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New Indole Alkaloids from the Bark of *Nauclea orientalis*

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Four new alkaloids, nauclealines A (**1**) and B (**2**) and naucleosides A (**3**) and B (**4**), together with six known compounds, strictosamide (**5**), vincosamide (**6**), pumiloside (**7**), kelampayoside A, sitosterol, and sitosteryl β-D-glucoside, were isolated from the bark of *Nauclea orientalis*. The structures of **1**−**4** were elucidated using 1D and 2D NMR spectral methods, including COSY, DEPT, HMQC, $^{13}C-^{1}H$ HMBC, and $^{15}N-^{1}H$ HMBC.

The genus *Nauclea* (Rubiaceae) is a rich source of alkaloids. To date, more than 40 monoterpenoid indole alkaloids have been isolated from this genus.[1] Some of these alkaloids were reported to have antiproliferative,[2] antiparasitic,[3] and antimicrobial activities.[4,5] *Nauclea orientalis* L. is a tree found in Papua New Guinea, Indonesia, Peru, and Queensland (Australia). Villagers of the Central Province of Papua New Guinea use the bark of this tree to treat abdominal pain, animal bites, and wounds, while its leaves are used traditionally by coastal Australian Aborigines as a pain-killer and fish poison.[6] Erdelmeier et al.[2,6] have isolated several indole alkaloids from the ammoniacal and ethyl acetate extracts of *N. orientalis* leaves.

As part of our ongoing program to identify inhibitors of *Candida albicans*-secreted aspartic protease (SAP) from higher plants, the $CHCl_3$ and BuOH fractions from the EtOH extract of the bark of *N. orientalis* were found to be active in a screening assay for inhibition of SAP. The secreted aspartic proteases of *Candida albicans* have been shown to be a major virulence factor in *Candida* infections.[7] Inhibition of SAP has been proposed as a new approach in the treatment of candidosis.[8] Bioassay-guided fractionation of the active $CHCl_3$ and BuOH fractions utilizing the SAP assay resulted in the isolation of four new indole alkaloids, nauclealines A (**1**) and B (**2**) and naucleosides A (**3**) and B (**4**), as well as six known compounds, strictosamide (**5**), vincosamide (**6**), pumiloside (**7**), kelampayoside A, sitosterol, and sitosteryl β-D-glucoside. In this report we describe the isolation and structural determination of **1**−**4**.

Results and Discussion

Compounds **5** and **6** were proven to be identical to strictosamide (C-3 α-epimer) and vincosamide (C-3 β-epimer), respectively, by comparison of their spectral data with the reported values.[6] The ^{13}C NMR spectra allowed for the differentiation of the epimers as the chemical shifts of carbons C-5 and C-14 of the β-epimer displayed significant chemical shift differences of $\Delta\delta$ −3.2 ppm and $\Delta\delta$ +5.6 ppm, respectively, when compared to those of the α-epimer[6] (Table 2). The stereochemistry at C-3 could also be determined from the CD spectrum (see Experimental Section), whereby a positive Cotton effect in the region 250−300 nm indicated an α-configuration, while a negative Cotton effect was suggestive of a β-configuration.[9,10] Strictosamide (**5**) was the main constituent of the $CHCl_3$ fraction of *N. orientalis* bark.

Compound **7** was established as pumiloside,[11] which was previously isolated from the whole plant of *Ophiorrhiza*

