Studies on Cardiac Ingredients of Plants. IX.[1]   Chemical Transformation of Proscillaridin by Utilizing Its 1,4-Cycloadducts as Key Compounds and Biological Activities of Their Derivatives

Takahiro Tanase,[a] Nobutoshi Murakami,[a] Shin-ichi Nagai,[a] Taisei Ueda,[a] Jinsaku Sakakibara,*,[a] Hiroaki Ando,[b] Yoshihiro Hotta,[b] and Kazumi Takeya[b]

Faculty of Pharmaceutical Sciences, Nagoya City University,[a] Tanabe-dori, Mizuho-ku, Nagoya 467, Japan and Department of Pharmacology, Aichi Medical University,[b] Nagakute, Aichi 480-11, Japan.   Received July 24, 1991
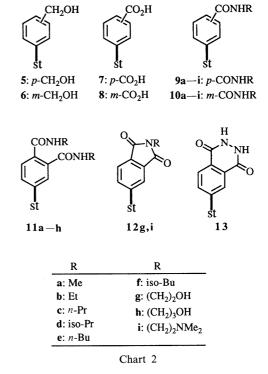
Three aromatic compounds (2—4) possessing a carbomethoxyl group or a dimethoxyphthaloyl group, prepared by the Diels–Alder reaction of the cardiac glycoside, proscillaridin (1), with dimethyl acetylenedicarboxylate and methyl propiolate, were transformed into alcohols, carboxylic acids and amides. The biological activities of the resulting derivatives were evaluated by the use of Na^+,K^+-adenosine triphosphatase (Na^+,K^+-ATPase) from dog kidney and isolated guinea-pig papillary muscle. Although the biological activities of the resulting derivatives were less potent than that of 1, a *para*-substituted benzylalcohol (5), methylbenzamides (9a and 10a), and ethylbenzamides (9b and 10b) inhibited the activity of Na^+,K^+-ATPase almost as potently as naturally occurring cardiac glycosides such as digoxin and digitoxin.

Keywords   proscillaridin; cardiac glycoside; chemical transformation; Na^+,K^+-ATPase; guinea-pig papillary muscle; positive inotropic effect

Although cardiac glycosides have been widely used in the treatment of congestive heart failure,[2] these drugs have a very narrow concentration range of positive inotropic effect (PIE) development, and sometimes causes arrhythmia.[3]

Bufadienolide glycosides such as proscillaridin and scillaren A have not been as well studied concerning their chemical transformation and the pharmacology of their derivatives as the cardenolide glycosides, (*e.g.* digitoxin, digoxin). In order to develop cardiac drugs with a higher margin of safety than clinically used cardiac glycosides by chemical modification of bufadienolide glycosides, we have been investigating chemical modification of proscillaridin (1). We previously reported that the diene portion in 1 readily undergoes the Diels–Alder reaction with methyl propiolate and dimethyl acetylenedicarboxylate (DMAD) to furnish methyl benzoate derivatives (2 and 3) and a phthalate compound (4) in one step and in high yields.[4] Thus, we have examined chemical modifications of these compounds to prepare various derivatives of proscillaridin (1). This paper describes the preparation of alcohols (5 and 6), carboxylic acids (7 and 8), and amides (9 and 10) from 1 by utilizing the methyl benzoate derivatives (2 and 3) and the dimethyl phthalate compound (4) as key intermediates, and presents their biological activities (Charts 1, and 2).

In a previous paper, we reported that the Diels–Alder adduct of proscillaridin (1) with DMAD gave the dimethyl phthalate derivative (4) in high yield and an alcohol obtained by lithium aluminum hydride (LiAlH₄) reduction of 4 showed enhanced cardiac potency in comparison with 4.[4a] Since this enhancement is assumed to be due to increased hydrophilicity, we undertook to prepare the corresponding alcohols from the methyl benzoate derivatives (2 and 3) and examine their biological activities. The LiAlH₄ reduction of 2 and 3 afforded the benzylalcohols (5 and 6) in 80%, and 76% yields, respectively. The infrared (IR) spectrum of 5 showed disappearance of the carbonyl absorption, while the signal due to a carbomethoxyl group was absent and a hydroxymethyl signal was newly observed at 4.97 ppm (2H, s) in the proton nuclear magnetic resonance (¹H-NMR) spectrum. In addition, the ¹H-NMR spectrum exhibited

5: *p*-CH₂OH
6: *m*-CH₂OH

7: *p*-CO₂H
8: *m*-CO₂H

9a—i: *p*-CONHR
10a—i: *m*-CONHR

proscillaridin (1)   2   3   4

11a—h   12g,i   13

R	R
a: Me	f: iso-Bu
b: Et	g: (CH₂)₂OH
c: *n*-Pr	h: (CH₂)₃OH
d: iso-Pr	i: (CH₂)₂NMe₂
e: *n*-Bu	

st=

Chart 1

Chart 2

the signal due to a hydroxy group at 5.10 ppm (1H, s, disappeared on addition of D_2O). Thus, the structure was confirmed to be 4-[3β-[(6-deoxy-α-L-mannopyranosyl)-oxy]-14β-hydroxyandrost-4-en-17β-yl]benzenemethanol. The physicochemical data of **6** were similar to those of **5** (see Experimental), so the structure of **6** was determined to be as depicted in Chart 2.

In order to increase further the hydrophylicity, the corresponding carboxylic acids (**7** and **8**) were prepared quantitatively by alkaline treatment (10% KOH–MeOH) of **2** and **3**. The ^{1}H-NMR spectra of **7** and **8** were very similar to those of **2** and **3** except for the lack of the signals due to carbomethoxyl groups and both IR spectra showed absorption bands attributed to a carboxyl group (3550—2550 cm^{-1}), so the structures of **7** and **8** were determined to be 4-[3β-[(6-deoxy-α-L-mannopyranosyl)oxy]-14β-hy-

droxyandrost-4-en-17β-yl]benzoic acid and 3-[3β-[(6-deoxy-α-L-mannopyranosyl)oxy]-14β-hydroxyandrost-4-en-17β-yl]benzoic acid. Furthermore, we attempted amidation of **2**, **3**, **7**, and **8** with various aliphatic amines in order to examine the correlation between dimensions of substituents and the biological activities. A mixture of **2** and 30% aqueous methylamine or ethylamine in methanol was heated at 100—110 °C in sealed tubes for 15 h or 5 d to give the corresponding amides (**9a** and **b**) in 70 and 72% yields, respectively. However, this method was inapplicable with other amines because the reaction times were too long to prepare the corresponding amides in satisfactory yields. Therefore, the amide derivatives (**9c—i** and **10c—i**) were prepared from the carboxylic acids (**7** and **8**) by the use of diphenylphosphoryl azide (DPPA).[5] A mixture of the

TABLE I. Reaction Times, Yields, and Appearances of **9a—i**, **10a—i**

Compound	Reaction time	Yield (%) 9	Yield (%) 10	Appearance (Recryst. solv.)
a	15 h[a]	70	67	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
b	5 d[a]	72	68	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
c	4.5 h[b]	95	96	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
d	5.0 h[b]	96	94	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
e	5.0 h[b]	90	92	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
f	5.0 h[b]	94	95	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
g	5.0 h[b]	91	90	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
h	4.5 h[b]	93	90	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
i	4.0 h[b]	95	92	Colorless crystalline powder (iso-Pr$_2$O–MeOH)

a) Prepared by method A.   b) Prepared by method B.

