.82 (3H, d, J=6.5 Hz)
H-23		5.60 (1H, m)	5.58 (1H, m)
H-24		5.60 (1H, br d, J=7.5 Hz)	5.58 (1H, br d, J=8.0 Hz)
H_3-26	1.73 (3H, s)	1.319 (3H, s)	1.31 (3H, s)
H_3-27		1.316 (3H, s)	1.31 (3H, s)
H_2-27	4.63, 4.97 (1H, br s, 24*R*,24*S*)		
	4.63, 4.93 (1H, br s, 24*R*,24*S*)		
H_3-28	0.88 (3H, s)	0.85 (3H, s)	0.85 (3H, s)
H_3-29	0.85 (3H, s)	0.89 (3H, s)	0.93 (3H, s)
H_3-30	0.89 (3H, s)	0.89 (3H, s)	0.98 (3H, s)
OAc	2.05 (3H, s)	2.05 (3H, s)	2.06 (3H, s)

Solvent: $CDCl_3$. δ in ppm from TMS [coupling constants (J) in Hz are given in parentheses].

and the mass fragment which was thought to be caused by cleavage between C-24 and C-25 (*m/z* 385 $[M-C_3H_7]^+$) was observed in its EI-MS, **6** was suggested to be a 24-hydroxyl derivative of **19**. The configuration of C-24 in **6** was suggested to be *S* by the comparison of chemical shifts of H_3-27, H_3-28 and H_3-29 (δ 0.889, 0.892, 0.853) with those of **5** (δ 0.888, 0.892, 0.852; 24*S*) and its 24*R*-isomer (δ 0.885, 0.891, 0.863).[1] Therefore, **6** was characterized as (24*S*)-24-hydroxystigmast-4-en-3-one.

Triterpenoid **13** ($C_{32}H_{52}O_3$, mp 120—124 °C) showed the $[M]^+$, $[M-H_2O]^+$, $[M-CH_3COOH]^+$ ion peaks at *m/z* 484, 466 and 424 in the EI-MS. The ^{1}H-NMR data (Table 3) showed one acetoxyl group at δ 2.05 and two proton doublets at δ 0.34 and 0.57 due to a cyclopropane ring. The ^{13}C- (Table 4) and ^{13}C–^{1}H COSY NMR spectral data of **13** revealed the presence of five *tert*-methyls, one *sec*-methyl, eleven methylenes, six methines (two of them were oxygenated), five quaternary carbons, one *exo*-methylene and one acetoxyl group. Therefore, **13** was believed to be a cycloartane-type triterpenoid having an acetoxyl and a hydroxyl group. Comparison of the ^{13}C-NMR data with those of cycloartane-type triterpenoids[3] allowed the assignment of ^{13}C-NMR signals of **13** as shown in Table 4. The signals C-24 to C-27 appeared as a doubling peak, and this fact suggested that **13** is a mixture of C-24 epimers. The ratio of these epimers was assumed to be 1 : 1 by the signal intensity of H_3-19. Actually, the ^{13}C-NMR spectrum of **13** showed good similarity with that of a C-24 epimeric mixture of cycloart-25-ene-3β,24-diol[4] except for the presence of acetoxyl carbons and the downfield shift of C-3. Therefore, **13** was characterized as a 1 : 1 mixture of a 24-epimeric pair of 3β-acetoxycycloart-25-en-24-ol.

Triterpenoid **14** ($C_{32}H_{52}O_3$, mp 145—146 °C, $[\alpha]_D^{23}$ +44.1°) showed the $[M]^+$, $[M-H_2O]^+$, $[M-H_2O-CH_3COOH]^+$ ion peaks at *m/z* 484, 466 and 406 in the EI-MS. The NMR spectral data (Tables 3, 4) suggested that **14** also has a cycloartane structure and differs from **13** only in the side-chain structure. The analysis of heteronuclear multiple-bond correlation (HMBC) spectral data (Fig. 1; shown in heavy lines) showed that the side-chain of **14** has a 23-en-25-ol structure, and the configuration of the double bond was assigned as *Z* since the olefinic protons signal appeared as narrow multiplets with half bandwidth of 7.5 Hz in the ^{1}H-NMR spectrum. Further, the ^{13}C-NMR spectrum of **14** showed good similarity with that of (23*Z*)-cycloart-23-ene-3β,25-diol[5] except for the presence of an acetoxyl group at C-3. Therefore, **14** was characterized as (23*Z*)-3β-acetoxycycloart-23-ene-25-ol.