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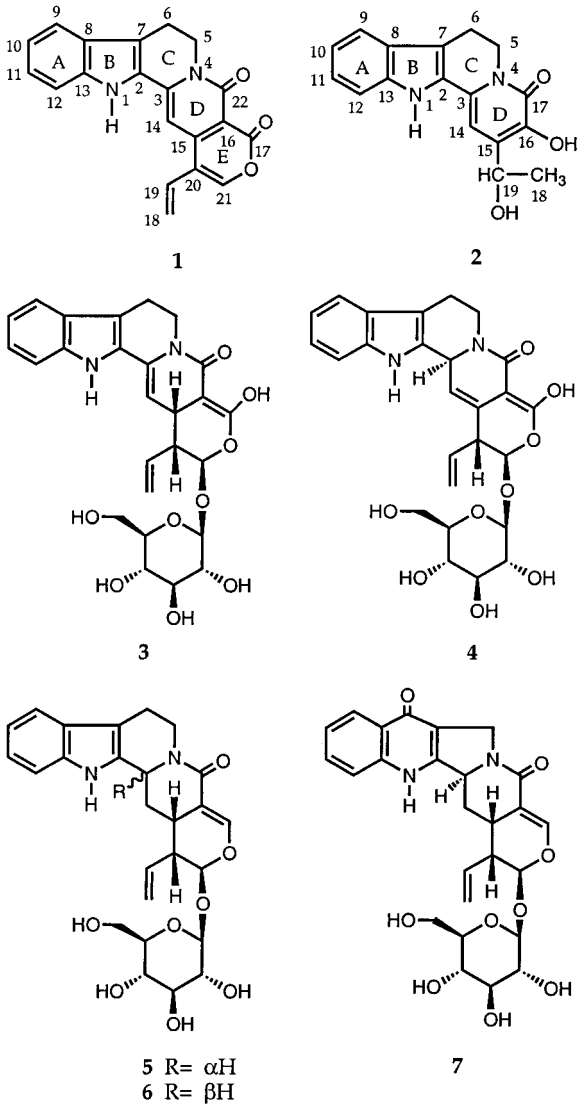

pumila (Rubiaceae). The ^{13}C NMR assignments were determined unambiguously and are reported for the first time. The stereochemistry at C-3 was indicated to be α from the positive Cotton effect in the region 300−350 nm.[11]

Kelampayoside A,[12] along with sitosterol[13] and sitosteryl β-D-glucoside, were identified by comparison of the spectral data with published values.

Compound **1** was isolated as a yellowish amorphous solid with strong fluoresence detected by TLC under UV light at 365 nm. Its IR spectrum showed an absorption band at 1642 cm^{-1} for an α,β-unsaturated carbonyl group. The HRESIMS of **1** displayed a molecular ion peak at *m/z* $[M + H]^+$ 331.1065, supporting a molecular formula of $C_{20}H_{14}N_2O_3$. Two nitrogen signals at δ 124.1 and 164.2, correlating with proton signals at δ 11.98 (H-1) and at δ 6.92 (H-14) and 3.11 (H-6), respectively, were observed in the $^{15}N-^{1}H$ HMBC spectrum and were assigned to N-1 and N-4. In the HMQC spectrum, the symmetrical triplets at δ 3.11 (2H, t, J = 7.0 Hz) and 4.31 (2H, t, J = 7.0 Hz) correlating with the carbon signals at δ 18.9 (t) and 40.1 (t) indicated the presence of C-5 and C-6 methylenes. Also, the proton signals at δ 5.43 (1H, dd, J = 10.6, 1.8 Hz, H-18_a), 5.69 (1H, dd, J = 17.0, 1.8 Hz, H-18_b), and 6.70 (1H, dd, J = 17.0, 10.6 Hz, H-19), correlating with the carbon signals at δ 117.9 (t) and 127.6 (d), were characteristic of a $-CH{=}CH_2$ functionality at C-20. The downfield region (δ 6.90−8.00) of the ^{1}H NMR spectrum of **1** showed six aromatic proton signals, of which two appeared as triplets at δ 7.11 and 7.30 and were attributed to H-10 and H-11, two were doublets at δ 7.48 and 7.66 and assigned to H-12 and H-9, and two were singlets at δ 6.92 and 7.94 and assigned to H-14 and H-21. In addition, a singlet at δ 11.98 indicated the presence of a NH unit. The structure of **1**, a new natural product named nauclealine A[14] (3,14,15,16,17,20-hexadehydro-16-ethenyloxayohimban-19,21-dione), was determined unambiguously using HMQC and HMBC NMR correlations (Figure 1).

Compound **2** has a molecular formula of $C_{17}H_{16}N_2O_3$, as deduced from the ESIMS ion at *m/z* $[M + H]^+$ 297 and from investigation of its ^{13}C and ^{15}N NMR spectra. In the ^{1}H NMR spectrum of **2**, two triplets at δ 7.09 (H-10) and 7.24 (H-11), two doublets at δ 7.47 (H-12) and 7.62 (H-9), and the two methylenes at δ 3.10 (2H, t, J = 6.5 Hz, H-6) and 4.37 (2H, m, H-5) verified the substitution pattern for rings A and C, while a singlet at δ 7.23 was assigned to H-14. The presence of a $-CH(OH)-CH_3$ group at C-15 was indicated by the proton signals at δ 1.50 (3H, d, J = 6.3 Hz, CH_3-18) and 5.31 (1H, q, J = 6.3 Hz, H-19), which correlated in the HMQC spectrum with the carbon signals at δ 25.1 (q, C-18) and 63.8 (d, C-19). The ^{1}H and ^{13}C NMR signals (Tables 1 and 2) of **2**, a new natural product named nauclealine B[15] (indolo[2,3-*a*]quinolizine-2-(1-hydoxyethyl)-3-hydroxy-4,6,7,12-tetrahydro-4-one), were unambiguously assigned using HMQC and HMBC NMR correlations (Figure 1).

