Taxuspines X—Z, New Taxoids from Japanese Yew *Taxus cuspidata*

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Three new taxoids, taxuspines X—Z (1—3), have been isolated from stems of the Japanese yew *Taxus cuspidata* SIEB. *et* ZUCC. and the structures elucidated on the basis of spectroscopic data. Taxuspine X (1) is a rare example of a bicyclic taxane-related diterpenoid consisting of 6/12-membered ring system with a cinnamoyl group at C-5. Taxuspine Y (2) is one of unique taxoids with 5/7/6-membered ring system, while taxuspine Z (3) is a new taxoid consisting of 6/8/6-ring system with a Winterstein's acid moiety at C-5.

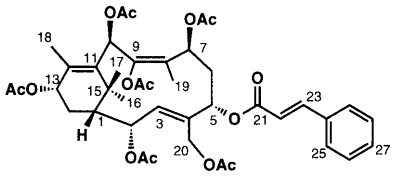
Key words taxoid; taxuspine X—Z; Japanese yew; bicyclic diterpenoid

Numerous taxoids have been isolated from various yew trees and some of them exhibit interesting biological activities.[1] In our continuing search for bioactive taxoids, we previously isolated a series of new taxoids, taxuspines A—H and J—W,[2-8] from stems and leaves of the Japanese yew *Taxus cuspidata* SIEB. *et* ZUCC. Further investigation of the extracts of stems of this yew led to the isolation of three new taxane and related diterpenoids, taxuspines X—Z (**1**—**3**). In this paper we describe the isolation and structure elucidation of **1**—**3**.

The methanolic extract of stems of the yew collected at Sapporo was partitioned between toluene and water. The toluene soluble portions were subjected to a silica gel column followed by reversed-phase column chromatographies to afford taxuspines X (**1**, 0.00014%), Y (**2**, 0.0002%), and Z (**3**, 0.000026%).

Taxuspine X (**1**) was obtained as a colorless amorphous solid and showed the molecular ion peak at m/z 766 (M^+) in the electron impact (EI)-MS spectrum. HR-EI-MS analysis revealed the molecular formula to be $C_{41}H_{50}O_{14}$ [m/z 766.3191 (M^+), Δ −1.0 mmu]. IR absorptions at 1740 and 1640 cm^{-1} implied that **1** possessed ester and unsaturated ester groups. Analyses of the ^1H- and ^{13}C-NMR data (Table 1) and heteronuclear multiple quantum coherence (HMQC) spectrum of **1** provided six acetyls, one unsaturated ester carbonyl, one monosubstituted benzene, one disubstituted olefin, one trisubstituted olefin, two tetrasubstituted olefin, five oxymethines, one oxymethylene, one sp^3 methine, two sp^3 methylenes, one sp^3 quaternary carbon, and four methyl groups. Proton signals due to a cinnamoyl group appeared at δ_H 7.40 (3H, m), 7.53 (2H, m), 6.60 (1H, d, J = 15.1 Hz; *trans*-oriented), and 7.87 (1H, d, J = 15.1 Hz). UV absorption at 279 nm also supported the presence of the cinnamoyl group. Since fifteen out of seventeen unsaturations were thus accounted for, **1** was inferred to contain two rings. Detailed analysis of the ^1H–^1H correlation spectroscopy (COSY) spectrum revealed connectivities of C-1 to C-3, C-5 to C-7, C-13 to C-1, and C-22 to C-23. Heteronuclear multiple-bond correlation spectroscopy (HMBC) correlations of H-13 to C-11 and C-12, H_3-16 and H_3-17 to C-1, C-11, and C-15 indicated the presence of a cyclohexene moiety (ring A), while the correlations of H_3-18 to C-11, C-12, and C-13 revealed that Me-18 was attached at C-12. HMBC correlations of H-3 to C-5, H-5 to C-3 and C-4, H-6a to C-8, H-7 to C-9, H-10 to C-9, and H-10 to C-11 suggested the presence of a cyclododecadiene moiety (ring B). The presence of an oxymethylene (C-20) at C-4 was deduced from the chemical shifts (δ_H 4.41, 4.93, H-20; δ_C 59.67, C-20) and HMBC correlations of H_2-20 to C-3 and C-4, and H-20a to C-5. HMBC cross-peaks of H_3-19 to C-7, C-8, and C-9 indicated that Me-19 was attached at C-8. Five out of six acetoxy groups were attached at C-2, C-7, C-10, C-13, and C-20 based on HMBC correlations of H-2 (δ_H 5.80), H-7 (δ_H 5.50), H-10 (δ_H 7.27), H-13 (δ_H 5.28), and H-20 (δ_H 4.41, 4.93) to acetyl carbonyls, while the cinnamoyl group was connected to C-5 from an HMBC correlation of H-5 to C-21 (δ_C 165.65). The remaining acetoxy group was an enol acetate connected to C-9, judging from the carbon chemical shift (δ_C 143.67).[8] Thus the structure of taxuspine X was assigned to be **1**. Relative stereochemistry of **1** was deduced from the nuclear Overhauser effect spectroscopy (NOESY) spectrum as shown in Fig. 1. The NOESY correlation of H-13/H_3-17 revealed that the ring A had a boat conformation and H-13 was β-oriented, while the correlations of H-2/H-16 and H-14a/H-17 indicated the β-orientation of H-14a and H-2. NOESY correlations of H-20a/H-2 and H-7/H-10 implied that the two double bonds at C-3 and C-8 had *E*-configuration. NOESY cross-peaks of H-7/H_3-18, H-10/H_3-18, and H_3-18/H-23 revealed that H-7, H-10, and the cinnamoyl group were α-oriented.