Triterpenoids **15** and **16** were identified as (24*RS*)-cycloart-25-ene-3β,24-diol and (23*Z*)-cycloart-23-ene-3β,25-diol, respectively, by comparison of their NMR data with published values.[5]

Triterpenoid **17** ($C_{32}H_{52}O_3$, mp 139—140 °C, $[\alpha]_D^{23}$ −3.2°) showed the $[M]^+$ ion peak at *m/z* 484 in the EI-MS. The NMR spectral data showed that **17** was a tetracyclic triterpenoid having two double bonds, one equatorial acetoxyl and

Table 4. ^{13}C-NMR Spectral Data for **13**—**16** and **17** [δ Values (ppm), 125 MHz]

	13	15	14	16	17
C-1	31.63	31.97	31.60	31.94	36.83
C-2	26.51	30.37	26.46	30.36	24.18
C-3	80.65	78.85	80.69	78.83	81.13
C-4	39.42	40.49	39.45	40.47	37.83
C-5	47.17	47.10	47.16	47.07	50.75
C-6	20.19	21.12	20.91	21.09	23.76
C-7	25.83	26.01	25.81	26.07	117.71
C-8	47.80	47.99	47.81	47.94	145.83
C-9	20.13	19.98	20.12	19.96	48.79
C-10	25.97	26.07	25.98	25.98	34.81
C-11	26.80	26.46	26.81	26.42	18.14
C-12	32.84	32.88	32.75	32.76	33.72
C-13	45.27	45.28	45.30	45.28	43.57
C-14	48.83	48.80	48.83	48.80	51.25
C-15	35.52	35.55	35.54	35.55	33.92
C-16	28.12	28.09	28.07	28.06	28.27
C-17	52.14	52.17	52.00	51.99	53.06
C-18	17.98	18.04	18.04	18.07	22.19
C-19	29.77	29.90	29.76	29.96	13.14
C-20	35.91	35.91	36.42	36.37	36.18
C-21	18.32	18.33	18.29	18.27	18.80
C-22	31.92	31.90	39.05	39.02	38.00
C-23	31.63	31.64	125.62	125.60	126.04
C-24	76.75 (24*R*)	76.36	139.36	139.31	139.12
	76.31 (24*S*)				
C-25	147.75 (24*R*)	147.47	70.76	70.75	70.74
	147.46 (24*S*)				
C-26	110.86 (24*R*)	110.91 (24*R*)	29.89	29.86	29.88
	111.35 (24*S*)	111.42 (24*S*)			
C-27	17.64 (24*R*)	17.61 (24*R*)	29.99	29.86	29.93
	17.20 (24*S*)	17.20 (24*S*)			
C-28	25.44	25.44	25.43	25.42	27.57
C-29	15.15	14.01	15.14	13.98	15.87
C-30	19.30	19.32	19.27	19.27	27.13
OAc	21.35		21.35		21.34
	171.01		171.01		171.02

Solvent: $CDCl_3$.

