Nine New Sesquiterpenes from *Dendrobium nobile*

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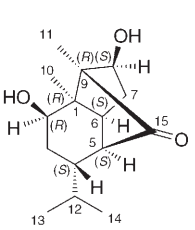
Nine new sesquiterpenes, *i.e.*, dendronobilins A–I (**1**–**9**), with copacamphane-type (**1**), picrotoxane-type (**2**–**6**), muurolene-type (**7**), alloaromadendrane-type (**8**), and cyclocopacamphane-type (**9**) skeletons, were isolated from the 60% EtOH extract of the stems of *Dendrobium nobile*. Their structures were established as (1*R*,2R,4*S*,5*S*,6*S*,8S,9*R*)-2,8-dihydroxycopacamphan-15-one (**1**), (2β,3β,4β,5β)-2,4,11-trihydroxypicrotoxano-3(15)-lactone (**2**), (2β,3β,5β,9α,11β)-2,11-epoxy-9,11,13-trihydroxypicrotoxano-3(15)-lactone (**3**), (2β,3β,5β,12*R**)-2,11,13-trihydroxypicrotoxano-3(15)-lactone (**4**), (2β,3β,5β,12*S**)-2,11,13-trihydroxypicrotoxano-3(15)-lactone (**5**), (2β,3β,5β,9α)-9,10-cyclo-2,11,13-trihydroxypicrotoxano-3(15)-lactone (**6**), (9β,10α)-muurol-4-ene-9,10,11-triol (**7**), (10α)-alloaromadendrane-10,12,14-triol (**8**), and (5β)-cyclocopacamphane-5,12,15-triol (**9**) on the basis of spectroscopic analysis. The absolute configuration of compound **1** was tentatively assigned as (1*R*,2*R*,4*S*,5*S*,6*S*,8*S*,9*R*) according to its CD spectrum and the octant rule. Compounds **1** and **4**–**9** were inactive in our preliminary *in vitro* immunomodulatory bioassay.

Introduction. – The genus *Dendrobium* includes about 1100 species widely distributed throughout Asia, Europe, and Oceania [1]. In China, there are 74 species and 2 variations, and several of them are used in traditional or folk medicine as a Yin tonic to nourish the stomach and promote the production of body fluid [2]. *Dendrobium nobile* LINDL. is one of the most famous *Dendrobium* plants and has been recorded in the Chinese Pharmacopeia (2005 edition) as one of the original materials of 'Shi Hu'. A series of chemical components including alkaloids, bibenzyls, phenanthrenes, sesquiterpenes, and sesquiterpene glycosides have been previously identified from this plant, and some of them exhibited antitumor, antimutagenic, and immunomodulatory activities [3–6]. In our previous study, some antioxidant bibenzyl derivatives and fluorenones have been isolated [7][8]. In continuation of the chemical and pharmacological investigation on *D. nobile*, nine new sesquiterpenes with the copacamphane-type (**1**), picrotoxane-type (**2**–**6**), muurolene-type (**7**), alloaromadendrane-type (**8**), and cyclocopacamphane-type (**9**) skeletons, were isolated from the

60% EtOH extract of the stems of this plant. In this paper, the isolation and structure elucidation of these nine compounds, and the result of an *in vitro* immunomodulatory bioassay of some compounds are reported.



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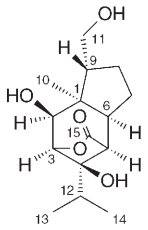

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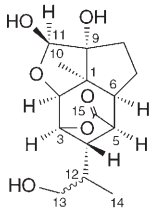

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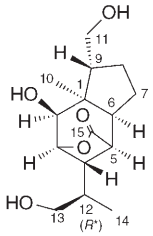

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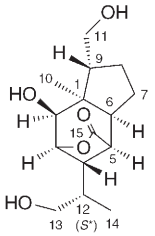

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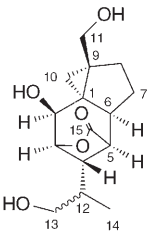

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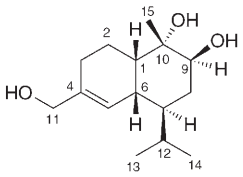

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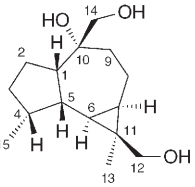

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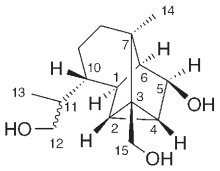

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Results and Discussion. – Compound **1** was obtained as a white amorphous powder. The HR-EI-MS (*m*/*z* 252.1737 (M^+)) and NMR analysis revealed the molecular formula $C_{15}H_{24}O_3$. Absorptions at 3464 and 1708 cm^{-1} in the IR spectrum of **1** were ascribed to OH and C=O groups, respectively. The ^{1}H- and ^{13}C-NMR data (*Tables 1* and *2*) were compatible with the proposed structure. The NOESY data established the relative configuration of **1** and the CD spectrum showed a positive *Cotton* effect at 294 nm and a negative *Cotton* effect at 211 nm. Application of the octant rule tentatively gave the configuration (*R*) at C(1), C(2), and C(9), and the configuration (*S*) at C(4), C(5), C(6), and C(8)[1]). Based on the above evidence, compound **1** was established as (1*R*,2*R*,4*S*,5*S*,6*S*,8*S*,9*R*)-2,8-dihydroxycopacamphan-15-one, which is a new sesquiterpene named dendronobilin A[1]).