TABLE II. Reaction Times, Yields, and Appearances of **11—13**

Compound	Reaction time	Yield (%)	Appearance (Recryst. solv.)
11a	2 d	80	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
11b	3 d	52	Colorless crystalline powder (iso-Pr$_2$O–acetone)
11c	4 d	64	Colorless crystalline powder (iso-Pr$_2$O–acetone)
11d	4 d	75	Colorless crystalline powder (iso-Pr$_2$O–acetone)
11e	4 d	44	Colorless crystalline powder (iso-Pr$_2$O–acetone)
11f	3 d	40	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
11g	4 d	38	Colorless crystalline powder (iso-Pr$_2$O–acetone)
11h	4 d	80	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
12g	4 d	37	Colorless needles (iso-Pr$_2$O–acetone)
12i	4 d	47	Colorless crystalline powder (iso-Pr$_2$O–MeOH)
13	30 h	92	Colorless crystalline powder (iso-Pr$_2$O–MeOH)

TABLE III. Physicochemical Data for **9a—i**

Compound	mp (°C)	$[\alpha]_D^{25}$ (°) (c, MeOH)	IR (ν_{max}^{KBr} cm^{-1})	UV (λ_{max}^{MeOH}) nm (ε)	Formula	Analysis (%) Calcd (Found) C	Analysis (%) Calcd (Found) H	Analysis (%) Calcd (Found) N
9a	178—180	−49.5 (0.4)	3400 (OH) 1635 (CO)	242.0 (1.6 × 10^4)	C$_{33}$H$_{47}$NO$_7$·2H$_2$O	65.43 (65.51	8.49 8.34	2.31 2.48)
9b	130—133	−44.3 (0.4)	3400 (OH) 1635 (CO)	242.0 (1.7 × 10^4)	C$_{34}$H$_{49}$NO$_7$·H$_2$O	67.86 (67.82	8.54 8.52	2.33 2.36)
9c	148—150	−52.0 (0.5)	3400 (OH) 1635 (CO)	243.0 (2.2 × 10^4)	C$_{35}$H$_{51}$NO$_7$·H$_2$O	68.27 (68.18	8.67 8.67	2.27 2.21)
9d	156—157	−48.9 (0.5)	3425 (OH) 1630 (CO)	243.0 (2.0 × 10^4)	C$_{35}$H$_{51}$NO$_7$·H$_2$O	68.27 (68.16	8.67 8.58	2.27 1.97)
9e	138—140	−51.2 (0.6)	3435 (OH) 1635 (CO)	242.7 (2.1 × 10^4)	C$_{36}$H$_{53}$NO$_7$·H$_2$O	68.64 (68.69	8.81 8.79	2.22 2.25)
9f	142—144	−48.7 (0.5)	3440 (OH) 1640 (CO)	242.3 (2.4 × 10^4)	C$_{36}$H$_{53}$NO$_7$·H$_2$O	68.64 (68.49	8.81 8.90	2.22 2.32)
9g	244—246	−50.5 (0.5)	3400 (OH) 1640 (CO)	243.0 (2.0 × 10^4)	C$_{34}$H$_{49}$NO$_8$·H$_2$O	66.10 (66.25	8.32 8.00	2.27 2.04)
9h	145—147	−50.1 (0.5)	3400 (OH) 1645 (CO)	243.0 (1.7 × 10^4)	C$_{35}$H$_{51}$NO$_8$·1/2H$_2$O	67.50 (67.53	8.42 8.65	2.25 2.20)
9i	128—130	−42.2 (0.6)	3410 (OH) 1635 (CO)	241.0 (1.6 × 10^4)	C$_{36}$H$_{55}$N$_2$O$_7$·H$_2$O	66.95 (67.07	8.90 8.72	4.32 4.36)

carboxylic acid (7) and *n*-propylamine was treated with DPPA to yield the amide (9c) in 95% yield. Similarly, the condensation of carboxylic acids (7 and 8) with isopropylamine, *n*-butylamine, isobutylamine, monoethanolamine, monopropanolamine, and, *N*,*N*-dimethylethylenediamine using DPPA led to the corresponding amides (9d—i and 10d—i) in 90—96% yields. Their structures were confirmed by the appearance of amide carbonyl absorption bands instead of ester carbonyl ones in their IR spectra, as well as the other physicochemical properties as shown in Tables IV, and V. Therefore, they were established as *N*-substituted-4 or 3-[3β-[(6-deoxy-α-L-mannopyranosyl)-oxy]-14β-hydroxyandrost-4-en-17β-yl]benzamide.

The dimethyl phthalate derivative (4) was also led to corresponding diamides (11a—h) in 38—80% yields by heating with the amines in sealed tubes for 2—4 d. In contrast, the reaction of 4 with monoethanolamine or *N*,*N*-dimethylethylenediamine gave imide derivatives (12g and i) in 37 and 47% yields, respectively. In the IR spectra, the absorption bands characteristic of an imide group (1770—1710 cm^{-1}) were observed, while the ^{1}H-NMR spectra of 12g and 12i exhibited a 4H signal in the range of 3.77—3.83 ppm. Based on physicochemical data, the structures of 12g and 12i were determined to be *N*-substituted-4-[3β-[(6-deoxy-α-L-mannopyranosyl)oxy]-14β-hydroxyandrost-4-en-17β-yl]phthalimide.

TABLE IV. Physicochemical Data for 10a—i

Compound	mp (°C)	$[\alpha]_D^{25}$ (°) (c, MeOH)	IR (ν_{max}^{KBr} cm^{-1})	UV (λ_{max}^{MeOH}) nm (ε)	Formula	Analysis (%) Calcd (Found)		
						C	H	N
10a	138—140	−61.1 (0.7)	3400 (OH) 1650 (CO)	233.0 (sh) (9.0 × 10³)	$C_{33}H_{47}NO_7 \cdot 2H_2O$	65.43 (65.45	8.49 8.48	2.31 1.99)
10b	128—130	−57.9 (0.6)	3400 (OH) 1640 (CO)	234.0 (sh) (1.1 × 10⁴)	$C_{34}H_{49}NO_7 \cdot H_2O$	67.86 (67.79	8.54 8.46	2.33 2.41)
10c	145—147	−59.6 (0.6)	3410 (OH) 1635 (CO)	234.0 (sh) (9.5 × 10³)	$C_{35}H_{51}NO_7 \cdot H_2O$	68.27 (68.54	8.67 8.58	2.27 1.99)
10d	154—156	−59.1 (0.5)	3445 (OH) 1635 (CO)	235.0 (sh) (1.2 × 10⁴)	$C_{35}H_{51}NO_7 \cdot H_2O$	68.27 (68.21	8.67 8.59	2.27 2.20)
10e	135—137	−53.2 (0.5)	3430 (OH) 1640 (CO)	234.0 (sh) (9.2 × 10³)	$C_{36}H_{53}NO_7 \cdot H_2O$	68.64 (68.59	8.81 8.84	2.22 2.31)
10f	140—142	−56.7 (0.6)	3425 (OH) 1635 (CO)	234.0 (sh) (8.7 × 10³)	$C_{36}H_{53}NO_7 \cdot H_2O$	68.64 (68.58	8.81 8.93	2.22 2.21)
10g	170—172	−57.8 (0.7)	3360 (OH) 1635 (CO)	234.0 (sh) (1.1 × 10⁴)	$C_{34}H_{49}NO_8 \cdot 2H_2O$	64.23 (64.24	8.24 8.24	2.04 2.04)
10h	144—146	−58.5 (0.7)	3400 (OH) 1640 (CO)	234.0 (sh) (1.1 × 10⁴)	$C_{35}H_{51}NO_8 \cdot H_2O$	66.54 (66.40	8.46 8.28	2.22 1.94)
10i	127—129	−50.4 (0.8)	3400 (OH) 1645 (CO)	233.0 (sh) (1.1 × 10⁴)	$C_{36}H_{55}N_2O_7 \cdot H_2O$	66.95 (66.87	8.90 8.85	4.32 4.21)