one *tert*-hydroxyl group. The results of HMBC spectral data (Fig. 1; shown in heavy lines) suggested that **17** was a euphane or tirucallane-type triterpenoid with double bonds at C-7 (8) and C-23 (24), equatorial acetoxyl at C-3 and *tert*-hydroxyl at C-25. This was also supported by the observed nuclear Overhauser effect (NOE) interactions between the proton signal of H-3α and H_3-28, H-9 and H_3-18, H_3-19 and H_3-29, H_3-19 and H_3-30, H_3-30 and H-17. The configuration of the side-chain double bond was found to be *Z* by the olefinic proton's signal which formed narrow multiplets with half bandwidth of 8 Hz in its ^{1}H-NMR spectrum. Further, the configuration of C-20 of **17** was confirmed as *R* by comparison of the ^{1}H chemical shift of H_3-21 with those of **14** (20*R*), eupha-7,24-diene (**19**; 20*R*) and tirucalla-7,24-diene (**20**; 20*S*).[6] That is, the chemical shift difference of H_3-21 between **17** and **14** [**17**—**14**; δ+0.04] showed good coincidence with that of **19** and **20** [**19**—**20**; δ+0.04]. From these facts, **17** was characterized as (23*Z*)-3β-acetoxyeupha-7,23-dien-25-ol. **6**, **13** and **17** are new compounds while **5** and **14** were newly isolated compounds from natural sources.

Experimental

Melting points were determined on a Yanagimoto micro melting point apparatus and are uncorrected. Optical rotations were measured on a JASCO DIP-370 digital polarimeter. MS were recorded with an HX-110 spectrometer, and in the case of FAB-MS, glycerol was used as matrix. ^{1}H- and ^{13}C-NMR spectra were taken on JEOL JNM A-500 and FX-100 spectrometers with tetramethylsilane as an internal standard, and chemical shifts are recorded in δ value. The ^{13}C–^{1}H COSY, HMBC and nuclear Overhauser enhancement spectroscopy (NOESY) spectra were obtained with the usual pulse sequence and data processing was performed with standard JEOL software. Column chromatography was carried out under TLC monitoring using Kieselgel 60 (70—230 mesh, Merck), Sephadex LH-20 (25—100 μm, Pharmacia), Lobar RP-8 column (Merck) and Amberlite XAD-II (Organo). TLC was performed on silica gel (Merck 5721) and spots were detected with *p*-anisaldehyde–H_2SO_4 reagent. HPLC separation was carried out on a JASCO chromatograph (980-system) with a JASCO 930 RI detector and ODS-3251-D [Senshu Pak; column size, 8×250 mm], Symmetry Prep C_{18} [Waters; column size, 7.8×300 mm], Megapak SIL C_{18}-10 [JASCO; column size, 7.5×250 mm] column.