Compound **3** was obtained as an orange-yellow amorphous solid. Its molecular formula of $C_{26}H_{28}N_2O_9$ was determined by HRESIMS. The IR spectrum of **3** showed an absorption band at 1645 cm^{-1} for an α,β-unsaturated carbonyl group. Analysis of the ^{1}H and ^{13}C NMR spectra indicated that **3** is related to strictosamide (**5**) and vincosamide (**6**) with an additional double bond in ring D. In the ^{13}C NMR spectrum of **3**, the signals for the C-3 (δ 137.8) and C-14 (δ 100.4) carbons appeared in the aromatic region, as compared with their positions in **5** and **6** at δ 52.7 (C-3) and C-14 (δ 25.7−31.3), clearly indicating the location of the double bond at C-3 to C-14. This was further confirmed by HMBC correlations (Figure 1). Acid hydrolysis of **3** yielded glucose, which was identified by co-TLC with an authentic sample. The signal of the anomeric proton of glucose at δ 4.86 (1H, d, J = 7.2 Hz) indicated a β-configuration. The sugar unit was attached to the C-21 position, as indicated by a correlation in the HMBC between the proton signal at δ 4.86 (1H, d, J = 7.2 Hz, H-1) and the carbon at δ 93.5 (C-21). The ^{1}H and ^{13}C NMR assignments (Tables 1 and 2) of **3**, a new natural product named naucleoside A[14](3,14,19,20-tetradehydro-16-ethenyl-17-(β-D-glucopyranosyloxy)-19-hydroxy-(15β,16α,17β)-oxayohimban-21-one), were unambiguously made using HMQC and HMBC NMR correlations (Figure 1).

Compound **4** was found to have the same molecular formula as **3**, $C_{26}H_{28}N_2O_9$. The UV and IR spectra are very similar (see Experimental Section), and detailed NMR analysis indicated that **4** differs from **3** only in the position of the double bond in ring D. In the ^{13}C NMR spectrum of **3**, the signals at δ 137.8 (s, C-3) and 29.0 (d, C-15) were replaced in **4** by signals at δ 57.0 (d, C-3) and 137.6 (s, C-15), indicating that in **4** the double bond is between C-14 and C-15. The C-3 configuration in **4** was determined as α by the positive Cotton effect observed in the region 250−300 nm.[9,10] Acid hydrolysis of **4** also afforded glucose, identified by co-TLC with an authentic sample. As in **3**, the glucose is of the β-configuration, as shown from the

Table 1. ^{1}H NMR Data for Compounds **1**−**4** and **7** (400 MHz in DMSO-d_6)