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[1]) Trivial numbering; for systematic names, see *Exper. Part.*

Table 1. *^{1}H-NMR* (400 MHz) *Data of Compounds* **1**–**3**[1]). δ in ppm, J in Hz.

	1 (CD_3OD)	**2** (CD_3OD)	**3** (CD_3OD)	**3** ((D_6)DMSO)
H–C(2)	3.44 (*t*, *J* = 3.8)	3.74 (*d*, *J* = 1.5)	4.04 (*d*, *J* = 4.3)	3.88 (*d*, *J* = 4.3)
CH_2(3) or H–C(3)	1.73–1.79 (*m*, H_α), 1.44–1.51 (*m*, H_β)	4.39 (*t*, *J* = 1.4)	4.73 (*t*, *J* = 4.7)	4.66 (*t*, *J* = 4.7)
H–C(4)	1.33–1.36 (*m*)	–	2.37–2.42 (*m*)	2.25–2.31 (*m*)
H–C(5)	2.21 (*dd*, *J* = 4.0, 1.3)	2.27–2.29 (*m*)	2.66 (*dd*, *J* = 6.8, 4.0)	2.56–2.59 (*m*)
H–C(6)	2.23 (*d*, *J* = 4.8)	2.11–2.16 (*m*)	2.27–2.31 (*m*)	2.15–2.19 (*m*)
CH_2(7)	2.44–2.51 (*m*, H_α), 1.09 (*dd*, *J* = 13.6, 3.4, H_β)	2.07–2.10 (*m*, H_α), 1.83–1.85 (*m*, H_β)	1.87–1.92 (*m*, H_α), 2.14–2.23 (*m*, H_β)	1.71–1.75 (*m*, H_α), 2.03–2.07 (*m*, H_β)
H–C(8) or CH_2(8)	4.20 (*dd*, *J* = 9.6, 3.4)	1.25–1.34 (*m*, H_α), 1.91–1.93 (*m*, H_β)	1.72–1.77 (*m*, H_α), 1.63–1.70 (*m*, H_β)	1.55–1.56 (*m*, H_α), 1.53–1.54 (*m*, H_β)
H–C(9)		2.64–2.72 (*m*)		
Me(10)	1.06 (*s*)	1.15 (*s*)	1.37 (*s*)	1.24 (*s*)
Me(11), CH_2(11), or H–C(11)	0.95 (*s*)	3.58 (*dd*, *J* = 10.7, 8.7), 3.39 (*dd*, *J* = 10.7, 5.3)	5.07 (*s*)	4.96 (br. *s*)
H–C(12)	2.05–2.11 (*m*)	1.88–1.91 (*m*)	2.05–2.10 (*m*)	1.86–1.92 (*m*)
Me(13) or CH_2(13)	0.96 (*d*, *J* = 5.0)	0.96 (*d*, *J* = 6.7)	3.51 (*dd*, *J* = 10.9, 5.2), 3.44 (*dd*, *J* = 10.9, 5.5)	3.27–3.34 (*m*)
Me(14)	0.88 (*d*, *J* = 5.0)	0.91 (*d*, *J* = 6.7)	1.01 (*d*, *J* = 6.7)	0.90 (*d*, *J* = 6.6)
OH–C(9)				4.21 (*s*)
OH–C(11)				6.50 (*s*)

Table 2. *^{13}C-NMR* (100 MHz) *Data of Compounds* **1**–**4**[1]). δ in ppm.

	1 (CD_3OD)	**2** (CD_3OD)	**3** (CD_3OD)	**3** ((D_6)DMSO)	**4** (CD_3OD)
C(1)	55.8	49.9	51.3	49.5	51.0
C(2)	72.0	75.2	82.6	80.2	73.4
C(3)	31.2	90.1	80.8	78.6	85.7
C(4)	45.1	82.2	48.8	46.9	49.1
C(5)	56.5	55.1	44.7	42.8	47.3
C(6)	38.3	48.8	44.9	42.9	46.9
C(7)	36.4	26.5	30.2	28.9	26.5
C(8)	76.4	27.9	37.8	36.4	27.9
C(9)	64.9	46.9	91.7	89.7	46.8
C(10)	14.7	24.2	26.2	25.6	23.7
C(11)	7.7	63.5	99.6	97.8	63.4
C(12)	30.1	29.9	33.2	31.5	33.8
C(13)	21.3	15.3	67.2	65.2	66.5
C(14)	21.4	16.2	15.4	15.0	14.6
C(15)	223.1	181.1	181.1	178.5	181.8

In the ^{13}C-NMR and DEPT spectra of **1**, 15 C-signals due to four Me, two CH_2, and six CH groups (two oxygenated), two quaternary C-atoms, and one ketone C=O group were observed.