TABLE V. Physicochemical Data for 11—13

Compound	mp (°C)	$[\alpha]_D^{25}$ (°) (c, MeOH)	IR (ν_{max}^{KBr} cm^{-1})	UV (λ_{max}^{MeOH}) nm (ε)	Formula	Analysis (%) Calcd (Found)		
						C	H	N
11a	252—254	−44.3 (0.5)	3400 (OH) 1645, 1630 (CO)	229.0 (1.7 × 10⁴)	$C_{35}H_{50}N_2O_8 \cdot 1/2H_2O$	66.14 (66.14	8.03 7.99	4.41 4.15)
11b	153—155	−45.1 (0.5)	3400 (OH) 1645, 1630 (CO)	228.0 (1.3 × 10⁴)	$C_{37}H_{54}N_2O_8 \cdot H_2O$	66.06 (66.09	8.33 8.11	4.17 4.20)
11c	143—145	−43.3 (0.5)	3430 (OH) 1640, 1630 (CO)	226.0 (1.5 × 10⁴)	$C_{39}H_{58}N_2O_8 \cdot H_2O$	66.86 (66.78	8.57 8.33	4.00 3.71)
11d	160—162	−42.9 (0.5)	3435 (OH) 1640 (CO)	226.0 (1.5 × 10⁴)	$C_{39}H_{58}N_2O_8 \cdot H_2O$	66.86 (66.99	8.57 8.24	4.00 3.70)
11e	136—139	−41.5 (0.5)	3420 (OH) 1640 (CO)	229.0 (1.4 × 10⁴)	$C_{41}H_{62}N_2O_8 \cdot H_2O$	67.58 (67.78	8.79 8.49	3.85 3.68)
11f	154—156	−30.3 (0.5)	3430 (OH) 1640, 1630 (CO)	227.0 (1.6 × 10⁴)	$C_{41}H_{62}N_2O_8 \cdot 1/2H_2O$	67.43 (68.41	8.76 8.75	3.89 3.82)
11g	175	−39.7 (0.5)	3415 (OH) 1635 (CO)	229.0 (1.5 × 10⁴)	$C_{37}H_{54}N_2O_8 \cdot H_2O$	63.07 (63.15	7.75 7.70	3.98 4.11)
11h	161—163	−40.7 (0.5)	3410 (OH) 1640, 1630 (CO)	229.0 (1.6 × 10⁴)	$C_{39}H_{58}N_2O_8 \cdot H_2O$	63.97 (64.24	8.20 8.16	3.83 3.64)
12g	225—226	−39.2 (0.4)	3400 (OH) 1770, 1710 (CO)	230.3 (3.7 × 10⁴) 305.0 (2.6 × 10³)	$C_{35}H_{47}NO_9 \cdot 1/2H_2O$	66.25 (66.06	7.57 7.33	2.21 2.09)
12i	139—141	−33.5 (0.5)	3445 (OH) 1765, 1700 (CO)	230.0 (3.1 × 10⁴) 306.0 (1.9 × 10³)	$C_{37}H_{52}N_2O_8 \cdot H_2O$	66.27 (66.02	8.06 7.79	4.00 3.78)
13	278—280	−42.3 (0.4)	3350 (OH) 1640 (CO)	243.0 (1.9 × 10⁴) 297.0 (6.1 × 10³)	$C_{33}H_{44}N_2O_8 \cdot H_2O$	64.50 (64.33	7.49 7.22	4.50 4.41)

Treatment of compound **4** possessing a 1,4-dicarbonyl moiety with hydrazine monohydrate afforded a pyridazinone (**13**) in 92% yield. The pyridazinone (**13**) showed the absorption due to amide carbonyl at $1640\,cm^{-1}$ in its IR spectrum, while the ^{1}H-NMR spectrum lacked a carbomethoxy proton signal in comparison with **4**. Further, elemental analysis revealed that **13** possessed two nitrogen atoms in the molecule. Thus, it was established as 6-[3β-[(6-deoxy-α-L-mannopyranosyl)oxy]-14β-hydroxy-androst-4-en-17β-yl]-2,3-dihydro-1,4-phthalazinedione.

Biological Results and Discussion

The Na^+,K^+-adenosine triphosphatase (ATPase) inhibitory activities of the resulting compounds were determined by use of the enzyme from dog kidney and are given as pIC_{50} values.[6,7] Compounds exhibiting pIC_{50} values of more than 6.0 were also investigated by means of measurement of PIE in isolated guinea-pig papillary muscle.[7] The results are summarized in Table VI.

The pIC_{50} values of the benzyl alcohols (**5** and **6**) were smaller than those of the corresponding methyl benzoates (**2** and **3**), a finding which differed from the result in the case of the dimethyl phthalate (**4**).[4a] The *para-*

or *meta*-substituted carboxylic acids (**7** and **8**) showed extremely small pIC_{50} values in comparison with the methyl benzoates (**2** and **3**). The *para*-substituted benzamides (**9a—i**) were more potent than the *meta*-substituted benzamides (**10a—i**). Among the resulting benzamides, the *para*-substituted methylamide (**9a**) and the ethylamide (**9b**) showed nearly the same Na^+, K^+-ATPase inhibitory activities as digoxin and digitoxin. As for the benzamide derivatives, large differences were observed between the pIC_{50} and the pD_2 values, and this result indicated that the mode of action for inhibition of the enzyme by the benzamides (**10a** and **b**) was different from that of proscillaridin (**1**). From the above findings, it appears that smaller substituents lead to more potent biological activities. In addition, *para*-substituted derivatives tend to show more potent enzyme-inhibitory activity and PIE than corresponding *meta*-substituted compounds. It may be of importance for the development of the cardiac remedies from proscillaridin derivatives that the results were similar to our previous findings.[4b]

TABLE VI. Biological Activities of Proscillaridin and Its Derivatives (**2—13**)

Compound	$pIC_{50}^{a)}\pm$S.E.	$pD_2^{b)}\pm$S.E.
1	7.44±0.02	7.41±0.14
2	6.38±0.10	5.92±0.04
3	5.95±0.09	
4	4.75±0.07	
5	6.57±0.06	6.38±0.03
6	5.80±0.03	
7	<5.0	
8	<5.0	
9a	6.49±0.001	5.53±0.03
9b	6.72±0.001	5.13±0.07
9c	6.07±0.001	4.81±0.04
9d	6.14±0.001	4.88±0.08
9e	5.95±0.001	
9f	5.56±0.001	
9g	5.32±0.001	
9h	6.35±0.001	5.14±0.04
9i	5.72±0.001	
10a	6.37±0.001	<4.5
10b	6.37±0.001	<4.5
10c	<5.0	
10d	<5.0	
10e	<5.0	
10f	<5.0	
10g	<5.0	
10h	5.75±0.001	
10i	<5.0	
11a	<5.0	
11b	<5.0	
11c	<5.0	
11d	<5.0	
11e	<5.0	
11f	<5.0	
11h	<5.0	
12g	5.46±0.01	
12i	<5.0	
13	<5.0	

a) pIC_{50} is the concentration of the test compounds required for 50% of the maximum inhibition of Na^+, K^+-ATPase from dog kidney. *b*) pD_2 is the concentration of the test compounds required for 50% of the maximum PIE in guinea-pig papillary muscles.

Experimental

All melting points were determined on a Yanagimoto micro melting point apparatus and are uncorrected. The ultraviolet (UV) spectra were recorded with a Shimadzu UV-2100 spectrometer, and the IR spectra with a JASCO IR-810 spectrometer. The NMR spectra were measured with JEOL GSX-400 spectrometers using tetramethylsilane as an internal standard. The following abbreviations are used; s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet. Optical rotations were measured on a JASCO DIP-140 digital polarimeter. High-performance liquid chromatography (HPLC) was performed using a JASCO 880-PU pump, and a Shodex RI, SE-11 differential refractometer. Thin layer chromatography (TLC) was carried out on Merck precoated Kieselgel 60F$_{254}$ silanized, and spots were detected by illumination with an ultraviolet lamp, or spraying 5% vanillin–70% HClO$_4$, 1% Ce(SO$_4$)$_2$–10% H$_2$SO$_4$ followed by heating. Column chromatography was performed on Silica gel BW-200 (Fuji Davison Chemicals Co., Ltd.).