position	1	2	3	4	7
1	11.98 (1H, s)	11.86 (1H, s)	11.73 (1H, s)	11.73 (1H, s)	
3				5.06 (1H, br s)	4.72 (1H, d, 11.8 Hz)
5	4.31 (2H, t, 7.0 Hz)	4.37 (2H, m)	4.02 (1H, m)	4.07 (1H, m)	4.31 (1H, d, 14.1 Hz)
			4.53 (1H, m)	4.44 (1H, m)	4.46 (1H, d, 14.1 Hz)
6	3.11 (2H, t, 7.0 Hz)	3.10 (2H, t, 6.5 Hz)	3.07 (2H, m)	2.95−3.07 (2H, m)	
9	7.66 (1H, d, 8.0 Hz)	7.62 (1H, d, 7.9 Hz)	7.59 (1H, d, 8.0 Hz)	7.59 (1H, d, 7.8 Hz)	8.10 (1H, d, 8.0 Hz)
10	7.11 (1H, t, 8.0 Hz)	7.09 (1H, t, 7.9 Hz)	7.06 (1H, t, 8.0 Hz)	7.05 (1H, t, 7.8 Hz)	7.31 (1H, t, 8.0 Hz)
11	7.30 (1H, t, 8.0 Hz)	7.24 (1H, t, 7.9 Hz)	7.21 (1H, t, 8.0 Hz)	7.21 (1H, t, 7.8 Hz)	7.64 (1H, t, 8.0 Hz)
12	7.48 (1H, d, 8.0 Hz)	7.47 (1H, d, 7.9 Hz)	7.41 (1H, d, 8.0 Hz)	7.41 (1H, d, 7.8 Hz)	7.60 (1H, d, 8.0 Hz)
14	6.92 (1H, s)	7.23 (1H, s)	6.57 (1H, br s)	6.57 (1H, br s)	1.98 (1H, q, 11.8 Hz)
					2.50 (overlapped)
15			3.34 (1H, m)		3.28 (1H, m)
17					7.04 (1H, d, 2.3 Hz)
18	5.43 (1H, dd, 10.6/1.8 Hz)	1.50 (3H, d, 6.3 Hz)	5.24 (1H, d, 10.0 Hz)	5.25 (1H, d, 10.9 Hz)	5.32 (1H, d, 10.9 Hz)
	5.69 (1H, dd, 17/1.8 Hz)		5.26 (1H, d, 17.2 Hz)	5.27 (1H, d, 18.6 Hz)	5.44 (1H, d, 17.2 Hz)
19	6.70 (1H, dd, 17/10.6 Hz)	5.31 (1H, q, 6.3 Hz)	5.73 (1H, m)	5.73 (1H, m)	5.74 (1H, m)
20			3.34 (1H, m)	3.38 (1H, m)	2.62 (1H, m)
21	7.94 (1H, s)		5.41 (1H, br s)	5.41 (1H, br s)	5.39 (1H, br s)
1′			4.86 (1H, d, 7.2 Hz)	4.63 (1H, d, 8.1 Hz)	4.54 (1H, d, 7.9 Hz)
2′			3.04 (1H, m)	2.95−3.07 (1H, m)	2.98 (1H, m)
3′			3.04 (1H, m)	2.95−3.07 (1H, m)	3.18 (1H, m)
4′			3.04 (1H, m)	2.95−3.07 (1H, m)	3.04 (1H, m)
5′			3.17 (1H, m)	3.20 (1H, m)	3.18 (1H, m)
6′			3.34 (1H, m),	3.20 (1H, m)	3.42 (1H, m)
			3.62 (1H, m)	3.68 (1H, m)	3.69 (1H, d, 11.3)

Table 2. ^{13}C NMR Data for Compounds **1**−**7** (100 MHz in DMSO-d_6)[a]

carbon	1	2	3	4	5	5[b]	6	6[b]	7
2	126.8	127.7	127.2	127.2	134.6	134.9	134.3	134.7	150.3
3	143.3	136.3	137.8	57.0	52.7	55.2	52.7	54.8	59.5
5	40.1	40.2	40.0	40.0	42.5	44.9	39.3	41.3	47.5
6	18.9	19.1	19.0	18.9	20.7	22.2	21.0	22.2	112.9
7	117.1	114.4	113.8	113.8	108.5	110.4	108.0	109.4	173.1
8	125.4	125.3	125.2	125.2	127.0	128.8	126.9	128.0	125.3
9	120.1	119.6	119.7	119.5	117.7	118.8	118.1	119.0	124.7
10	120.2	119.8	120.0	119.7	118.8	120.3	119.0	120.2	123.3
11	124.9	124.3	124.1	124.1	121.1	122.6	121.4	122.6	131.6
12	112.2	111.8	111.9	111.9	111.4	112.4	111.4	112.2	118.7
13	139.1	138.3	138.3	138.2	135.7	137.8	136.5	138.3	140.6
14	93.5	93.7	100.4	99.8	25.7	27.4	31.3	32.7	28.3
15	149.0	147.4	29.0	137.6	23.5	25.0	25.9	27.4	23.7
16	103.1	149.0	117.1	118.8	107.6	109.3	107.6	109.2	109.0
17	156.8	161.1	147.3	144.0	146.7	149.3	147.0	149.1	145.2
18	117.9	25.1	119.5	118.9	120.0	120.8	120.4	120.7	120.6
19	127.6	63.8	134.5	135.5	133.4	134.4	133.0	134.0	132.5
20	115.1		47.0	45.8	42.9	44.8	42.6	44.5	43.6
21	149.2		93.5	94.7	95.8	98.2	95.3	97.5	94.9
22	158.4		158.7	158.7	163.5	167.2	163.0	166.1	164.1
1′			97.7	97.7	99.0	100.6	98.2	99.7	97.8
2′			74.4	73.4	72.7	74.3	73.4	74.9	73.2
3′			79.4	77.4	77.2	78.2	77.4	78.4	77.3
4′			70.4	69.9	69.9	71.4	70.3	71.7	70.1
5′			78.1	76.6	76.8	78.0	76.7	78.1	76.5
6′			60.8	61.1	61.0	62.7	61.3	62.8	61.1