Analysis of the ^{1}H,^{1}H-COSY and HSQC data led to the deduction of the fragments C(2)–C(3)–C(4), C(6)–C(7)–C(8), and C(4)–C(12)(–C(14))–C(13). In the HMBC plot, the ^{1}H,^{13}C long-range correlations Me(10)/C(1), C(2), C(6), and C(9), Me(11)/C(1), C(8), C(9), and C(15), and H–C(5)/C(1), C(3), C(4), C(7), and C(15) were observed, consistent with a copacamphane sesquiterpene skeleton [9]. The NOE correlations H–C(2)/Me(10) and Me(11), H–C(6)/H_α–C(3),

H–C(5), H_α–C(7), Me(10), H–C(12), and Me(14), H–C(8)/H_α–C(7), Me(10), and Me(11) revealed the relative configuration of **1**.

Compound **2** was obtained as a white amorphous powder. The molecular formula $C_{15}H_{24}O_5$ was determined by HR-TOF-MS (*m/z* 307.1533 ($[M+Na]^+$)) and NMR analysis (*Tables 1* and *2*). The structure of **2** was established as (2*β*,3*β*,4*β*,5*β*)-2,4,11-trihydroxypicrotoxano-3(15)-lactone, a new sesquiterpene designated as dendronobilin B[1]).

In the ^{13}C-NMR and DEPT spectra of **2**, 15 C-signals belonging to three Me, three CH_2 (one oxygenated), and six CH groups (two oxygenated), two quaternary C-atoms (one oxygenated), and one lactone C=O group were observed. Analysis of the ^{1}H,^{1}H-COSY and HSQC data allowed the deduction of the fragments C(2)–C(3), C(5)–C(6)–C(7)–C(8)–C(9)–C(11), and C(13)–C(12)–C(14). The ^{1}H,^{13}C long-range correlations Me(10)/C(1), C(2), C(6), and C(9), H–C(3)/C(4), C(5), and C(15), H–C(5)/C(3), C(4), and C(15), H–C(6)/C(15), Me(13)/C(4), and Me(14)/C(4) in the HMBC plots suggested a picrotoxane sesquiterpene skeleton [10][11]. The relative configuration of **2** was determined on the basis of its NOESY plot, in which the NOE correlations H–C(2)/H–C(3), Me(10), H–C(12) and Me(13), H–C(6)/H–C(5), H_α–C(8), Me(10), H–C(12), and Me(14), and Me(10)/H_α–C(7), and CH_2(11) were observed.

Compound **3** was obtained as colorless oil. Its molecular formula was established as $C_{15}H_{22}O_6$ by HR-TOF-MS giving a quasimolecular-ion peak $[M+Na]^+$ at *m/z* 321.1326 and by NMR analysis (*Tables 1* and *2*). On the basis of further spectroscopic data, compound **3** was established as (2*β*,3*β*,5*β*,9*α*,11*β*)-2,11-epoxy-9,11,13-trihydroxypicrotoxano-3(15)-lactone, a new sesquiterpene, named dendronobilin C[1]).

The ^{13}C-NMR and DEPT spectra of **3** exhibited 15 C-signals due to two Me, three CH_2 (one oxygenated), and seven CH groups (three oxygenated), two quaternary C-atoms (one oxygenated), and one lactone C=O group. Analysis of the ^{1}H,^{1}H-COSY and HSQC data enabled the establishment of the fragments C(2)–C(3)–C(4)–C(5)–C(6)–C(7)–C(8) and C(4)–C(12)(–C(13))–C(14). The constitution of **3** was deduced on the basis of the HMBC plot, in which the ^{1}H,^{13}C long-range correlations Me(10)/C(1), C(2), C(6), and C(9), H–C(11)/C(1), C(2), C(8), C(9), H–C(3)/C(15), and H–C(6)/C(15) were observed. Thus compound **3** possesses a picrotoxane sesquiterpene skeleton [10][11]. In the NOESY plot, the NOE correlations H–C(2)/H–C(3), Me(10), and H–C(12), H–C(6)/H–C(5), H_α–C(7), and Me(10), and H_β–C(8)/H_β–C(7) allowed to determine the relative configuration of all chiral C-atoms of **3** except for C(9) and C(12). To elucidate the configuration at C(9), the NMR spectra were measured in (D_6)DMSO; interpretation of the ^{1}H,^{1}H-COSY, HSQC, and HMBC data resulted in the assignments of all H- and C-signals. Thus, the OH–C(9) signal was observed at δ 4.21 in the ^{1}H-NMR spectrum. The NOE correlations OH–C(9)/Me(10) and H–C(11) suggested that OH–C(9) had α configuration. However, we could not determine the configuration at C(12) since it lies outside of the ring system with a freely rotating C(4)–C(12) single bond, and the NMR data are insufficient to establish its configuration.

Compound **4** was obtained as colorless oil. The HR-TOF-MS (*m/z* 307.1534 ($[M+Na]^+$)) and NMR analysis (*Tables 2* and *3*) indicated the molecular formula $C_{15}H_{24}O_5$. The structure of **4** was established as (2*β*,3*β*,5*β*,12*R**)-2,11,13-trihydroxypicrotoxano-3(15)-lactone, a new sesquiterpene which we name dendronobilin D[1]).