The molecular formula of taxuspine Y (**2**) was established to be $C_{31}H_{38}O_9$ by the HR-EI-MS [m/z 494.1907 ($M-i$-PrOH)$^+$, Δ −3.3 mmu] and ^{13}C-NMR data. The IR spectrum indicated the presence of hydroxy



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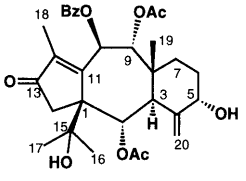

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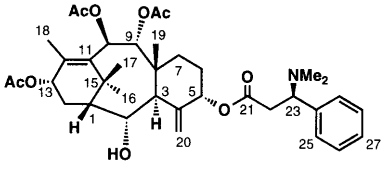

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Table 1. ^{1}H- and ^{13}C-NMR Data of Taxuspine X (**1**) in $CDCl_3$

Position	^{1}H[a]	J (Hz)	^{13}C[a]	H coupled with C[b]
1	1.81 m		46.47 d	H-2, H-13, H-14a, H-16, H-17
2	5.80 dd	11.1, 4.8	68.85 d	
3	5.99 d	11.1	124.48 d	H-5, H-20a, H-20b
4			132.21 s	H-5, H-20a, H-20b
5	5.75 m		69.42 d	H-3, H-7, H-20a
6 (a)	2.57 m		35.11 t	H-7
(b)	2.14 m			
7	5.50 d	9.1	66.74 d	H-5, H-6a, H-19
8			125.45 s	H-6a, H-19
9			143.67 s	H-7, H-10, H-19
10	7.27 s		68.56 d	
11			136.92 s	H-10, H-13, H-16, H-17, H-18
12			135.40 s	H-10, H-13, H-14a, H-18
13	5.28 d	9.1	69.99 d	H-18
14 (a)	2.55 m		25.80 t	H-2, H-13
(b)	2.03 m			
15			36.19 s	H-10, H-16, H-17
16	1.12 s		33.20 q	H-17
17	1.30 s		25.17 q	H-16
18	2.28 s		17.01 q	
19	1.65 s		12.23 q	H-7
20 (a)	4.93 d	12.9	59.67 t	H-3
(b)	4.41 d	12.9		
21			165.65 s	H-5, H-22, H-23
22	6.60 d	15.1	117.87 d	
23	7.87 d	15.1	145.93 d	
24			134.10 s	H-22
25	7.53 m		128.12 d	H-27
26	7.40 m		129.08 d	
27	7.40 m		130.70 d	H-25
2-AcO	1.98 s		169.18 s	H-2
			20.84 q	
7-AcO	1.98 s		169.71 s	H-7
			21.01 q	
9-AcO	1.88 s		167.83 s	
			20.41 q	
10-AcO	2.06 s		167.84 s	H-10
			21.05 q	
13-AcO	1.95 s		170.58 s	H-13
			21.05 q	
20-AcO	2.20 s		170.63 s	H-20
			21.30 q	

a) In ppm; *b*) in HMBC spectrum.