[a] Assigments were based on DEPT, COSY, HMQC, and HMBC experiments. [b] Data were recorded in MeOH-d_4.

resonance of the anomeric proton at δ 4.63 (1H, d, J = 8.1 Hz). Analysis of the HMQC and HMBC spectral data allowed for the complete assignments of the proton and carbon signals of **4**[14] (14,15,19,20-tetradehydro-16-ethenyl-17-(β-D-glucopyranosyloxy)-19-hydroxy-(3α,16α,17β)-oxayohimban-21-one), a new natural product named naucleoside B.

Crude extracts were initially tested in the *Candida*-secreted aspartic proteases (SAP) inhibition assay[16] at one concentration of 200 μg/mL to identify active extracts (≥80% inhibition). Active extracts were confirmed by retesting at three concentrations (50, 10, 2 μg/mL). The aspartic protease inhibitor pepstatin A was used as a positive control (IC_{50} 0.006 μg/mL). The crude EtOH extract was tested in the SAP assay and found to have an IC_{50} of 3 μg/mL. As the extract was detannified, the tannin-free fraction had an IC_{50} of 16 μg/mL. Further purification through solvent partitioning resulted in two active fractions, a $CHCl_3$ fraction with an IC_{50} of 30 μg/mL and an *n*-butanol fraction with an IC_{50} of 3 μg/mL. The $CHCl_3$ fraction was chromatographed over a Si gel column, and fractions were combined according to their TLC similarities to yield five fractions, A−E. Fraction D had an IC_{50} of 40 μg/mL. Further chromatography of fraction D provided compounds **4**, **5**, and **6**. None of these compounds were active. The *n*-butanol fraction was further purified using reversed-phase chromatography to provide **7** and kelampayoside A, which were also inactive in the SAP assay. It appeared that the activity observed in the initial extract was the result of synergetic action of several components

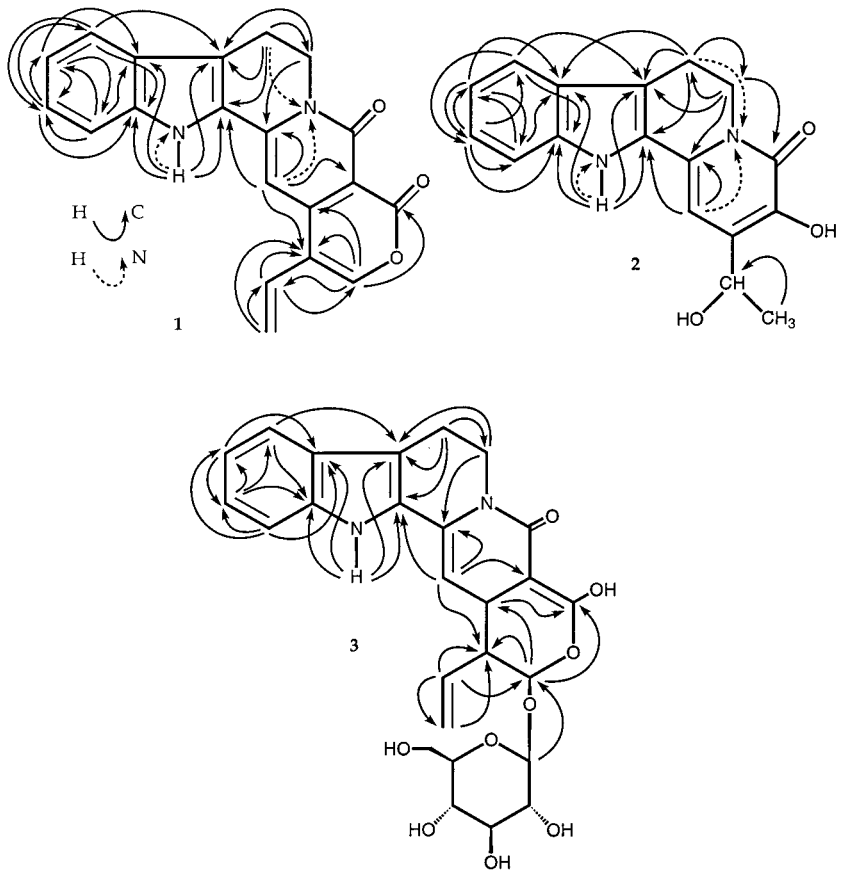

Figure 1. HMBC correlations of **1**−**3**.