The ^{13}C-NMR and DEPT spectra of **4** showed the presence of two Me, four CH_2 (two oxygenated), and seven CH groups (two oxygenated), one quaternary C-atom and one lactone C=O group. The

Table 3. *^{1}H-NMR* (400 MHz, CD_3OD) *Data of Compounds* **4**–**6**[1]). δ in ppm, J in Hz.

	4	**5**	**6**
H–C(2)	3.70 (*d*, *J* = 1.5)	3.89 (*d*, *J* = 1.5)	4.47 (br. *s*)
H–C(3)	4.55 (*d*, *J* = 5.6)	4.69–4.71 (*m*)	4.69–4.71 (*m*)
H–C(4)	2.28–2.30 (*m*)	2.28–2.31 (*m*)	2.34–2.36 (*m*)
H–C(5)	2.46 (*t*, *J* = 3.8)	2.31–2.34 (*m*)	2.38–2.39 (*m*)
H–C(6)	2.32–2.35 (*m*)	2.21 (*dt*, *J* = 9.4, 2.4)	2.40–2.43 (*m*)
CH_2(7)	2.03–2.12 (*m*, H_α), 1.83–1.88 (*m*, H_β)	2.04–2.14 (*m*, H_α), 1.78–1.84 (*m*, H_β)	1.63–1.68 (*m*)
CH_2(8)	1.23–1.33 (*m*, H_α), 1.89–1.93 (*m*, H_β)	1.25–1.33 (*m*, H_α), 1.90–1.96 (*m*, H_β)	1.60–1.63 (*m*, H_α), 2.02–2.05 (*m*, H_β)
H–C(9)	2.62–2.67 (*m*)	2.62–2.66 (*m*)	
Me(10)	1.16 (*s*)	1.16 (*s*)	0.87 (*d*, *J* = 4.3), 0.61 (*d*, *J* = 4.7)
CH_2(11)	3.57 (*dd*, *J* = 10.9, 8.8), 3.39 (*dd*, *J* = 10.9, 5.5)	3.57 (*dd*, *J* = 10.8, 8.8), 3.39 (*dd*, *J* = 10.9, 5.2)	3.92 (*d*, *J* = 11.6), 3.70 (*d*, *J* = 11.6)
H–C(12)	1.76–1.84 (*m*)	1.83–1.89 (*m*)	2.05–2.08 (*m*)
CH_2(13)	3.50 (*dd*, *J* = 11.1, 4.8), 3.40 (*dd*, *J* = 11.1, 5.8)	3.51 (*dd*, *J* = 5.7, 2.5)	3.54 (*d*, *J* = 5.5)
Me(14)	1.01 (*d*, *J* = 6.6)	0.94 (*d*, *J* = 6.6)	0.94 (*d*, *J* = 6.6)

^{13}C-NMR data of **4** were similar to those of **2**, except for the loss of a Me group and an OH-substituted quaternary C-atom and the appearance of a CH and a CH_2OH group. Analysis of the ^{1}H,^{1}H-COSY, HSQC, and HMBC data of **4** also allowed to deduce a picrotoxane-type sesquiterpene skeleton similar to that of **2**. The difference between these two compounds lay in the substitution by an OH group at C(13) in **4** instead of at C(4) in **2**. The relative configuration of **4** was established by the NOE correlations H–C(2)/H–C(3), Me(10), and H–C(12), H–C(6)/H–C(5), H_α–C(7), and Me(10), H_α–C(8)/H_α–C(7), and CH_2(11). But we still could not determine the configuration at C(12) for the same reasons as those mentioned above in the case of **3**.

Compound **5** was obtained as a white amorphous powder. The HR-TOF-MS (*m*/*z* 307.1544 ($[M+Na]^+$)) and NMR analysis (*Tables 3* and *4*) revealed the molecular formula $C_{15}H_{24}O_5$. The IR spectrum of **5** indicated the presence of OH group(s) (3321 cm^{-1}) and C=O group(s) (1770 cm^{-1}). The ^{1}H- and ^{13}C-NMR data of **5** were very similar to those of **4**. Analysis of the ^{1}H,^{1}H-COSY, HSQC, and HMBC data of **5** established an identical planar structure to that of **4**. A further comparison of the ^{13}C-NMR data of these two compounds revealed differences in the chemical shifts of C(4), C(12), C(13), and C(14), which indicated that **5** was an epimer at C(12) of **4**. Compound **5** was thus established to be (2*β*,3*β*,5*β*,12*S**)-2,11,13-trihydroxypicrotoxano-3(15)-lactone, a new sesquiterpene named dendronobilin E[1]).

Compound **6** was obtained as colorless oil. The molecular formula $C_{15}H_{22}O_5$ was determined by HR-TOF-MS (*m*/*z* 305.1359 ($[M+Na]^+$)) and NMR analysis (*Tables 3* and *4*). The structure of compound **6** was established as (2*β*,3*β*,5*β*,9*α*)-9,10-cyclo-2,11,13-trihydroxypicrotoxano-3(15)-lactone, a new picrotoxane sesquiterpene designated as dendronobilin F[1]).

In the ^{13}C-NMR and DEPT spectra of **6**, 15 C-signals belonging to one Me, five CH_2 (two oxygenated), and six CH groups (two oxygenated), two quaternary C-atoms, and one lactone C=O

Table 4. *^{13}C-NMR* (100 MHz, CD_3OD) *Data of Compounds* **5**–**9**. δ in ppm.