(3455 cm^{-1}), ester (1715 cm^{-1}), and unsaturated carbonyl (1700 cm^{-1}) groups. The ^{1}H- and ^{13}C-NMR data showed signals due to one α,β-unsaturated carbonyl, one exomethylene, two acetyls, one benzoyl, one tetrasubstituted olefin, one oxygenated quaternary carbon, four oxymethines, two sp^3 quaternary carbons, one sp^3 methine, three methylenes, and four methyl groups. Since eight out of eleven unsaturations were thus accounted for, **2** was inferred to contain three rings. HMBC correlations of H-14a to C-1, C-11, and C-13 and H_3-18 to C-11, C-12, and C-13 revealed the presence of a cyclopentenone ring (ring A), which was also supported by the UV absorption at 235 nm (ε 15700). Signals due to two methyl groups (δ_H 1.18, 1.23), a deuterium-exchangeable proton (δ_H 2.50), and an oxygenated quaternary carbon (δ_C 75.81, C-15) indicated the presence of a hydroxyisopropyl group, which was attached at C-1 from HMBC correlations of H_3-16 and H_3-17 to C-1. The hydroxyisopropyl group was

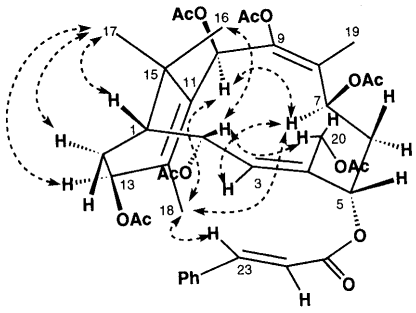

Fig. 1. Relative Stereochemistry of Taxuspine X (**1**)
Dotted arrows denote NOESY correlations.

inferred to be β-oriented, since NOESY correlations of H_3-16/H-2 and H_3-17/H-14a were observed. HMBC correlations of H-2 to C-8, H-6b to C-8, H_2-7 to C-9, and H_3-19 to C-8 and NOESY correlations of H-2/H_3-19, H-3/H-7b, and H-7b/H-10 revealed the presence of a seven-membered ring (ring B) and a six-membered ring (ring C). Two acetoxy groups were connected to C-2 and C-9, judging from HMBC correlations of H-2 and H-9 to acetyl carbonyls (δ_C 170.00 and 171.52, respectively), while one benzoate group was attached at C-10 based on the HMBC cross-peak between H-10 and a benzoyl carbonyl carbon (δ_C 164.77). The signals at δ_H 4.45 and 5.12 (each 1H, s) indicated the presence of an exomethylene at C-4 by HMBC correlation of H-20a to C-3 and C-5, while a hydroxyl group was attached at C-5, judging from a proton signal at δ_H 4.25 (br s, H-5). Thus the structure of taxuspine Y was assigned to be **2**. Relative stereochemistry of **2** was elucidated to have a chair/boat conformation at B/C ring by the NOESY spectrum.