Extraction and Separation of 1 to 18 *F. pumila* L. was collected at Gushikawa City in Okinawa Prefecture of Japan in March, 1994. The fresh fruit (28 kg) was extracted with methanol (32 l) at room temperature. After evaporation of the solvent, the residue (986.5 g) was suspended in water and successively extracted with ether, ethyl acetate and *n*-butanol to give an ether-soluble (43.1 g), ethyl acetate-soluble (6.5 g), *n*-butanol-soluble (35.1 g) and an aqueous (888.7 g) portion after removal of the solvents. The ether-soluble portion was extracted with hot *n*-hexane (500 ml×2) and **18** (3.4 g) was obtained by repeated recrystallization from methanol of the hot *n*-hexane insoluble portion (8.7 g). The hot *n*-hexane soluble portion (34.4 g) was chromatographed on silica gel [*n*-hexane–EtOAc (9 : 1→4 : 1→7 : 3)→EtOAc→MeOH] which furnished eight fractions (frs. 1 to 8). Fraction 2 (11.5 g) was purified by silica gel [*n*-hexane–EtOAc (9 : 1)] column chromatography and recrystallized from methanol to afford a monoacetyl triterpenoid mixture (fr. 2-1; 4.7 g). A part of fr. 2-1 (21 mg) was subjected to HPLC [ODS; CH_3CN–$CHCl_3$ (4 : 1)] to give **7** (7 mg) and **8** (14 mg). Fraction 4 (3.3 g) was subjected to repeated silica gel [*n*-hexane–EtOAc (4 : 1), benzene] column chromatography to give four fractions (fr. 4-1 to 4-4). Fraction 4-2 was subjected to silica gel [*n*-hexane–EtOAc (9 : 1), $CHCl_3$] column chromatography to give a mixture of **9**, **10**, **11** and **12** (fr. 4-2-2; 420 mg). A part of fr. 4-2-2 (50 mg) was subjected to HPLC [ODS; CH_3CN–$CHCl_3$ (4 : 1)] to give **9** (14 mg) and a mixture of **10**, **11**, **12** (fr. 4-2-2-2; 36 mg). Fraction 4-2-2-2 was recrystallized from methanol to give **10** (12 mg) and the mother liquor was acetylated; they were then separated to **11**-acetate (10 mg) and **12**-acetate (16 mg) by HPLC [ODS; CH_3CN–$CHCl_3$ (4 : 1)]. Fraction 4-4 was subjected to Sephadex LH-20 (MeOH), silica gel [*n*-hexane–EtOAc (4 : 1), $CHCl_3$] and a Lobar RP-8 [MeOH–H_2O (9 : 1)→MeOH] column chromatography to give **13** (20.6 mg). Fraction 5 (2.2 g) was recrystallized from methanol to afford **1** (712 mg) and the mother liquor was subjected to silica gel [*n*-hexane–EtOAc (4 : 1), $CHCl_3$] column chromatography to give a fraction containing a sterol mixture (fr. 5-2-2; 500 mg); this fraction was purified by HPLC (Symmetry Prep C_{18}; MeOH, 2 ml/min) to give **2** (26.1 min, 9 mg), **4** (27.1 min, 4 mg), **3** (28.5 min, 2 mg), **1** (30.0 min, 320 mg) and a fraction containing **14** (fr. 5-2-2-7; 12.4 min, 60 mg). Fraction 5-2-2-7 was purified by HPLC [Megapak SIL C_{18}-10, CH_3CN–MeOH (19 : 1)] to afford **14** (32 mg). Fraction 6 (1.8 g) was recrystallized from methanol to afford **18** (188 mg) and the mother liquor was subjected to silica gel [*n*-hexane–EtOAc (4 : 1), $CHCl_3$], a Lobar RP-8 [MeOH–H_2O (9 : 1)→MeOH] column chromatography and HPLC [ODS, MeOH–H_2O (19 : 1)] to give **17** (12 mg). Fraction 7 (1.5 g) was recrystallized from methanol to afford **18** (173 mg) and the mother liquor was subjected to silica gel [*n*-hexane–EtOAc (4 : 1→3 : 2), $CHCl_3$] to give four fractions (frs. 7-2-1 to 7-2-4). From fr. 7-2-2, **6** (6 mg) and **15** (12 mg) were obtained by HPLC (Symmetry Prep C_{18}, MeOH). From fr. 7-2-4, **5** (10 mg) and **16** (3 mg) were isolated by silica gel [$CHCl_3$, $CHCl_3$–MeOH (9 : 1)] and HPLC [Megapak SIL C_{18}-10, CH_3CN–$CHCl_3$ (9 : 1)].

The following compounds were identified by comparison with authentic compounds.

β-Sitosterol (**1**), cholesterol (**2**), campesterol (**3**), stigmasterol (**4**), lupenyl acetate (**7**), β-amyrin acetate (**8**; **10**-acetate), lupeol (**9**), α-amyrin acetate (**11**-acetate), taraxerol (**12**), psoralene (**18**).

(24*S*)-Stigmast-5-ene-3β,24-diol (5) Colorless needles, mp 178—180 °C, $[\alpha]_D^{23}$ −54.5° (*c*=0.8, $CHCl_3$), [ref. 1; mp 178.5—180 °C, $[\alpha]_D$ −30.7°]. Positive FAB-MS *m/z*: 453 $[M+Na]^+$. EI-MS *m/z*: 430.3753 $[M]^+$ (Calcd for $C_{29}H_{50}O_2$: 430.3808), 412.3691 $[M-H_2O]^+$ (Calcd for $C_{29}H_{48}O$: 412.3702), 387 $[M-C_3H_7]^+$, 369 $[M-C_3H_7-H_2O]^+$, 314 $[M-C_7H_{16}O]^+$ (base).