	5	**6**	**7**	**8**	**9**
C(1)	51.0	36.1	49.6	54.3	42.6
C(2)	73.6	68.1	23.0	25.3	20.1
C(3)	85.9	85.7	27.7	30.1	31.0
C(4)	49.9	50.4	139.8	39.5	23.8
C(5)	47.4	47.7	123.9	40.8	78.9
C(6)	46.7	40.1	40.4	24.8	52.6
C(7)	26.6	26.3	46.2	30.4	45.3
C(8)	27.9	30.8	30.8	19.2	36.0
C(9)	46.9	39.9	79.0	32.9	24.1
C(10)	23.7	14.6	76.3	77.2	39.3
C(11)	63.4	65.6	67.3	25.4	40.2
C(12)	34.1	33.4	27.0	64.0	66.5
C(13)	66.7	66.6	15.4	24.4	15.3
C(14)	15.3	15.3	21.9	71.5	22.3
C(15)	181.5	180.9	14.1	16.9	60.3

group were observed. Analysis of the ^{1}H,^{1}H-COSY and HSQC data of **6** enabled the deduction of the fragments C(2)–C(3)–C(4)–C(5), C(6)–C(7)–C(8), and C(4)–C(12)(–C(13))–C(14). The ^{1}H,^{13}C long-range correlations H–C(3)/C(1) and C(15), H–C(2)/C(9) and C(10), CH_2(10)/C(6), C(8), and C(11), H–C(4)/C(6), and H–C(6)/C(15) in the HMBC plot led to the planar structure of **6**. The relative configuration, except for C(12), was determined by the NOE correlations H–C(2)/H–C(3), CH_2(10), and H–C(12), H–C(5)/H–C(12), and CH_2(10)/H–C(6) and H_β–C(8).

Compound **7** was obtained as colorless oil. The HR-TOF-MS (*m*/*z* 277.1787 ([*M* + Na]$^+$)) and NMR analysis (*Tables 4* and *5*) revealed the molecular formula $C_{15}H_{26}O_3$. Compound **7** was established to be (9β,10α)-muurol-4-ene-9,10,11-triol, a new sesquiterpene designated as dendronobilin G[1]).

The ^{13}C-NMR and DEPT spectra of **7** exhibited 15 C-signals due to three Me, four sp^3 CH_2 (one oxygenated), and five sp^3 CH groups (one oxygenated), one sp^3 quaternary C-atom (oxygenated), one sp^2 CH group, and one sp^2 quaternary C-atom. Analysis of the ^{1}H,^{1}H-COSY and HSQC data led to the establishment of the fragments C(2)–C(3), C(1)–C(6)–C(7)–C(8)–C(9), and C(13)–C(12)–C(14). In the HMBC plot, the ^{1}H,^{13}C long-range correlations Me(13)/C(7), Me(14)/C(7), Me(15)/C(1), C(9), and C(10), CH_2(11)/C(3), C(4), and C(5), H–C(5)/C(1), C(6), and C(7), and H–C(1)/C(2) and C(3) were observed. Thus, compound **7** was identified as a sesquiterpene possessing a muurolene-type skeleton [12][13]. The relative configuration of **7** was determined by the NOE correlations H–C(1)/H_β–C(2) and Me(15), H–C(6)/H_β–C(8), Me(13), and Me(15), H_α–C(8)/H–C(9) and Me(14), and H_β–C(8)/H–C(12).

Compound **8** was obtained as a white amorphous powder. Its molecular formula was established as $C_{15}H_{26}O_3$ by HR-TOF-MS giving a quasimolecular-ion peak [*M* + Na]$^+$ at *m*/*z* 277.1790, and by NMR analysis (*Tables 4* and *5*). An OH absorption was observed at 3313 cm^{-1} in the IR spectrum. Compound **8** was established as (10α)-alloaromadendrane-10,12,14-triol. An alloaromadendrane-type sesquiterpene with the opposite configuration at C(11) has been isolated from *Dendrobium moniliforme* before [11]. To the best of our knowledge, compound **8** is a new sesquiterpene, and was assigned the name dendronobilin H[1]).

Table 5. *^{1}H-NMR* (400 MHz, CD_3OD) *Data of Compounds* **7**–**9**[1]). δ in ppm, J in Hz.