LiAlH$_4$ Reduction of 2 and 3 LiAlH$_4$ (40 mg, 1.05 mmol) was added portionwise to a solution of **2** (50 mg, 0.087 mmol) in dry tetrahydrofuran (THF) (8 ml) and the resulting mixture was stirred at room temperature for 10 min under N$_2$. The reaction mixture was treated with H$_2$O-saturated ether, H$_2$O, and 5% aqueous NaOH. The precipitate was removed by filtration, and removal of solvent from the filtrate gave a product, which was purified by column chromatography (SiO$_2$, 5% CHCl$_3$–MeOH (7:1)) to furnish **5** (41 mg, 80%). In the same manner, the alcohol (**6**, 39 mg) was prepared from **3** (50 mg, 0.087 mmol) in 76% yield. **5**: colorless crystalline powder, mp 254—255 °C (iso-Pr$_2$O–MeOH). $[\alpha]_D^{25}$ −63.9° (c = 0.6, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3440 (OH). UV λ_{max}^{MeOH} nm (ε): 221.0 (1.2 × 10^4). ^{1}H-NMR (400 MHz, pyridine-d_5) δ: 0.94, 0.95 (each 3H, both s, 10′-CH$_3$, 13′-CH$_3$), 1.72 (3H, d, J = 5.9 Hz, 5″-CH$_3$), 2.97 (1H, dd, J = 7.9, 9.2 Hz, 17′-H), 4.33—4.39 (2H, m, 4″-H, 5″-H), 4.45 (1H, dd, J = 5.8, 7.7 Hz, 3′-H), 4.55 (1H, dd, J = 3.5, 8.8 Hz, 3″-H), 4.59 (1H, dd, J = 1.7, 3.5 Hz, 2″-H), 4.97 (2H, s, CH$_2$OH), 5.10 (1H, s, CH$_2$OH), 5.56 (1H, s, 4′-H), 5.58 (1H, s, 1″-H), 7.69 (2H, d, J = 7.9 Hz, 2-H, 6-H), 7.73 (2H, d, J = 7.9 Hz, 3-H, 5-H). *Anal.* Calcd for C$_{32}$H$_{46}$O$_7$·H$_2$O: C, 68.55; H, 8.63. Found: C, 68.71; H, 8.38. **6**: colorless crystalline powder, mp 266—267 °C (iso-Pr$_2$O–MeOH). $[\alpha]_D^{25}$ −54.5° (c = 0.4, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3440 (OH). UV λ_{max}^{MeOH} nm (ε): 236.0 (sh) (1.0 × 10^4). ^{1}H-NMR (400 MHz, pyridine-d_5) δ: 0.94, 0.95 (each 3H, both s, 10′-CH$_3$, 13′-CH$_3$), 1.71 (3H, d, J = 5.9 Hz, 5″-CH$_3$), 2.99 (1H, dd, J = 8.4, 8.8 Hz, 17′-H), 4.31—4.41 (2H, m, 4″-H, 5″-H), 4.45 (1H, dd, J = 6.7, 9.5 Hz, 3′-H), 4.55 (1H, dd, J = 3.5, 8.6 Hz, 3″-H), 4.59 (1H, s, 2″-H), 4.97 (2H, s, CH$_2$OH), 5.18 (1H, s, CH$_2$OH), 5.56 (1H, s, 4′-H), 5.58 (1H, d, J = 1.3 Hz, 1″-H), 7.38 (1H, t, J = 7.7, 7.7 Hz, 5-H), 7.51 (1H, d, J = 7.7 Hz, 4-H), 7.69 (1H, d, J = 7.7 Hz, 6-H), 7.93 (1H, s, 2-H). *Anal.* Calcd for C$_{32}$H$_{46}$O$_7$: C, 68.55; H, 8.63. Found: C, 68.27; H, 8.52.

Alkaline Hydrolysis of 2 and 3 A solution of **2** (50 mg, 0.087 mmol) in 10% KOH–MeOH (10 ml) was heated under reflux for 20 min. After cooling, the reaction mixture was neutralized with ion-exchange resin (Dowex CHR-W2, H$^+$-form). The resin was removed by filtration and

the filtrate was concentrated under reduced pressure to give the product, which was purified by column chromatography (SiO$_2$, CHCl$_3$–MeOH (4:1)) to furnish **7** (49 mg, quant.). In the same manner, the carboxylic acid **8** (49 mg) was prepared from **3** (50 mg, 0.087 mmol) quantitatively. **7**: colorless crystalline powder, mp 173—175 °C (iso-Pr$_2$O–MeOH), $[\alpha]_D^{25}$ −54.5° (c = 0.8, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3450 (OH), 1695 (CO). UV λ_{max}^{MeOH} nm (ε): 242.0 (1.8 × 10^4). ^{1}H-NMR (400 MHz, pyridine-d_5) δ: 0.94, 0.95 (each 3H, both s, 10′-CH$_3$, 13′-CH$_3$), 1.72 (3H, d, J = 5.9 Hz, 5″-CH$_3$), 3.01 (1H, dd, J = 8.1, 8.8 Hz, 17′-H), 4.31—4.40 (2H, m, 4″-H, 5″-H), 4.46 (1H, dd, J = 6.1, 9.6 Hz, 3′-H), 4.55 (1H, dd, J = 3.5, 8.6 Hz, 3″-H), 4.60 (1H, dd, J = 1.7, 3.5 Hz, 2″-H), 5.56 (1H, s, 4′-H), 5.58 (1H, d, J = 1.3 Hz, 1″-H), 7.85 (2H, d, J = 8.5 Hz, 3-H, 5-H), 8.84 (2H, d, J = 8.5 Hz, 2-H, 6-H). *Anal.* Calcd for C$_{32}$H$_{44}$O$_8$: C, 69.04; H, 7.97. Found: C, 69.11; H, 7.99. **8**: colorless crystalline powder, mp 167—169 °C (iso-Pr$_2$O–MeOH), $[\alpha]_D^{25}$ −53.0° (c = 0.9, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3440 (OH), 1705 (CO). UV λ_{max}^{MeOH} nm (ε): 233.0 (1.0 × 10^4). ^{1}H-NMR (400 MHz, pyridine-d_5) δ: 0.92, 0.95 (each 3H, both s, 10′-CH$_3$, 13′-CH$_3$), 1.71 (3H, d, J = 5.9 Hz, 5″-CH$_3$), 3.03 (1H, dd, J = 8.2, 8.4 Hz, 17′-H), 4.30—4.41 (2H, m, 4″-H, 5″-H), 4.45 (1H, dd, J = 7.7, 8.1 Hz, 3′-H), 4.55 (1H, dd, J = 3.5, 8.7 Hz, 3″-H), 4.59 (1H, dd, J = 1.5, 3.5 Hz, 2″-H), 5.56 (1H, s, 4′-H), 5.58 (1H, d, J = 1.5 Hz, 1″-H), 7.45 (1H, t, J = 7.7, 7.7 Hz, 5-H), 7.95 (1H, d, J = 7.7 Hz, 4-H), 8.33 (1H, d, J = 7.7 Hz, 6-H), 8.82 (1H, s, 2-H). *Anal.* Calcd for C$_{32}$H$_{44}$O$_8$: C, 69.04; H, 7.97. Found: C, 68.77; H, 7.97.