such that as purification proceeded, the activity diminished, with none of the pure compounds being active.

Experimental Section

General Experimental Procedures. Melting points were measured with a Thomas-Hoover capillary melting point apparatus and were uncorrected. Optical rotations were determined on a JASCO DIP-370 digital polarimeter. UV spectra were recorded on a Hewlett-Packard 8435 spectrometer. CD spectra were recorded on a JASCO J 715 spectropolarimeter. IR spectra were obtained on an ATI Mattson Genesis Series FTIR spectrometer. The NMR spectra were recorded in DMSO-d_6 on a Bruker Avance DRX-400 spectrometer operating at 400 MHz for ^{1}H and 100 MHz for ^{13}C. Proton and carbon chemical shifts are relative to the internal standard TMS. Nitrogen (^{15}N) chemical shifts are reported relative to liquid ammonia using a nitromethane chemical shift of δ 380.2. HMQC, gradient ^{1}H−^{13}C HMBC (J, 10 Hz), and gradient ^{1}H−^{15}N HMBC experiments (J_{NH}, 5 Hz) were performed with standard pulse programs on a Bruker Advance DPX 500. HRESIMS were measured on a Bruker-Magnex BioAPEX 30es ion cyclotron high-resolution HPLC-FT spectrometer by direct injection into an electrospray interface. Si gel (40 μm, J. T. Baker) and RP silica gel (RP-18, 40 μm, J. T. Baker) were used for low-pressure chromatography. HPLC was performed using an ODS column (column A, μ-Bondapak C_{18}, 3.9 mm i.d. × 300 mm, 10 μm; column B, Phenomenex Prodigy ODS prep, 21.2 mm i.d. × 250 mm, 10 μm). TLC was performed on silica gel 60 F_{254} (EM Science) using $CHCl_3$−MeOH (4:1, solvent A) and EtOAc−acetone (2:3, solvent B) or reversed-phase KC_{18} F silica gel 60A (Whatman) using MeOH/H_2O (80:20, solvent C).

Plant Material. *Nauclea orientalis* L. (bark) was collected in June 1996 in Papua New Guinea and identified by Topal Rali. A voucher specimen is on deposit at the National Center for Natural Products Research, University of Mississippi (NWG 62).

Extraction and Isolation. The powdered material (230 g) was percolated with 95% EtOH (1500 mL × 5), and the alcoholic extracts were combined and evaporated to dryness (34 g, IC_{50} 3 μg/mL). Part of the ethanolic extract (26 g) was detannified by passage over a polyamide column and washing with MeOH. The MeOH wash (tannin-free fraction, 8.1 g, IC_{50} 16 μg/mL) was suspended in MeOH−H_2O (7:3) (150 mL) and then partitioned successively with hexane (100 mL × 3, 0.75 g), $CHCl_3$ (100 mL × 3, 3.6 g), EtOAc (100 mL × 3, 0.53 g), and *n*-BuOH (100 mL × 3, 1.5 g). The activity was concentrated in the chloroform (IC_{50} 30 μg/mL) and BuOH fractions (IC_{50} 3 μg/mL). Part of the chloroform fraction (3.0 g) was chromatographed over a silica gel column (250 g) using mixtures of $CHCl_3$−MeOH of increasing polarity (9:1 → 3:2). Based on TLC analysis, five combined fractions were obtained: A (340 mg), B (150 mg), C (150 mg), D (500 mg, IC_{50} 40 μg/mL), and E (1500 mg). Part of fraction D (400 mg) was rechromatographed over a silica gel column (100 g) using $CHCl_3$−MeOH (9:1 and 6:1, each 1000 mL) to afford 56 fractions. Fractions 19−22, 25−35, and 37−45 were combined to give pooled fractions D_1 (20 mg), D_2 (200 mg), and D_3 (20 mg), respectively. Fractions D_1, D_2, and D_3 were each purified by HPLC (column B, MeOH−H_2O, 88:12, 3 mL/min, UV 365 nm) to yield **4** (1.5 mg, t_R 19 min), **5** (126 mg, t_R 23 min), and **6** (1.7 mg, t_R 27 min). Part of A (100 mg) was chromatographed over a silica gel column (50 g) using $CHCl_3$−MeOH (15:1, 200 mL) to give 15 fractions, from which fractions 7−8 were combined and crystallized from $CHCl_3$−MeOH (1:1) to afford β-sitosterol (20 mg). Fraction B was separated on a Si gel column (80 g) using $CHCl_3$−EtOAc mixtures of increasing polarity (9:1, 4:1, 2:1, and 1:1, each 200 mL) to give 70 fractions. Fractions 26−30 were combined and then purified by HPLC (column A, MeOH−H_2O, 75:25, 0.8 mL/min, UV 365 nm) to give **1** (1.5 mg, t_R 17 min), and fractions 63−64 were