	7	**8**	**9**
H–C(1)	1.18–1.22 (*m*)	1.93–1.96 (*m*)	1.80 (br. *s*)
CH_2(2) or	1.26–1.32 (*m*, H_α),	1.50–1.55 (*m*, H_α),	1.15–1.17 (*m*)
H–C(2)	2.09–2.10 (*m*, H_β)	1.63–1.66 (*m*, H_β)	
CH_2(3)	2.11–2.13 (*m*),	1.25–1.34 (*m*, H_α),	–
	2.00–2.08 (*m*)	1.79–1.85 (*m*, H_β)	
H–C(4)	–	1.97–2.01 (*m*)	1.23–1.25 (*m*)
H–C(5)	5.77 (*s*)	1.92–1.95 (*m*)	3.74 (br. *s*)
H–C(6)	1.75–1.79 (*m*)	0.29 (*t*, $J = 9.1$)	1.26–1.27 (*m*)
H–C(7)	1.16–1.18 (*m*)	0.75–0.82 (*m*)	–
CH_2(8)	1.72–1.75 (*m*, H_α),	1.73–1.79 (*m*, H_α),	1.41–1.44 (*m*, H_α),
	1.10–1.14 (*m*, H_β)	1.60–1.63 (*m*, H_β)	1.68–1.72 (*m*, H_β)
H–C(9) or	3.39–3.43 (*m*)	1.46–1.50 (*m*, H_α),	1.44–1.47 (*m*)
CH_2(9)		1.55–1.59 (*m*, H_β)	
H–C(10)	–	–	1.35–1.40 (*m*)
CH_2(11) or	3.92 (br. *s*)		1.46–1.49 (*m*)
H–C(11)			
H–C(12) or	2.19–2.23 (*m*)	3.69 (*d*, $J = 11.4$),	3.57 (*dd*, $J = 10.7, 4.2$),
CH_2(12)		3.61 (*d*, $J = 11.3$)	3.38 (*dd*, $J = 10.7, 6.5$)
Me(13)	0.81 (*d*, $J = 6.9$)	1.10 (*s*)	0.92 (*d*, $J = 6.8$)
Me(14) or	0.94 (*d*, $J = 6.9$)	3.31 (*d*, $J = 11.2$),	1.24 (*s*)
CH_2(14)		3.21 (*d*, $J = 11.1$)	
Me(15) or	1.01 (*s*)	0.97 (*d*, $J = 6.6$)	3.75 (br. *s*)
CH_2(15)			

In the ^{13}C-NMR and DEPT spectra of **8**, 15 C-signals due to two Me, six CH_2 (two oxygenated), and five CH groups and two quaternary C-atoms (one oxygenated) were observed. Analysis of the ^{1}H,^{1}H-COSY and HSQC data of **8** enabled the deduction of the fragment C(9)–C(8)–C(7)–C(6)–C(5)–C(4)(–C(15))–C(3)–C(2)–C(1)–C(5). The HMBC plot exhibited the ^{1}H,^{13}C long-range correlations Me(13)/C(6), C(7), C(11) and C(12), and CH_2(14)/C(1), C(9), and C(10), consistent with an alloaromadendrane-type sesquiterpene skeleton [5]. The following NOE correlations were observed: H_β–C(3)/H–C(1), H_β–C(2), H–C(4), and H–C(5), H–C(6)/H–C(7), Me(13), and Me(15), H_α–C(8)/H–C(7) and H_α–C(9), CH_2(12)/H–C(5), and CH_2(14)/H–C(1).

Compound **9** was obtained as a white amorphous powder. The molecular formula $C_{15}H_{24}O_3$ was determined by HR-TOF-MS (*m*/*z* 275.1644 ($[M+Na]^+$)) and NMR analysis (*Tables 4* and *5*). The absorption at 3363 cm^{-1} in the IR spectrum of **9** indicated the presence of OH group(s). Compound **9** was established as (5β)-cyclocopacamphane-5,12,15-triol, which is a new sesquiterpene named dendronobilin I[1]).

In the ^{13}C-NMR and DEPT spectra of **9**, 15 C-signals belonging to two Me, four CH_2 (two oxygenated), and seven CH groups (one oxygenated) and two quaternary C-atoms were observed. Analysis of the ^{1}H,^{1}H-COSY and HSQC data enabled the deduction of the fragments C(10)–C(1)–C(2)–C(4)–C(5)–C(6)–C(1) and C(8)–C(9)–C(10)–C(11)(–C(13))–C(12). In the HMBC plot, the ^{1}H,^{13}C long-range correlations Me(14)/C(3), C(6), C(7), and C(8), and CH_2(15)/C(2), C(3), C(4), and C(7) were observed, which suggested a cyclocopacamphane sesquiterpene skeleton [5]. The relative configuration, except for C(11), was determined by the NOE correlations H–C(1)/H–C(5), H–C(6), and H–C(11), H–C(6)/H_α–C(8), and CH_2(15)/H–C(2), H–C(4), and H_β–C(8).

Previously, many sesquiterpene glycosides isolated from *Dendrobium* plants were reported to have immunomodulatory activity [6][9][14]. However, in our preliminary *in vitro* bioassay, the sesquiterpenes **1** and **4**–**9** were inactive, suggesting that the sugar unit of the corresponding glycosides plays an important role in its immunomodulatory activity.

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Experimental Part

General. Anal. and prep. HPLC: *Shimadzu Pak* with RI detector; *Shim-pack-VP-ODS* column (4.6 × 250 mm) and *Shim-pack-PREP-ODS* column (20 × 250 mm), resp. Column chromatography (CC): silica gel *H60* (*Qingdao Haiyang Chemical Group Corp.*, Qingdao, China), *Sephadex LH-20* (*Amersham Biosciences AB*), or *ODS* (60–80 μm; *YMC*) as packing materials. Anal. TLC: silica gel *G*. Optical rotations: *Jasco P-1020* polarimeter. UV Spectra: *Shimadzu UV2401PC-UV-Vis* spectrophotometer; in MeOH. CD Spectra: λ ([θ]) in nm. IR Spectra: *Shimadzu FTIR-8400* spectrophotometer; KBr disks; in cm^{-1}. NMR Spectra: *Bruker Avance-400* spectrometer; at 400 (^{1}H) and 100 MHz (^{13}C). MS: *Bruker Esquire-2000* spectrometer for ESI; *Finnigan MAT95* spectrometer for HR-EI; *Micromass* spectrometer for HR-TOF; in *m*/*z*.