Preparation of Amides by Method A   A solution of **4** (50 mg, 0.080 mmol) in methanol (2.25 ml) and alkylamine (0.5 ml, excess) was heated at 100—110 °C in a sealed tube. After cooling, the reaction mixture was evaporated and the residue was purified by column chromatography (SiO$_2$, CHCl$_3$–MeOH (85:15) [**11a—h**], CHCl$_3$–MeOH (5:1) [**12g, i**]) to furnish the corresponding amides and imides. In the same manner, the corresponding benzamides were prepared from **2** and **3** (SiO$_2$ column chromatography, CHCl$_3$–MeOH (6:1) [**9a, b, 10a, b**]). **9a**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.56 (3H, s, 13′-CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.30 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.89 (1H, dd, J = 7.1, 9.0 Hz, 17′-H), 2.99 (3H, d, J = 4.8 Hz, CONHCH$_3$), 3.44 (1H, t, J = 9.3, 9.3 Hz, 4″-H), 3.73—3.77 (2H, m, 3″-H, 5″-H), 3.90 (1H, dd, J = 1.5, 3.3 Hz, 2″-H), 4.13 (1H, t, J = 7.9, 7.9 Hz, 3′-H), 4.95 (1H, d, J = 1.5 Hz, 1″-H), 5.33 (1H, s, 4′-H), 6.43 (1H, q, J = 4.8 Hz, NH), 7.41 (2H, d, J = 8.5 Hz, 3-H, 5-H), 7.65 (2H, d, J = 8.5 Hz, 2-H, 6-H). **10a**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.57 (3H, s, 13′-CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.30 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.89 (1H, dd, J = 7.2, 9.3 Hz, 17′-H), 2.98 (3H, d, J = 4.8 Hz, CONHCH$_3$), 3.45 (1H, t, J = 9.5, 9.5 Hz, 4″-H), 3.73—3.77 (2H, m, 3″-H, 5″-H), 3.90 (1H, dd, J = 1.5, 3.5 Hz, 2″-H), 4.14 (1H, t, J = 7.8, 7.8 Hz, 3′-H), 4.95 (1H, d, J = 1.5 Hz, 1″-H), 5.33 (1H, s, 4′-H), 6.62 (1H, q, J = 4.8 Hz, NH), 7.29 (1H, t, J = 7.7, 7.7 Hz, 5-H), 7.45 (1H, d, J = 7.7 Hz, 4-H), 7.59 (1H, d, J = 7.7 Hz, 6-H), 7.80 (1H, s, 2-H). **9b**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.56 (3H, s, 13′-CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.24 (3H, t, J = 7.3 Hz, NHCH$_2$CH$_3$), 1.30 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.90 (1H, dd, J = 7.3, 9.3 Hz, 17′-H), 3.44—3.49 (3H, m, NHCH$_2$CH$_3$, 4″-H), 3.72—3.76 (2H, m, 3″-H, 5″-H), 3.95 (1H, s, 2″-H), 4.14 (1H, t, J = 7.6, 7.6 Hz, 3′-H), 4.95 (1H, d, J = 1.5 Hz, 1″-H), 5.32 (1H, s, 4′-H), 6.53 (1H, t, J = 4.8 Hz, NH), 7.42 (2H, d, J = 8.2 Hz, 3-H, 5-H), 7.66 (2H, d, J = 8.5 Hz, 2-H, 6-H). **10b**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.57 (3H, s, 13′-CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.24 (3H, t, J = 7.3 Hz, NHCH$_2$CH$_3$), 1.30 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.91 (1H, dd, J = 7.3, 9.5 Hz, 17′-H), 3.42—3.50 (3H, m, NHCH$_2$CH$_3$, 4″-H), 3.73—3.77 (2H, m, 3″-H, 5″-H), 3.90 (1H, dd, J = 1.4, 3.3 Hz, 2″-H), 4.14 (1H, t, J = 7.7, 7.7 Hz, 3′-H), 4.95 (1H, d, J = 1.4 Hz, 1″-H), 5.32 (1H, d, J = 6.1 Hz, 4′-H), 6.48 (1H, br s, NH), 7.30 (1H, t, J = 7.6, 7.6 Hz, 5-H), 7.48 (1H, d, J = 7.6 Hz, 4-H), 7.58 (1H, d, J = 7.6 Hz, 6-H), 7.77 (1H, s, 2-H). **11a**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.58 (3H, s, 13′-CH$_3$), 1.02 (3H, s, 10′-CH$_3$), 1.25 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.87, 2.89 (each 3H, both s, CONHCH$_3$ × 2), 2.89 (1H, dd, J = 7.1, 8.2 Hz, 17′-H), 3.42 (1H, t, J = 9.4, 9.4 Hz, 4″-H), 3.63—3.68 (2H, m, 3″-H, 5″-H), 3.81 (1H, s, 2″-H), 4.09 (1H, dd, J = 7.3, 8.2 Hz, 3′-H), 4.87 (1H, s, 1″-H), 5.29 (1H, s, 4′-H), 7.23 (2H, s, 5-H, 6-H), 7.62 (1H, s, 3-H). **11b**: ^{1}H-NNR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.59 (3H, s, 13′-CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.20, 1.21 (each 3H, both s, CONHCH$_2$CH$_3$ × 2), 1.30 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.87 (1H, dd, J = 7.0, 8.0 Hz, 17′-H), 3.38—3.47 (5H, m, NHCH$_2$CH$_3$ × 2, 4″-H), 3.72—3.80 (2H, m, 3″-H, 5″-H), 3.90 (1H, s, 2″-H), 4.13 (1H, dd, J = 7.3, 9.2 Hz, 3′-H), 4.95 (1H, s, 1″-H), 5.32 (1H, s, 4′-H), 7.50 (2H, s, 5-H, 6-H), 7.60 (1H, s, 3-H). **11c**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.59 (3H, s, 13′-CH$_3$), 0.97 (6H, t, J = 7.4 Hz, CH$_2$CH$_2$CH$_3$ × 2), 1.03 (3H, s, 10′-CH$_3$), 1.30 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.89 (1H, dd, J = 7.1, 8.1 Hz, 17′-H), 3.33—3.38 (4H, m, NHCH$_2$CH$_2$CH$_3$ × 2), 3.44 (1H, t, J = 9.4, 9.4 Hz, 4″-H), 3.74—3.79 (2H,