The molecular formula of taxuspine Z (**3**) was revealed to be $C_{37}H_{51}NO_9$ by HR-EI-MS [m/z 653.3589 (M^+), Δ +2.6 mmu]. The ^{1}H- and ^{13}C-NMR signals (δ_H 2.83, 3.01, 3.75, 7.33, 7.40; δ_C 40.32, 42.86, 67.89, 127.70, 128.23, 128.37, 139.08, 170.94) indicated the presence of a 3-*N*,*N*-dimethylamino-3-phenylpropanoyl group (Winterstein's acid[9]). Prominent fragment ion peaks at m/z 134 and 192 in the EI-MS corresponded to fission of the Winterstein's acid moiety from **3**. HMBC correlations of H_3-16 and H_3-17 to C-1 and C-15, H_3-18 to C-11, C-12, and C-13, H_3-19 to C-3, C-7, C-8, and C-9, and H-20a to C-3 and C-5 revealed that **3** possessed a 6/8/6-ring system. Three acetoxy groups were attached at C-9, C-10, and C-13 based on the HMBC correlations of H-9, H-10, and H-13 to acetyl carbonyls (δ_C 169.88, 170.23, and 170.55, respectively) and the oxymethine proton resonances (δ_H 5.82, 5.96, and 5.91, respectively), while a hydroxyl group was attached at C-2 based on the HMBC correlation of an oxymethine proton (δ_H 4.16) to C-2. The Winterstein's acid part was attached at C-5 by an HMBC correlation of H-5 to C-21. Thus the structure of taxuspine Z was assigned to be **3** and the relative stereochemistry was elucidated from the NOESY spectrum. The absolute stereochemistry at C-23 of the Winterstein's acid moiety was determined to be *R* by chiral HPLC analysis (SUMICHIRAL OA-5000) of the acid hydrolysis products of **3**.

Taxuspines X—Z (**1**—**3**) are new taxoids from stems of

the Japanese yew *Taxus cuspidata* SIEB. *et* ZUCC. Taxuspine X (**1**) is a rare example of bicyclic taxane-related diterpenoids consisting of 6/12-membered ring system with a cinnamoyl group at C-5.[10,11] Taxuspine Y (**2**) is one of unique taxoids with 5/7/6-membered ring system, while taxuspine Z (**3**) is a new taxoid consisting of a 6/8/6-ring system with a Winterstein's acid moiety at C-5.[12] Compounds **1**—**3** exhibited cytotoxicity *in vitro* against L1210 murine leukemia cells with IC_{50} values of 4.2, 5.4, and $>10\,\mu g/ml$, respectively, and against KB human epidermoid carcinoma cells with IC_{50} values of >10, >10, and $6.2\,\mu g/ml$, respectively.

Experimental

General Methods Optical rotations were determined on a JASCO DIP-370 polarimeter. UV and IR spectra were obtained on JASCO Ubest-35 and JASCO IR Report-100 spectrometers, respectively. ^1H- and ^{13}C-NMR spectra were recorded on Bruker ARX-500 and AMX-600 spectrometers. The 7.26 ppm resonance of residual $CHCl_3$ and 77.0 ppm of $CDCl_3$ were used as internal references, respectively. EI-MS was obtained on a JEOL DX-303 spectrometer operating at 70 eV. FAB-MS was measured on a JEOL HX-110 spectrometer by using glycerol matrix.