Plant Material. The fresh stems of *D. nobile* were collected in Yunnan province in 2004 and identified by Ms. *Li-Ping Xiao* of the Hongkong Kadoorie Farm and Botanic Garden. A voucher specimen (YZXDN-2004) is deposited at the Key Laboratory for Research & Development of New Drugs from Traditional Chinese Medicine & Natural Products in Shenzhen, China.

Extraction and Isolation. The powdered air-dried stems of *Dendrobium nobile* (5 kg) were refluxed with 60% EtOH three times. After evaporation of EtOH, the aq. residue was extracted successively with AcOEt and BuOH. The AcOEt extract (63 g) was first subjected to CC (silica gel, $CHCl_3$/MeOH 100:0 → 0:100): *Fractions 1–12*. *Fr. 7* (6.9 g) was subjected to CC (*Sephadex LH-20*, $CHCl_3$/MeOH 1:1) and then to MPLC (silica gel, cyclohexane/AcOEt 8:2 → 0:1): *Fr. 7.1–7.9*. *Fr. 7.4* (1.3 g) was subjected to CC (*ODS*, MeOH/H_2O 4:6 → 7:3), and the fraction eluted with 40% MeOH was further purified by prep. HPLC (35% MeOH): **1** (25.7 mg). *Fr. 9* (3.8 g) was subjected to CC (*Sephadex LH-20*, $CHCl_3$/MeOH 1:1) and then to MPLC (silica gel, cyclohexane/acetone 8:2 → 0:1): *Fr. 9.1–9.7*. *Fr. 9.5* (1.3 g) was subjected to CC (*ODS*, MeOH/H_2O 3:7 → 6:4), and the fraction obtained with 50% MeOH was purified by prep. HPLC (45% MeOH): **7** (11.4 mg).

Fr. 10 (4.7 g) was subjected to CC (*Sephadex LH-20*, $CHCl_3$/MeOH 1:1) and then to MPLC (silica gel, cyclohexane/acetone 8:2 → 0:1): *Fr. 10.1–10.8*. *Fr. 10.3* (380 mg) was subjected to CC (*ODS*, MeOH/H_2O 3:7 → 7:3) and the fraction obtained with 40% and 50% MeOH was purified by prep. HPLC (48% and 55% MeOH): **2** (4.3 mg) and **8** (31.4 mg). *Fr. 10.5* (263 mg) was subjected to CC (*ODS*, MeOH/H_2O 3:7 → 7:3) and then purified by prep. HPLC (30% MeOH): **3** (5.8 mg) and **4** (11.2 mg). *Fr. 10.6* (948 mg) was subjected to CC (*ODS*, MeOH/H_2O 3:7 → 7:3), and **5**, **6**, and **9** (15.1, 148.6, and 76.0 mg, resp.) were finally obtained with 30% MeOH and purified by prep. HPLC (30%, 30%, and 37% MeOH, resp.).

Dendronobilin A (= *(1*R*,2*S*,3a*S*,4*S*,5*S*,7*R*,7a*R*)-Octahydro-2,7-dihydroxy-1,7a-dimethyl-5-(1-methylethyl)-1,4-methano-1*H*-inden-8-one*; **1**): White amorphous powder. $[\alpha]_D^{26} = +82.7$ (c = 1.0, MeOH). CD: 211 (−4765), 294 (+5862). IR (KBr): 3464, 1708. ^{1}H-NMR: *Table 1*. ^{13}C-NMR: *Table 2*. ESI-MS: 275 ($[M+Na]^+$), 251 ($[M-H]^-$). HR-EI-MS: 252.1737 (M^+, $C_{15}H_{24}O_3^+$; calc. 252.1725).

Dendronobilin B (= *(1*R*,4*S*,5*S*,5a*R*,6*R*,8a*S*,9*R*)-Octahydro-5,9-dihydroxy-6-(hydroxymethyl)-5a-methyl-9-(1-methylethyl)-1,4-methano-2*H*-cyclopent[*d*]oxepin-2-one*; **2**): White amorphous powder. $[\alpha]_D^{26} = +5.8$ (c = 0.5, MeOH). ^{1}H-NMR: *Table 1*. ^{13}C-NMR: *Table 2*. ESI-MS: 285 ($[M+H]^+$), 307

([$M+Na]^+$), 283 ([$M-H]^-$), 319 ([$M+Cl]^-$). HR-TOF-MS: 307.1533 ([$M+Na]^+$, $C_{15}H_{24}NaO_5^+$; calc. 307.1521).