m, 3″-H, 5″-H), 3.91 (1H, s, 2″-H), 4.14 (1H, dd, J = 7.1, 7.7 Hz, 3′-H), 4.95 (1H, s, 1″-H), 5.32 (1H, s, 4′-H), 6.70, 6.77 (each 1H, both br s, NH × 2) 7.48 (2H, s, 5-H, 6-H), 7.58 (1H, s, 3-H). **11d**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.59 (3H, s, 13′-CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.23 (12H, d, J = 6.6 Hz, CH(CH$_3$)$_2$ × 2), 1.30 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.89 (1H, dd, J = 7.1, 8.4 Hz, 17′-H), 3.44 (1H, t, J = 9.5, 9.5 Hz, 4″-H), 3.74—3.81 (2H, m, 3″-H, 5″-H), 3.91 (1H, s, 2″-H), 4.14 (1H, dd, J = 6.4, 9.5 Hz, 3′-H), 4.19 (2H, m, NH × 2), 4.95 (1H, s, 1″-H), 5.32 (1H, s, 4′-H), 6.50, 6.72 (each 1H, both d, J = 7.8 Hz, NH × 2), 7.42 (1H, d, J = 8.1 Hz, 5-H), 7.48 (1H, d, J = 8.1 Hz, 6-H), 7.57 (1H, s, 3-H). **11e**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.59 (3H, s, 13′-CH$_3$), 0.94 (6H, t, J = 7.3 Hz, CH$_2$CH$_3$ × 2), 1.03 (3H, s, 10′-CH$_3$), 1.30 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.89 (1H, dd, J = 7.0, 8.6 Hz, 17′-H), 3.34—3.39 (4H, m, NHCH$_2$ × 2), 3.44 (1H, t, J = 9.1, 9.1 Hz, 4″-H), 3.72—3.78 (2H, m, 3′-H, 5″-H), 3.90 (1H, s, 2″-H), 4.13 (1H, dd, J = 7.2, 7.9 Hz, 3′-H), 4.95 (1H, s, 1″-H), 5.32 (1H, s, 4′-H), 7.18, 7.24 (each 1H, both t, J = 5.7 Hz, NH × 2), 7.51 (2H, s, 5-H, 6-H), 7.57 (1H, s, 3-H). **11f**: ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 0.60 (3H, s, 13′-CH$_3$), 0.95 (6H, d, J = 6.6 Hz, CH(CH$_3$)$_2$ × 2), 0.96 (6H, d, J = 6.6 Hz, CH(CH$_3$)$_2$ × 2), 1.03 (3H, s, 10′-CH$_3$), 1.29 (3H, d, J = 6.0 Hz, 5″-CH$_3$), 2.88 (1H, dd, J = 7.1, 8.2 Hz, 17′-H), 3.17—3.24 (4H, m, NHCH$_2$ × 2), 3.45 (1H, t, J = 9.5, 9.5 Hz, 4′-H), 3.75—3.79 (2H, m, 3″-H, 5″-H), 3.90 (1H, s, 2″-H), 4.13 (1H, dd, J = 7.3, 8.2 Hz, 3′-H), 4.95 (1H, s, 1″-H), 5.31 (1H, s, 4′-H), 6.83, 6.96 (each 1H, both t, J = 5.9 Hz, NH × 2), 7.44 (1H, d, J = 7.8 Hz, 5-H), 7.45 (1H, d, J = 7.8 Hz, 6-H), 7.62 (1H, s, 3-H). **11g**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.59 (3H, s, 13′-CH$_3$), 1.03, (3H, s, 10′-CH$_3$), 1.28 (3H, d, J = 6.4 Hz, 5″-CH$_3$), 2.89 (1H, dd, J = 7.2, 8.4 Hz, 17′-H), 3.43 (1H, t, J = 9.3, 9.3 Hz, 4″-H), 3.50 (4H, m, NHCH$_2$ × 2), 3.70—3.75 (6H, m, CH$_2$OH × 2, 3″-H, 5″-H), 3.87 (1H, d, J = 1.5 Hz, 2″-H), 4.13 (1H, dd, J = 6.8, 9.3 Hz, 3′-H), 4.92 (1H, s, 1″-H), 5.31 (1H, s, 4′-H), 7.45 (2H, s, 5-H, 6-H), 7.63, 7.65 (each 1H, both t, J = 5.8 Hz, NH × 2), 7.68 (1H, s, 3-H). **11h**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.58 (3H, s, 13′-CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.29 (3H, d, J = 6.3 Hz, 5″-CH$_3$), 2.88 (1H, dd, J = 7.0, 8.4 Hz, 17′-H), 3.44 (1H, t, J = 9.3, 9.3 Hz, 4″-H), 3.52—3.55 (4H, m, NHCH$_2$ × 2), 3.70—3.78 (6H, m, CH$_2$OH × 2, 3″-H, 5″-H), 3.89 (1H, s, 2″-H), 4.13 (1H, dd, J = 5.9, 7.7 Hz, 3′-H), 4.94 (1H, s, 1″-H), 5.32 (1H, s, 4′-H), 7.43 (1H, d, J = 8.2 Hz, 5-H), 7.48 (1H, d, J = 8.2 Hz, 6-H), 7.61, 7.65 (each 1H, both t, J = 5.8 Hz, NH × 2), 7.65 (1H, s, 3-H). **12g**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.58 (3H, s, 13′-CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.29 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.97 (1H, dd, J = 6.9, 8.9 Hz, 17′-H), 3.43 (1H, t, J = 9.3, 9.3 Hz, 4″-H), 3.69—3.75 (2H, m, 3″-H, 5″-H), 3.76—3.82 (4H, m, NH(CH$_2$)$_2$OH), 3.86 (1H, s, 2″-H), 4.12 (1H, dd, J = 7.3, 8.2 Hz, 3′-H), 4.92 (1H, s, 1″-H), 5.32 (1H, s, 4′-H), 7.63 (2H, s, 5-H, 6-H), 8.00 (1H, s, 3-H). **12i**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.58 (3H, s, 13′-CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.28 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.27 (6H, s, N(CH$_3$)$_2$), 2.56 (2H, t, J = 6.6 Hz, CH$_2$N(CH$_3$)$_2$), 2.96 (1H, dd, J = 6.9, 7.9 Hz, 17′-H), 3.42 (1H, t, J = 9.4, 9.4 Hz, 4″-H), 3.68—3.72 (2H, m, 3″-H, 5″-H), 3.75 (2H, t, NCH$_2$CH$_2$N(CH$_3$)$_2$), 3.86 (1H, s, 2″-H), 4.12 (1H, dd, J = 7.1, 8.3 Hz, 3′-H), 4.91 (1H, s, 1″-H), 5.31 (1H, s, 4′-H), 7.65 (1H, d, J = 7.7 Hz, 5-H), 7.68 (1H, d, J = 7.7 Hz, 6-H), 7.99 (1H, s, 3-H).

Preparation of Amides by Method B   The carboxylic acid (**7**) (80 mg, 0.142 mmol) was dissolved in dry N,N-dimethylformamide (DMF) (1 ml) under an N$_2$ atmosphere and the solutions was cooled at 0 °C. Diphenylphosphoryl azide (62 μl, 0.284 mmol) and n-propylamine (42 μl, 0.426 mmol) were added and the reaction mixture was stirred at 0 °C for 30 min, then at room temperature for a further 4 h. After removal of the solvent, the residue was purified by column chromatography (SiO$_2$, CHCl$_3$–MeOH (6:1)) to furnish **9c**. In the same manner, the corresponding amides were prepared from **7** and **8** (SiO$_2$ column chromatography, CHCl$_3$–MeOH (6:1), [**9d—f, 10d—f**], CHCl$_3$–MeOH (5:1) [**9g, h, 10g, h**], CHCl$_3$–MeOH–H$_2$O (6:4:1) [**9i** and **10i**]). **9c**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.56 (3H, s, 13′-CH$_3$), 0.98 (3H, t, J = 7.4 Hz, NH(CH$_2$)$_2$CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.30 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.90 (1H, dd, J = 7.5, 9.5 Hz, 17′-H), 3.27—3.47 (3H, m, CONHCH$_2$, 4″-H), 3.73—3.77 (2H, m, 3″-H, 5″-H), 3.90 (1H, dd, J = 1.4, 3.5 Hz, 2″-H), 4.14 (1H, dd, J = 7.7, 7.9 Hz, 3′-H), 4.95 (1H, d, J = 1.4 Hz, 1″-H), 5.33 (1H, s, 4′-H), 6.28 (1H, t, J = 5.7 Hz, NH), 7.41 (2H, d, J = 8.4 Hz, 3-H, 5-H), 7.64 (2H, d, J = 8.4 Hz, 2-H, 6-H). **10c**: ^{1}H-NMR (400 MHz, CDCl$_3$ + DMSO-d_6) δ: 0.57 (3H, s, 13′-CH$_3$), 0.98 (3H, t, J = 7.5 Hz, NH(CH$_2$)$_2$CH$_3$), 1.03 (3H, s, 10′-CH$_3$), 1.30 (3H, d, J = 6.2 Hz, 5″-CH$_3$), 2.91 (1H, dd, J = 7.5, 9.5 Hz, 17′-H), 3.26—3.47 (3H, m, CONHCH$_2$, 4″-H), 3.71—3.79 (2H, m, 3″-H, 5″-H), 3.90 (1H, s, 2″-H), 4.14 (1H, t, J = 7.7, 7.7 Hz, 3′-H), 4.95 (1H, d, J = 1.1 Hz, 1″-H), 5.32 (1H, d, J = 3.9 Hz, 4′-H), 6.57 (1H, t, J = 5.7 Hz, NH), 7.29 (1H, t, J = 7.7, 7.7 Hz, 5-H), 7.48 (1H, d, J = 7.7 Hz, 4-H), 7.57 (1H, d, J = 7.7 Hz, 6-H), 7.78 (1H, s, 2-H).