also combined and purified by HPLC (column A, MeOH−H_2O, 70:30, 0.8 mL/min, UV 365 nm) to give **2** (3.8 mg, t_R 15 min). Part of C (80 mg) was subjected to a low-pressure ODS column (10 g), washing with MeOH−H_2O (70:30, 200 mL) to give five fractions. Fraction 5 contained sitosteryl-β-D-glucoside (14.6 mg), while fraction 1 was purified by HPLC (column A, MeOH−H_2O, 70:30, 1 mL/min, UV 365 nm) to give **3** (1.6 mg, t_R 13 min). The BuOH fraction was applied onto a Diaion HP-20 column, washing with H_2O and then MeOH to furnish a H_2O fraction and a MeOH fraction (600 mg). Part of the MeOH fraction (500 mg) was chromatographed over Si gel (100 g), eluting with $CHCl_3$−MeOH−H_2O (4:1:0.1 and 3:2:0.2, each 1000 mL), to furnish 32 fractions. Fractions 4−7 were combined and separated using a low-pressure ODS column (10 g) with mixtures of MeOH−H_2O (30:70, 50:50, and 100:0, each 100 mL) to furnish a 30% MeOH fraction (10 mg) and a 50% MeOH fraction (20 mg). Fractions were further purified by HPLC to give kelampayoside A (2.5 mg, column B, MeOH−H_2O, 50:50, 3 mL/min, UV 236 nm, t_R 19 min) and **7** (6 mg, column B, MeOH−H_2O, 60:40, 3 mL/min, UV 245 nm, t_R 22 min), respectively.

Naucleafine A (1): yellowish amorphous solid; mp 267−268 °C (MeOH); UV (MeOH) λ_{max} (log ϵ) 206 (3.28), 288 (2.67), 424 (3.01) nm; IR (KBr) ν_{max} 3451, 1642, 1071, 703, 627 cm^{-1}; ^1H and ^{13}C NMR data (Tables 1 and 2); ^{15}N NMR (DMSO-d_6) δ 124.1 (N-1), 164.2 (N-4); HRESIMS *m/z* 331.1065 $[M + H]^+$ (calcd for $C_{20}H_{15}N_2O_3$, 331.1042); R_f 0.70 and 0.80 (Si gel, solvents A and B, respectively), 0.29 (reversed-phase KC_{18} F, solvent C).

Naucleafine B (2): yellowish amorphous solid; mp 222−223 °C (MeOH); $[\alpha]^{22}_D$ −11.4° (*c* 0.07, MeOH); UV (MeOH) λ_{max} (log ϵ) 214 (4.98), 290 (3.19), 374 (3.62), 394 (3.61) nm; IR (KBr) ν_{max} 3395, 2924, 1667, 1604, 1274, 1070, 879, 828, 746 cm^{-1}; ^1H and ^{13}C NMR data (Tables 1 and 2); ^{15}N NMR (DMSO-d_6) δ 123.0 (N-1), 151.7 (N-4); ESIMS *m/z* 297 $[M + H]^+$ and 295 $[M - H]^+$; R_f 0.70 and 0.80 (Si gel, solvents A and B, respectively), 0.29 (reversed-phase KC_{18} F, solvent C).