(24*S*)-24-Hydroxystigmast-4-en-3-one (6) Amorphous powder, $[\alpha]_D^{23}$ +29.1° (*c*=0.5, $CHCl_3$). Positive FAB-MS *m/z*: 429.3735 $[M+H]^+$ (base,

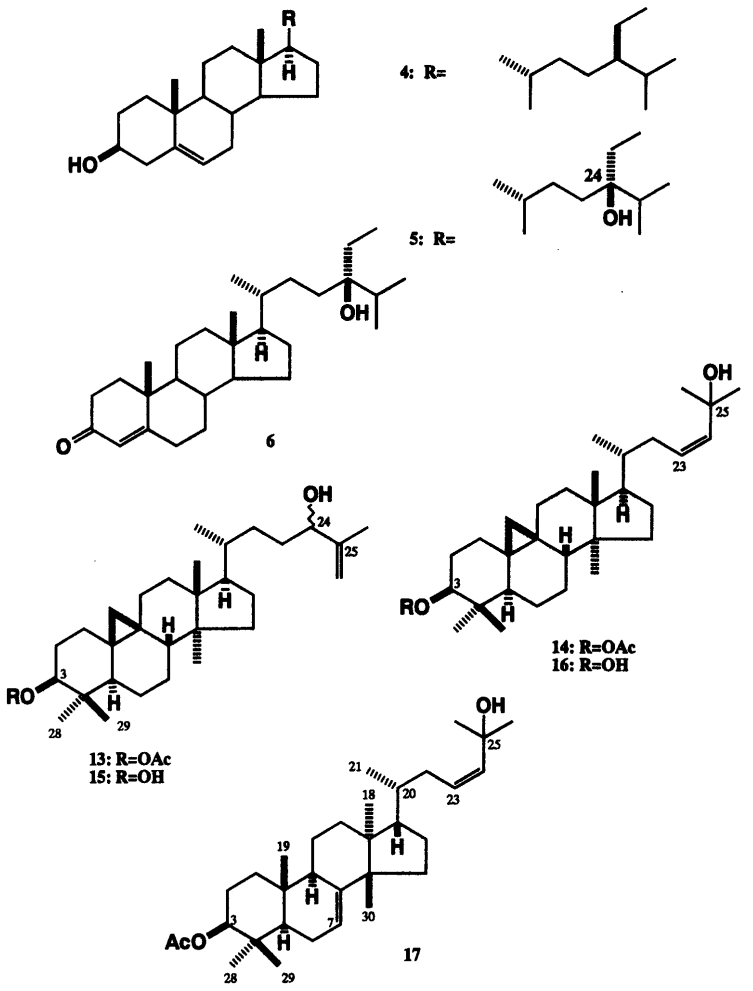

Chart 1. Structures of **4** to **6** and **13** to **17**

Calcd for $C_{29}H_{49}O_2$: 429.3732). EI-MS *m/z*: 429 $[M+H]^+$ (base), 411 $[M+H-H_2O]^+$, 385 $[M-C_3H_7]^+$, 367 $[M-C_3H_7-H_2O]^+$, 312 $[M-C_7H_{16}O]^+$.

(24*RS*)-3β-Acetoxycycloart-25-en-24-ol (13) Colorless needles, mp 120—124 °C. EI-MS *m/z*: 484.3934 [M]+ (Calcd for $C_{32}H_{52}O_3$: 484.3913), 466 $[M-H_2O]^+$, 424.3708 $[M-CH_3COOH]^+$ (base, Calcd for $C_{30}H_{48}O$: 424.3705), 409 $[M-CH_3COOH-CH_3]^+$.