9d: ^{1}H-NMR (400 MHz, CDCl$_3$+DMSO-d_6) δ: 0.56 (3H, s, 13'-CH$_3$), 1.03 (3H, s, 10'-CH$_3$), 1.26 (6H, d, $J=6.6$ Hz, NHCH(C$\underline{\text{H}}_3$)$_2$), 1.31 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.90 (1H, dd, $J=7.9$, 9.3 Hz, 17'-H), 3.45 (1H, t, $J=9.3$, 9.3 Hz, 4''-H), 3.76—3.81 (2H, m, 3''-H, 5''-H), 3.93 (1H, s, 2''-H), 4.14 (1H, dd, $J=7.7$, 7.9 Hz, 3'-H), 4.28 (1H, m, NHC$\underline{\text{H}}$(CH$_3$)$_2$), 4.96 (1H, d, $J=1.5$ Hz, 1''-H), 5.32 (1H, s, 4'-H), 5.81 (1H, d, $J=8.1$ Hz, NH), 7.40 (2H, d, $J=8.4$ Hz, 3-H, 5-H), 7.62 (2H, d, $J=8.4$ Hz, 2-H, 6-H). **10d**: ^{1}H-NMR (400 MHz, CDCl$_3$+DMSO-d_6) δ: 0.57 (3H, s, 13'-CH$_3$), 1.04 (3H, s, 10'-CH$_3$), 1.26 (6H, d, $J=6.6$ Hz, NHCH(C$\underline{\text{H}}_3$)$_2$), 1.33 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.92 (1H, dd, $J=7.5$, 9.5 Hz, 17'-H), 3.46 (1H, dd, $J=9.3$, 9.3 Hz, 4''-H), 3.74—3.80 (2H, m, 3''-H, 5''-H), 3.93 (1H, s, 2''-H), 4.14 (1H, dd, $J=7.5$, 7.9 Hz, 3'-H), 4.28 (1H, m, NHC$\underline{\text{H}}$(CH$_3$)$_2$), 4.97 (1H, d, $J=1.3$ Hz, 1''-H), 5.32 (1H, s, 4'-H), 5.92 (1H, d, $J=8.2$ Hz, NH), 7.30 (1H, t, $J=7.7$, 7.7 Hz, 5-H), 7.50 (1H, d, $J=7.7$ Hz, 4-H), 7.52 (1H, d, $J=7.7$ Hz, 6-H), 7.73 (1H, s, 2-H). **9e**: ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 0.56 (3H, s, 13'-CH$_3$), 0.96 (3H, t, $J=7.3$ Hz, CH$_2$C$\underline{\text{H}}_3$), 1.03 (3H, s, 10'-CH$_3$), 1.31 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.90 (1H, dd, $J=7.0$, 9.2 Hz, 17'-H), 3.42—3.40 (3H, m, NHC$\underline{\text{H}}_2$, 4''-H), 3.74—3.80 (2H, m, 3''-H, 5''-H), 3.93 (1H, d, $J=1.5$ Hz, 2''-H), 4.14 (1H, dd $J=7.5$, 9.2 Hz, 3'-H), 4.97 (1H, d, $J=1.5$ Hz, 1''-H), 5.32 (1H, s, 4'-H), 6.06 (1H, t, $J=5.8$ Hz, NH), 7.40 (2H, d, $J=8.3$ Hz, 3-H, 5-H), 7.62 (2H, d, $J=8.3$ Hz, 2-H, 6-H). **10e**: ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 0.57 (3H, s, 13'-CH$_3$), 0.96 (3H, t, $J=7.3$ Hz, CH$_2$C$\underline{\text{H}}_3$), 1.03 (3H, s, 10'-CH$_3$), 1.29 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.91 (1H, dd, $J=7.0$, 9.3 Hz, 17'-H), 3.41—3.49 (3H, m, NHC$\underline{\text{H}}_2$, 4''-H), 3.74—3.81 (2H, m, 3''-H, 5''-H), 3.93 (1H, dd, $J=1.3$, 3.1 Hz, 2''-H), 4.14 (1H, dd, $J=7.1$, 8.2 Hz, 3'-H), 4.96 (1H, d, $J=1.3$ Hz, 1''-H), 5.32 (1H, s, 4'-H), 6.16 (1H, t, $J=5.8$ Hz, NH), 7.28 (1H, t, $J=7.7$, 7.7 Hz, 5-H), 7.48 (1H, d, $J=7.7$ Hz, 4-H), 7.51 (1H, d, $J=7.7$ Hz, 6-H), 7.80 (1H, s, 2-H). **9f**: ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 0.56 (3H, s, 13'-CH$_3$), 0.98 (6H, d, $J=6.6$ Hz, CH(C$\underline{\text{H}}_3$)$_2$), 1.03 (3H, s, 10'-CH$_3$), 1.31 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.90 (1H, dd, $J=7.1$, 9.3 Hz, 17'-H), 3.28 (2H, t, $J=6.6$ Hz, NHC$\underline{\text{H}}_2$), 3.45 (1H, t, $J=9.2$, 9.2 Hz, 4''-H), 3.75—3.82 (2H, m, 3''-H, 5''-H), 3.93 (1H, s, 2''-H), 4.14 (1H, dd, $J=7.5$, 9.4 Hz, 3'-H), 4.96 (1H, d, $J=1.1$ Hz, 1''-H), 5.32 (1H, s, 4'-H), 6.13 (1H, t, $J=6.1$ Hz, NH), 7.41 (2H, d, $J=8.4$ Hz, 3-H, 5-H), 7.64 (2H, d, $J=8.4$ Hz, 2-H, 6-H). **10f**: ^{1}H-NMR (400 MHz, CDCl$_3$) δ: 0.57 (3H, s, 13'-CH$_3$), 0.93 (3H, d, $J=6.8$ Hz, CH(CH$_3$)$_2$), 1.03 (3H, s, 10'-CH$_3$), 1.31 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.91 (1H, dd, $J=7.2$ 9.4 Hz, 17'-H), 3.27 (2H, t, $J=6.7$ Hz, NHC$\underline{\text{H}}_2$), 3.47 (1H, t, $J=9.4$, 9.4 Hz, 4''-H), 3.74—3.80 (2H, m, 3''-H, 5''-H), 3.93 (1H, s, 2''-H), 4.14 (1H, dd, $J=7.7$, 8.1 Hz, 3'-H), 4.97 (1H, s, 1''-H), 5.32 (1H, s, 4'-H), 6.21 (1H, t, $J=5.9$ Hz, NH), 7.30 (1H, t, $J=7.7$, 7.7 Hz, 5-H), 7.49 (1H, d, $J=7.7$ Hz, 4-H), 7.51 (1H, d, $J=7.7$ Hz, 6-H), 7.78 (1H, s, 2-H). **9g**; ^{1}H-NMR (400 MHz, CDCl$_3$+DMSO-d_6) δ: 0.56 (3H, s, 13'-CH$_3$), 1.03 (3H, s, 10'-CH$_3$), 1.29 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.89 (1H, dd, $J=7.5$, 9.0 Hz, 17'-H), 3.44 (1H, t, $J=9.3$, 9.3 Hz, 4''-H), 3.57 (2H, t, $J=5.3$ Hz, NHC$\underline{\text{H}}_2$), 3.71—3.77 (4H, m, C$\underline{\text{H}}_2$OH, 3''-H, 5''-H), 3.89 (1H, d, $J=1.5$ Hz, 2''-H), 4.13 (1H, t, $J=7.5$, 7.5 Hz, 3'-H), 4.94 (1H, d, $J=1.5$ Hz, 1''-H), 5.32 (1H, s, 4'-H), 7.42 (2H, d, $J=8.2$ Hz, 3-H, 5-H), 7.72 (2H, d, $J=8.2$ Hz, 2-H, 6-H). **10g**: ^{1}H-NMR (400 MHz, CDCl$_3$+DMSO-d_6) δ: 0.57 (3H, s, 13'-CH$_3$), 1.03 (3H, s, 10'-CH$_3$), 1.29 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.90 (1H, dd, $J=7.1$, 9.4 Hz, 17'-H), 3.44 (1H, t, $J=9.3$, 9.3 Hz, 4''-H), 3.58 (2H, t, $J=5.3$ Hz, NHC$\underline{\text{H}}_2$), 3.72—3.77 (4H, m, C$\underline{\text{H}}_2$OH, 3''-H, 5''-H), 3.89 (1H, dd, $J=1.7$, 3.5 Hz, 2''-H), 4.13 (1H, dd, $J=7.5$, 7.7 Hz, 3'-H), 4.94 (1H, d, $J=1.7$ Hz, 1''-H), 5.32 (1H, s, 4'-H), 7.28 (1H, t, $J=7.7$, 7.7 Hz, 5-H), 7.43 (1H, d, $J=7.7$ Hz, 4-H), 7.64 (1H, d, $J=7.7$ Hz, 6-H), 7.89 (1H, s, 2-H). **9h**: ^{1}H-NMR (400 MHz, CDCl$_3$+DMSO-d_6) δ: 0.56 (3H, s, 13'-CH$_3$), 1.03 (3H, s, 10'-CH$_3$), 1.30 (3H, d, $J=6.4$ Hz, 5''-CH$_3$), 2.89 (1H, dd, $J=7.5$, 9.0 Hz, 17'-H), 3.44 (1H, t, $J=9.5$, 9.5 Hz, 4''-H), 3.59 (2H, t, $J=6.0$ Hz, NHC$\underline{\text{H}}_2$),