Collection, Extraction, and Separation The Japanese yew *Taxus cuspidata* SIEB. *et* ZUCC. was collected at Sapporo, Hokkaido. The stems (1.2 kg) of the yew was extracted with MeOH (15 l × 4). The MeOH extract was partitioned between toluene (1 l × 4) and H_2O (750 ml). The toluene soluble portions were evaporated under reduced pressure to give a residue (24.5 g), part of which (15.9 g) was subjected to a silica gel column (hexane/acetone, 8 : 1) to give a fraction (3.22 g), part of which (1.11 g) was subjected to a silica gel column ($CHCl_3$/acetone, 10 : 1) to afford a fraction (38.6 mg). The fraction was separated by a reversed-phase column (YMC GEL ODS 60; CH_3CN/H_2O, 1 : 1) to give a fraction (17.7 mg), which was purified by reversed-phase HPLC (YMC-Pack ODS AL-323, 10 × 250 mm; flow rate 2.5 ml/min; UV detection at 227 nm; CH_3CN/H_2O, 1 : 1) to give taxuspine Y (**2**, 1.0 mg, t_R 20.0 min). Toluene extract (115 g) obtained from the stems (11 kg) of the yew was subjected to a silica gel column with hexane/acetone to give fractions I (29.2 g; hexane/acetone, 2 : 1) and II (7.52 g; hexane/acetone, 1 : 1). The fraction II was subjected to a silica gel column (hexane/acetone, 8 : 1) followed by a reversed-phase HPLC (YMC-Pack ODS AM-323, 10 × 250 mm; flow rate 2.5 ml/min; UV detection at 227 nm; CH_3OH/H_2O, 70 : 30) to afford taxuspine Z (3, 2.0 mg, t_R 28.0 min). The fraction I was separated by a reversed-phase column (YMC GEL ODS 60; CH_3CN/H_2O, 4 : 1) followed by a silica gel column ($CHCl_3$/acetone, 10 : 1) to give a fraction (0.90 g), which was separated by a silica gel column ($CHCl_3$/MeOH, 40 : 1) followed by a reversed-phase column (YMC-GEL ODS 60; MeOH/H_2O, 80 : 20) and then reversed-phase HPLC (YMC-Pack ODS AL-323, 10 × 250 mm; flow rate 2.5 ml/min; UV detection at 227 nm; MeOH/H_2O, 70 : 30) to give taxuspine X (1, 11.7 mg, t_R 27.2 min).

Taxuspine X (**1**): A colorless amorphous solid; $[\alpha]_D^{22}$ +31.7° (c = 0.13, $CHCl_3$); UV (MeOH) λ_{max} 279 (ε 15300), 217 (19000), 205 (17900) nm; IR (film) ν_{max} 3450, 1740, 1640, 1370, 1240 cm^{-1}; ^1H- and ^{13}C-NMR (Table 1); EI-MS *m/z* 766 (M^+); HR-EI-MS *m/z* 766.3191 (M^+), Calcd for $C_{41}H_{50}O_{14}$, 766.3201; ^1H–^1H COSY correlations ($CDCl_3$, H/H): 1/2, 1/14a, 2/3, 5/6a, 5/6b, 6a/6b, 6a/7, 6b/7, 9/10, 13/14a, 13/14b, 14a/14b, 20a/20b, 22/23, 25/26, 26/27; HMBC correlations (Table 1); NOESY correlations ($CDCl_3$ H/H): 1/2, 1/14a, 1/16, 1/17, 2/16, 2/20a, 3/5, 3/7, 3/16, 3/22, 5/6a, 5/6b, 5/19, 6a/19, 6a/20b, 7/10, 7/18, 7/19, 10/18, 13/14a, 13/17, 13/18, 13/20a, 14a/17, 18/22, 18/23, 19/20b, 22/25, 23/25.