(23*Z*)-3β-Acetoxycycloart-23-en-25-ol (14) Colorless needles, mp 145—146 °C. $[\alpha]_D^{23}$ +44.1° (*c*=2.3, $CHCl_3$). EI-MS *m/z*: 484.3902 $[M]^+$ (Calcd for $C_{32}H_{52}O_3$: 484.3913), 466.3875 $[M-H_2O]^+$ (Calcd for $C_{32}H_{50}O_2$: 466.3810), 424 $[M-CH_3COOH]^+$, 406 $[M-CH_3COOH-H_2O]^+$ (base), 391 $[M-CH_3COOH-H_2O-CH_3]^+$.

(24*RS*)-Cycloart-25-en-3β,24-diol (15) Amorphous powder. ^{1}H-NMR ($CDCl_3$) δ: 3.24 (1H, m, H-3), 0.97 (3H, s, H_3-18), 0.33, 0.56 (each 1H, d, *J*=4.0 Hz, H_2-19), 0.85 (3H, d, *J*=6.5 Hz, H_3-21), 4.02 (1H, t, *J*=6.5 Hz, H-24), 1.73 (3H, s, H_3-26), 4.84, 4.92 (each 1H, br s, H_2-27), 0.81 (3H, s, H_3-28), 0.97 (3H, s, H_3-29), 0.89 (3H, s, H_3-30).

(23*Z*)-Cycloart-23-en-3β,25-diol (16) Colorless needles, mp 140—141 °C. $[\alpha]_D^{23}$+23.3° (*c*=0.2, $CHCl_3$). ^{1}H-NMR ($CDCl_3$) δ: 3.31 (1H, m, H-3), 0.97 (3H, s, H_3-18), 0.33, 0.55 (each 1H, d, *J*=4.0 Hz, H_2-19), 0.93 (3H, d, *J*=6.5 Hz, H3-21), 5.59 (1H, m, H-23), 5.59 (1H. br s, *J*=8.0 Hz, H-24), 1.32 (6H, s, H_3-26, H_3-27), 0.81 (3H, s, H_3-28), 0.97 (3H, s, H_3-29), 0.88 (3H, s, H_3-30).

(23*Z*)-3β-Acetoxyeupha-7,23-dien-25-ol (17) Colorless needles, mp 139—140 °C. $[\alpha]_D^{23}$ −3.2° (*c*=0.9, $CHCl_3$). EI-MS *m/z*: 484.3911 $[M]^+$ (Calcd for $C_{32}H_{52}O_3$: 484.3913), 466.3820 $[M-H_2O]^+$ (Calcd for $C_{32}H_{50}O_2$: 466.3810), 451 $[M-CH_3-H_2O]$+ (base), 391 $[M-CH_3COOH-H_2O-CH_3]^+$.

Acknowledgements The authors thank Messrs. Y. Takase and H. Suzuki of the Central Analytical Center of Showa College of Sciences for NMR and MS measurements.

References

1) Catalan C. A. N., Kokke W. C. M. C., Carmenza D., Djerassi C., *J. Org. Chem.*, **48**, 5207—5214 (1983).
2) Greca M. D., Monaco P., Previtera L., *J. Nat. Prod.*, **53**, 1430—1435 (1990).
3) Akihisa T., Matumoto T., *Yukagaku*, **36**, 301—319 (1987); Khuong H, F., Sangare M., Chari V. M., Bekaert A., Devys M., Barbier M., Lukaecs G., *Tetrahedron Lett.*, **1975**, 1787—1790; Radics L., Peredy M. K., Corsano S., Standoli L., *ibid.*, **1975**, 4287—4290; Blunt J. W., Munro M. H. G., *Org. Magn. Reson.*, **13**, 26—29 (1980).
4) Anjaneyulu V., Prasad K. H., Ravi K., Connolly J. D., *Phytochemistry*, **24**, 2359—2367 (1985).
5) Della M., Fiorentino A., Monaco P., Previtera L., *Phytochemistry*, **35**, 1017—1022 (1994).
6) Masuda K., Shiojima K., Ageta H., *Chem. Pharm. Bull.*, **31**, 2530—2533 (1983).