3.63—3.76 (4H, m, C$\underline{\text{H}}_2$OH, 3''-H, 5''-H), 3.89 (1H, dd, $J=1.5$, 3.5 Hz, 2''-H), 4.13 (1H, dd, $J=7.5$, 7.5 Hz, 3'-H), 4.94 (1H, d, $J=1.5$ Hz, 1''-H), 5.32 (1H, s, 4'-H), 7.42 (2H, d, $J=8.4$ Hz, 3-H, 5-H), 7.69 (2H, d, $J=8.4$ Hz, 2-H, 6-H). **10h**: ^{1}H-NMR (400 MHz, CDCl$_3$+DMSO-d_6) δ: 0.57 (3H, s, 13'-CH$_3$), 1.03 (3H, s, 10'-CH$_3$), 1.30 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.90 (1H, dd, $J=7.7$, 8.8 Hz, 17'-H), 3.44 (1H, t, $J=9.4$, 9.4 Hz, 4''-H), 3.59 (2H, t, $J=6.0$ Hz, NHC$\underline{\text{H}}_2$), 3.67—3.76 (4H, m, C$\underline{\text{H}}_2$OH, 3''-H, 5''-H), 3.89 (1H, dd, $J=1.5$, 3.5 Hz, 2''-H), 4.13 (1H, t, $J=7.5$, 7.5 Hz, 3'-H), 4.94 (1H, d, $J=1.5$ Hz, 1''-H), 5.32 (1H, s, 4'-H), 7.28 (1H, t, $J=7.7$, 7.7 Hz, 5-H), 7.43 (1H, d, $J=7.7$ Hz, 4-H), 7.62 (1H, d, $J=7.7$ Hz, 6-H), 7.87 (1H, s, 2-H). **9i**: ^{1}H-NMR (400 MHz, CDCl$_3$+DMSO-d_6) δ: 0.56 (3H, s, 13'-CH$_3$), 1.03 (3H, s, 10'-CH$_3$), 1.26 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.31 (6H, s, N(C$\underline{\text{H}}_3$)$_2$), 2.87 (1H, dd, $J=7.5$, 9.3 Hz, 17'-H), 3.40 (1H, t, $J=9.5$, 9.5 Hz, 4''-H), 3.50 (2H, br s, NHC$\underline{\text{H}}_2$), 3.60—3.67 (2H, m, 3''-H, 5''-H), 3.83 (1H, br s, 2''-H), 4.11 (1H, dd, $J=7.5$, 7.9 Hz, 3'-H), 4.89 (1H, s, 1''-H), 5.31 (1H, s, 4'-H), 7.44 (2H, d, $J=8.2$ Hz, 3-H, 5-H), 7.71 (2H, d, $J=8.2$ Hz, 2-H, 6-H). **10i**: ^{1}H-NMR (400 MHz, CDCl$_3$+DMSO-d_6) δ: 0.56 (3H, s, 13'-CH$_3$), 1.02 (3H, s, 10'-CH$_3$), 1.26 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 2.60 (6H, s, N(C$\underline{\text{H}}_3$)$_2$), 2.89 (1H, dd, $J=7.5$, 9.3 Hz, 17'-H), 3.40 (1H, t, $J=9.3$, 9.3 Hz, 4''-H), 3.51 (2H, t, $J=6.0$ Hz, NHC$\underline{\text{H}}_2$), 3.67—3.70 (2H, m, 3''-H, 5''-H), 3.84 (1H, s, 2''-H), 4.11 (1H, dd, $J=7.5$, 7.9 Hz, 3'-H), 4.89 (1H, d, $J=1.3$ Hz, 1''-H), 5.31 (1H, s, 4'-H), 7.27 (1H, 7.7, 7.7 Hz, 5-H), 7.47 (1H, d, $J=7.7$ Hz, 4-H), 7.61 (1H, d, $J=7.7$ Hz, 6-H), 7.88 (1H, s, 2-H).

13: A solution of **4** (200 mg, 0.314 mmol) and hydrazine monohydrate (10 ml, excess) was heated under reflux for 30 h under an N$_2$ atmosphere. After cooling, the reaction mixture was evaporated and the residue was purified by recrystallization from isopropylether–methanol to furnish a colorless crystalline powder (190 mg, 92%). ^{1}H-NMR (400 MHz, CDCl$_3$+DMSO-d_6) δ: 0.55 (3H, s, 13'-CH$_3$), 1.02 (3H, s, 10'-CH$_3$), 1.24 (3H, d, $J=6.2$ Hz, 5''-CH$_3$), 3.02 (1H, dd, $J=8.2$, 8.3 Hz, 17'-H), 3.36 (1H, t, $J=8.8$, 8.8 Hz, 4''-H), 3.61—3.67 (2H, m, 3''-H, 5''-H), 3.79 (1H, s, 2''-H), 4.10 (1H, t, $J=7.2$, 7.2 Hz, 3'-H), 4.35, 4.44 (each 1H, both br s NH$\times$2), 4.86 (1H, s, 1''-H), 5.30 (1H, s, 4'-H), 7.97 (1H, d, $J=8.4$ Hz, 6-H), 8.01 (1H, d, $J=8.4$ Hz, 5-H), 8.08 (1H, s, 3-H).

References

1) Part VIII: N. Murakami, Y. Sato, T. Tanase, T. Nagai, T. Ueda, J. Sakakibara, H. Ando, Y. Hotta, K. Takeya, and M. Asano, *Yakugaku Zasshi*, **111**, 436 (1991).
2) H. G. Kurbjuweit, *Arzneim.-Forsch*, **14**, 716 (1964).
3) A. Schwartz, G. E. Lindenmayer, and J. C. Allen, *Pharmacol. Rev.*, **27**, 3 (1975).
4) *a*) J. Mori, S. Nagai, J. Sakakibara, K. Takeya, and Y. Hotta, *Chem. Pharm. Bull.*, **36**, 48 (1988); *b*) N. Murakami, T. Tanase, S. Nagai, T. Ueda, J. Sakakibara, H. Ando, Y. Hotta, and K. Takeya, *ibid.*, **39**, 1962 (1991).
5) T. Shioiri, K. Ninomiya, and S. Yamada, *J. Am. Chem. Soc.*, **94**, 6203 (1972).
6) W. Schorner and R. Reitz, *Justus Liebigs Ann. Chem.*, **1976**, 610.
7) J. Sakakibara, J. Mori, S. Nagai, Y. Hotta, and K. Takeya, *Nippon Yakurigaku Zasshi*, **90**, 115 (1987).