Taxuspine Y (**2**): A colorless amorphous solid; $[\alpha]_D^{31}$ −25.4° (c = 0.17, $CHCl_3$); UV (MeOH) λ_{max} 235 (ε 15700), 204 (11100) nm; IR (film) ν_{max} 3455, 1715, 1700, 1370, 1250 cm^{-1}; ^1H-NMR ($CDCl_3$): δ 7.91 (2H, d, J = 9.2 Hz, H-23, H-27), 7.58 (1H, m, H-25), 7.45 (2H, m, H-24, H-26), 6.62 (1H, d, J = 10.3 Hz, H-10), 6.17 (1H, d, J = 9.2 Hz, H-2), 6.12 (1H, d, J = 10.3 Hz, H-9), 5.12 (1H, s, H-20a), 4.45 (1H, s, H-20b), 4.25 (1H, br s, H-5), 3.29 (1H, d, J = 9.2 Hz, H-3), 2.74 (1H, d, J = 18.8 Hz, H-14b), 2.50 (1H, br s, 15-OH), 2.48 (1H, d, J = 18.8 Hz, H-14a), 2.04 (3H, s, 2-AcO), 1.96 (3H, s, H_3-18), 1.84 (1H, m, H-6b), 1.82 (3H, s, 9-AcO), 1.81 (1H, m, H-7b), 1.77 (1H, m, H-6a), 1.64 (1H, m, H-7a), 1.23 (3H, s, H_3-16), 1.18 (3H, s, H_3-17), 1.02 (3H, s, H_3-19); ^{13}C-NMR ($CDCl_3$): δ 207.4 (s, C-13), 171.52 (s, 2-$\underline{C}OCH_3$), 170.00 (s, 9-$\underline{C}OCH_3$), 164.77 (s, C-21), 161.75 (s, C-11), 155.43 (s, C-4), 146.85 (s, C-12), 133.72 (s, C-22), 129.64 (d, C-25), 128.87 (d, C-24, C-26), 128.74 (d, C-23, C-27), 111.38 (t, C-20), 76.02 (d, C-9), 75.81 (s, C-15), 75.81 (s, C-5), 68.87 (d, C-10), 68.02 (d, C-2), 65.00 (s, C-1), 44.21 (t, C-3), 44.21 (d, C-14), 42.50 (s, C-8), 30.45 (q, C-17), 30.25 (t, C-6), 26.66 (t, C-7), 26.66 (q, C-16), 21.69 (q, 2-$\underline{C}H_3CO$), 20.52 (q, 9-$\underline{C}H_3CO$), 16.50 (q, C-19), 8.87 (q, C-18); EI-MS *m/z* 494 ($M-i$-PrOH)$^+$; HR-EI-MS *m/z* 494.1907 ($M-i$-PrOH)$^+$, Calcd for $C_{28}H_{30}O_8$, 494.1940; ^1H–^1H COSY correlations ($CDCl_3$, H/H): 2/3, 5/6a, 5/6b, 6a/6b, 6b/7, 9/10, 14a/14b, 20a/20b; HMBC correlations ($CDCl_3$, H/C): 2/3, 2/8, 2/15, 2/$CH_3\underline{C}O$, 6b/8, 7/9, 9/$CH_3\underline{C}O$, 10/1, 10/9, 10/12, 10/Ph$\underline{C}O$, 20a/3, 20a/5, 14a/1, 14a/11, 14a/12, 14a/13, 14a/15, 16/1, 16/15, 16/17, 17/1, 17/15, 17/16, 18/11, 18/12, 18/13, 19/8; NOESY correlations ($CDCl_3$, H/H): 2/3, 2/16, 2/19, 2/20a, 3/7b, 3/10, 3/14b, 5/6b, 6b/19, 7/10, 7a/19, 9/16, 9/19, 10/18, 14a/17, 19/20a, 19/20b.