Naucleoside A (3): orange-yellow amorphous solid; mp 171−172 °C (MeOH); $[\alpha]^{22}_D$ − 48.6° (*c* 0.15, MeOH); UV (MeOH) λ_{max} (log ϵ) 216 (4.02), 260 (3.57), 372 (3.92), 390 (3.89) nm; IR (KBr) ν_{max} 3401, 2922, 1645, 1067, 885, 829 cm^{-1}; ^1H and ^{13}C NMR data (Tables 1 and 2); HRESIMS *m/z* 513.1837 $[M + H]^+$ (calcd for $C_{26}H_{29}N_2O_9$, 513.1867); R_f 0.47 and 0.27 (Si gel, solvents A and B, respectively), 0.42 (reversed-phase KC_{18} F, solvent C).

Naucleoside B (4): orange-yellow amorphous solid; mp 189−190 °C (MeOH); $[\alpha]^{22}_D$ −58.2° (*c* 0.15, MeOH); UV (MeOH) λ_{max} (log ϵ) 212 (3.68), 260 (3.20), 372 (3.55), 390 (3.52) nm; IR (KBr) ν_{max} 3417, 2923, 1645, 1276, 1068, 885, 829, 688 cm^{-1}; CD (*c* 0.4 × 10^{-3} M, MeOH): $[\theta]_{270}$ +15813 (max); ^1H and ^{13}C NMR data (Tables 1 and 2); HRESIMS *m/z* 513.1837 $[M + H]^+$ (calcd for $C_{26}H_{29}N_2O_9$, 513.1867); R_f 0.22 and 0.20 (silica gel, solvents A and B, respectively), 0.48 (reversed-phase KC_{18} F, solvent C).

Strictosamide (5): yellowish amorphous solid; mp 176−177 °C (MeOH); $[\alpha]^{22}_D$ −56.3° (*c* 0.15, MeOH); CD (*c* 0.4 × 10^{-3} M, MeOH) $[\theta]_{258}$ +39604 (max), $[\theta]_{270}$ +69350 (max), $[\theta]_{295}$ +18897 (max); R_f 0.20 and 0.15 (Si gel, solvents A and B, respectively), 0.54 (reversed-phase KC_{18} F, solvent C). The UV, IR, and ^1H and ^{13}C NMR data (recorded in MeOH-d_4) were identical with the reported values.[6]

Vincosamide (6): yellowish amorphous solid; mp 194−196 °C (MeOH) [lit.[17] 201−202 °C (MeOH); $[\alpha]^{22}_D$ −48.6° (*c* 0.15, MeOH); CD (*c* 0.4 × 10^{-3} M, MeOH) $[\theta]_{261}$ −34830 (max); $[\theta]_{272}$ −29425 (max); R_f 0.30 and 0.26 (Si gel, solvents A and B, respectively), 0.50 (reversed-phase KC_{18} F, solvent C). The UV, IR, and ^1H and ^{13}C NMR data (recorded in MeOH-d_4) were identical with the reported values.[6]

Pumiloside (7): colorless amorphous solid; mp 307−308 °C (MeOH) [lit.[11] >300 °C]; $[\alpha]^{22}_D$ −39.8° (*c* 0.15, MeOH); UV (MeOH) λ_{max} (log ϵ) 214 (3.89), 244 (4.09), 303 sh, 316 (3.31), 328 (3.58) nm; IR (KBr) ν_{max} 3412, 2924, 1648, 1631, 1583, 1470, 1069, 897, 828, 767 cm^{-1}; CD (*c* 0.15 × 10^{-3} M, MeOH) $[\theta]_{264}$ −50943 (max), $[\theta]_{312}$ + 40485 (max), $[\theta]_{327}$ + 45670 (max); ^1H and ^{13}C NMR data (Tables 1 and 2); HRESIMS *m/z* 513.1826 $[M + H]^+$ (calcd for $C_{26}H_{29}N_2O_9$, 513.1867); R_f 0.08 (Si gel, solvent A), 0.67 (reversed-phase KC_{18} F, solvent C).

Acid Hydrolysis of Compounds 3 and 4. Compounds **3**, **4**, and the authentic sugar D-glucose were spotted on a silica gel TLC plate and hydrolyzed in situ by exposure to HCl vapor at 70 °C for 20 min. The TLC plate was then developed with $CHCl_3$−MeOH−AcOH−H_2O (14:6:2:1) and sprayed with 10% H_2SO_4 for detection. Glucose was detected with an R_f value of 0.20 from **3** and **4**.

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