Dendronobilin C (= *(2*R*,2a*R*,4a*S*,5*R*,8*R*,8a*S*,8b*S*,9*S*)-Octahydro-2,2a-dihydroxy-9-(2-hydroxy-1-methylethyl)-8b-methyl-5,8-methano-1,7-dioxacyclopent[*cd*]azulen-6(2*H*)-one*; **3**): Colorless oil. $[\alpha]_D^{27} = +16.2$ ($c = 0.6$, MeOH). ^{1}H-NMR: *Table 1*. ^{13}C-NMR: *Table 2*. ESI-MS: 321 ([$M+Na]^+$), 297 ([$M-H]^-$). HR-TOF-MS: 321.1326 ([$M+Na]^+$, $C_{15}H_{22}NaO_6^+$; calc. 321.1314).

Dendronobilin D (= *(1*R*,4*R*,5*S*,5a*R*,6*R*,8a*S*,9*S*)-Octahydro-5-hydroxy-6-(hydroxymethyl)-9-[(1*R**)-2-hydroxy-1-methylethyl)-5a-methyl-1,4-methano-2*H*-cyclopent[*d*]oxepin-2-one*; **4**): Colorless oil. $[\alpha]_D^{27} = +4.7$ ($c = 1.0$, MeOH). ^{1}H-NMR: *Table 3*. ^{13}C-NMR: *Table 2*. ESI-MS: 307 ([$M+Na]^+$), 283 [$M-H]^-$). HR-TOF-MS: 307.1534 ([$M+Na]^+$, $C_{15}H_{24}NaO_5^+$; calc. 307.1521).

Dendronobilin E (= *(1*R*,4*R*,5*S*,5a*R*,6*R*,8a*S*,9*S*)-Octahydro-5-hydroxy-6-(hydroxymethyl)-9-[(1*S**)-2-hydroxy-1-methylethyl)-5a-methyl-1,4-methano-2*H*-cyclopent[*d*]oxepin-2-one*; **5**). White amorphous powder. $[\alpha]_D^{27} = +14.4$ ($c = 1.0$, MeOH). IR (KBr): 3321, 1770. ^{1}H-NMR: *Table 3*. ^{13}C-NMR: *Table 4*. ESI-MS: 307 ([$M+Na]^+$), 319 ([$M+Cl]^-$). HR-TOF-MS: 307.1544 ([$M+Na]^+$, $C_{15}H_{24}NaO_5^+$; calc. 307.1521).

Dendronobilin F (= *(1*R*,4*R*,5*S*,5a*R*,6a*R*,8a*S*,9*S*)-Octahydro-5-hydroxy-6a-(hydroxymethyl)-9-(2-hydroxy-1-methylethyl)-1,4-methano-2*H*-cyclopropan[1,5]cyclopent[1,2-*d*]-oxepin-2-one*; **6**). Colorless oil. $[\alpha]_D^{27} = -10.2$ ($c = 1.0$, MeOH). ^{1}H-NMR: *Table 3*. ^{13}C-NMR: *Table 4*. ESI-MS: 305 ([$M+Na]^+$), 281 ([$M-H]^-$). HR-TOF-MS: 305.1359 ([$M+Na]^+$, $C_{15}H_{22}NaO_5^+$; calc. 305.1365).

Dendronobilin G (= *(1*S*,2*S*,4*S*,4a*R*,8a*S*)-1,2,3,4,4a,7,8,8a-Octahydro-6-(hydroxymethyl)-1-methyl-4-(1-methylethyl)naphthalene-1,2-diol*, **7**): Colorless oil. $[\alpha]_D^{26} = -19.1$ ($c = 1.0$, MeOH). ^{1}H-NMR: *Table 5*. ^{13}C-NMR: *Table 4*. ESI-MS: 277 ([$M+Na]^+$), 507 ([$2M-H]^-$). HR-TOF-MS: 277.1787 ([$M+Na]^+$, $C_{15}H_{26}NaO_3^+$; calc. 277.1780).

Dendronobilin H (= *(1*R*,1a*R*,4*R*,4a*S*,7*R*,7a*S*,7b*R*)-Decahydro-4-hydroxy-1,7-dimethyl-1*H*-cyclopro[e]azulene-1,4-dimethanol*; **8**). White amorphous powder. $[\alpha]_D^{26} = -5.7$ ($c = 1.0$, MeOH). IR (KBr): 3313. ^{1}H-NMR: *Table 5*. ^{13}C-NMR: *Table 4*. ESI-MS: 277 ([$M+Na]^+$), 253 ([$M-H]^-$). HR-TOF-MS: 277.1790 ([$M+Na]^+$, $C_{15}H_{26}NaO_3^+$; calc. 277.1780).

Dendronobilin I (= *(1*R*,2*R*,3*R*,3a*S*,4*R*,5*R*,7a*S*,8*R*)-Octahydro-3-hydroxy-1-(hydroxymethyl)-β,7a-dimethyl-1,2,4-metheno-1*H*-indene-5-ethanol*; **9**): White amorphous powder. $[\alpha]_D^{27} = +41.7$ ($c = 1.0$, MeOH). IR (KBr): 3363. ^{1}H-NMR: *Table 5*. ^{13}C-NMR: *Table 4*. ESI-MS: 275 ([$M+Na]^+$), 503 ([$2M-H]^-$). HR-TOF-MS: 275.1644 ([$M+Na]^+$, $C_{15}H_{24}NaO_3^+$; calc. 275.1623).

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