Taxuspine Z (**3**): A colorless amorphous solid; $[\alpha]_D^{28}$ +31.2° (c = 0.08, $CHCl_3$); UV (MeOH) λ_{max} 265 (ε 2330), 210 (15400) nm; IR (film) ν_{max} 3530, 1740, 1640 cm^{-1}; ^1H-NMR ($CDCl_3$): δ 7.40 (2H, m, H-26, H-28), 7.40 (1H, m, H-27), 7.33 (2H, m, H-25, H-29), 5.96 (1H, d, J = 10.5 Hz, H-10), 5.91 (1H, dd, J = 15.4, 8.0 Hz, H-13), 5.82 (1H, d, J = 10.5 Hz, H-9), 5.42 (1H, s, H-20a), 5.39 (1H, s, H-20b), 5.21 (1H, s, H-5), 4.16 (1H, dd, J = 6.8, 3.0 Hz, H-2), 3.75 (1H, t, J = 6.8 Hz, H-23), 3.01 (1H, m, H-22a), 2.97 (1H, d, J = 6.8 Hz, H-3), 2.83 (1H, m, H-22b), 2.56 (1H, m, H-14a), 2.20 (3H, s, H_3-18), 2.18 (6H, s, NMe_2), 2.10 (1H, m, H-1), 2.05 (3H, s, 13-AcO), 2.04 (3H, s, 9-AcO), 2.01 (3H, s, 10-AcO), 1.68 (3H, s, H_3-16), 1.59 (1H, m, H-7a), 1.45 (1H, m, H-6a), 1.38 (1H, dd, J = 11.0, 4.5 Hz, H-7b), 1.30 (1H, m, H-14b), 1.15 (3H, s, H_3-17), 1.10 (1H, m, H-6b), 0.84 (3H, s, H_3-19); ^{13}C-NMR ($CDCl_3$): δ 170.94 (s, C-21), 170.55 (s, 13-$CH_3\underline{C}O$), 170.23 (s, 10-$CH_3\underline{C}O$), 169.88 (s, 9-$CH_3\underline{C}O$), 143.46 (s, C-4), 139.08 (s, C-24), 136.71 (s, C-12), 132.95 (s, C-11), 128.37 (d, C-25, C-29), 128.23 (d, C-26, C-28), 127.70 (d, C-27), 119.44 (t, C-20), 77.81 (d, C-5), 76.75 (d, C-9), 72.38 (d, C-10), 70.38 (d, C-13), 70.04 (d, C-2), 67.89 (d, C-23), 51.33 (d, C-1), 46.10 (d, C-3), 44.33 (s, C-8), 42.86 (q, NMe_2), 40.32 (t, C-22), 37.29 (s, C-15), 31.61 (q, C-17), 28.68 (t, C-6), 28.02 (t, C-7), 27.41 (t, C-14), 27.10 (q, C-16), 21.34 (q, 13-$\underline{C}H_3CO$), 21.00 (q, 9-$\underline{C}H_3CO$), 20.81 (q, 10-$\underline{C}H_3CO$), 17.97 (q, C-19), 15.24 (q, C-18); EI-MS *m/z* 653 (M^+), 192, 134; HR-EI-MS *m/z* 653.3589 (M^+), Calcd. for $C_{37}H_{51}NO_9$, 653.3563; ^1H–^1H COSY correlations ($CDCl_3$, H/H): 1/2, 1/14a, 2/3, 5/6a, 5/6b, 6a/6b, 6b/7, 9/10, 13/14a, 14a/14b, 20a/20b, 22a/22b, 22/23, 25/27; HMBC correlations ($CDCl_3$, H/C); 3/2, 3/4, 3/19, 5/3, 5/7, 5/21, 6b/7, 7/19, 9/7, 9/8, 9/10, 9/19, 9/$CH_3\underline{C}O$, 10/9, 10/11, 10/12, 10/$CH_3\underline{C}O$, 13/$CH_3\underline{C}O$, 14a/12, 14a/13, 14b/13, 16/1, 16/11, 16/15, 16/17, 17/1, 17/11, 17/15, 17/16, 18/11, 18/12, 18/13, 19/3, 19/7, 19/8, 19/9, 20a/3, 20a/5, 22/21, 22/23, 22/24, 23/22; NOESY correlations ($CDCl_3$, H/H): 1/14a, 1/16, 1/17, 2/9, 2/16, 2/19, 3/14b, 3/18, 5/6a, 5/6b, 7b/10, 9/16, 9/19, 10/18, 13/14a, 13/17, 16/17, 18/22b, 19/20a, 25/26, 26/27.

Determination of Absolute Configuration of 3-*N*,*N*-Dimethylamino-3-phenylpropanoic Acid in Taxuspine Z (3) Compound **3** (0.1 mg) was refluxed in 10% HCl aq. (0.1 ml) for 1 h. The reaction mixture was extracted with Et_2O (1 ml × 2), and the aqueous layer was concentrated under reduced pressure. The residue was dissolved in H_2O for chiral HPLC analysis using a SUMICHIRAL OA-5000 column (Sumitomo Chemical Industry, 0.46 × 15 cm; flow rate 1.0 ml/min; UV detection at 254 nm; eluent: 2.0 mM $CuSO_4$ in H_2O/CH_3OH, 98 : 2). Retention times of standard 3-(*S*)- and 3-(*R*)-*N*,*N*-dimethylamino-3-phenylpropanoic acids[9] were 14.7 and 15.9 min, respectively, and that of 3-*N*,*N*-dimethylamino-3-phenylpropanoic acid contained in the hydrolysate of **3** was found to be 15.9 min.

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