Heronamides D-F, Polyketide Macrolactams from the Deep-Sea-Derived Streptomyces sp SCSIO 03032

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Heronamides D−F, Polyketide Macrolactams from the Deep-Sea-Derived *Streptomyces* sp. SCSIO 03032

Wenjun Zhang,[†] Sumei Li,[†] Yiguang Zhu,[†] Yuchan Chen,[‡] Yaolong Chen,[†] Haibo Zhang,[†] Guangtao Zhang,[†] Xinpeng Tian,[†] Yu Pan,[§] Si Zhang,[†] Weimin Zhang,[‡] and Changsheng Zhang*[,†]

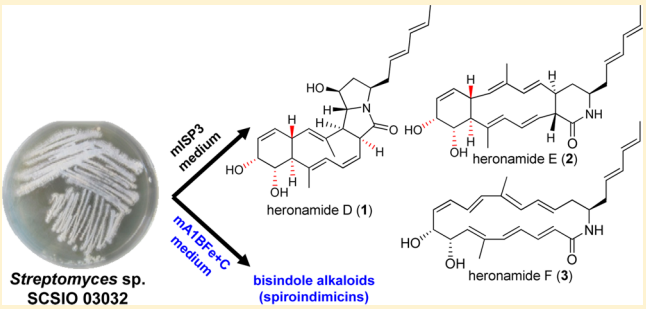
[†]Key Laboratory of Tropical Marine Bio-resources and Ecology, RNAM Center for Marine Microbiology, Guangdong Key Laboratory of Marine Materia Medica, South China Sea Institute of Oceanology, Chinese Academy of Sciences, 164 West Xingang Road, Guangzhou 510301, People's Republic of China

[‡]Guangdong Institute of Microbiology, 100 Central Xianlie Road, Guangzhou 510070, People's Republic of China

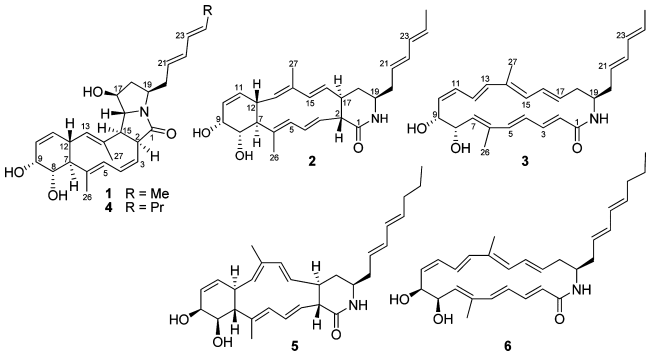
[§]Medical Research Center, Guangdong General Hospital, Guangdong Academy of Medical Sciences, 96 Dongchuan Road, Weilun Bldg., Guangzhou 510080, People's Republic of China

Supporting Information

ABSTRACT: Three new macrolactams, heronamides D−F (**1**−**3**), were isolated from the deep-sea-derived *Streptomyces* sp. SCSIO 03032 upon changing cultivation conditions. The planar structures of heronamides D−F (**1**−**3**) were elucidated by extensive MS and NMR spectroscopic analyses and comparisons with the closely related heronamides A−C. The relative configurations of **1**−**3** were deduced by detailed analysis of $^{3}J_{HH}$ values and NOESY data. The absolute configurations of **1** and **2** were determined by chemical modifications and application of the modified Mosher's method. None of the compounds exhibited obvious antimicrobial or cytotoxic activities.



The deep sea has long been thought of as a biological desert, owing to extreme variations in pressure, salinity, and temperature.[1] The recent advancements in marine technologies have allowed humans to access the deep sea, the earth's largest and least-explored ecosystem,[2] and revealed the presence of bacteria in this otherworldly ecosystem by isolating actinomycetes from sediments collected at a depth of 10 898 m in the Mariana Trench[3] and by *in situ* measurement of microbial carbon turnover rates in sediments at Challenger Deep in the Mariana Trench.[4] Recently, we reported unusual *spiro*-containing bisindole alkaloids, spiroindimicins A−D, and α-pyridone antibiotic piericidin E1 from the actinomycete species *Streptomyces* sp. SCSIO 03032 that was isolated from a sediment sample at the depth of 3412 m from the Bay of Bengal in the Indian Ocean.[5] Genome sequencing of numerous actinomycetes has revealed their genetic potential to encode multiple biosynthetic gene clusters, most of which have not been linked to chemically defined natural products.[6] Genome mining,[7] transcriptome mining,[8] proteomics,[9] and a combination of these strategies[10] have been successful in connecting some silent gene clusters to new metabolites. However, we should not ignore the "one strain−many compounds" approach, for its ease and usefulness in discovering new natural products, simply by altering cultivation parameters under laboratory conditions.[11] To this end, the cultivation of *Streptomyces* sp. SCSIO 03032 in an alternative medium led to the isolation of three new macrolactams of polyketide origin, designated heronamides D−F (**1**−**3**), on the basis of their close structural similarity to heronamides A−C (**4**−**6**).[12] Herein we report the isolation, structure elucidation, and biological activities of these three new macrocyclic lactams.



We have previously shown that the strain *Streptomyces* sp. SCSIO 03032 was capable of producing four new alkaloids, spiroindimicins A−D, along with the two known compounds lynamicins A and D, and a new α-pyridone antibiotic, piericidin E1, when cultivated in the modified A1BFe+C medium.[5] To search for more compounds, we investigated metabolite profiles

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Table 1. NMR Spectroscopic Data (^{1}H 500 MHz; ^{13}C 125 MHz) for Heronamides D−F (1−3)

	1[a]		2[a]		3[b]	
no.	δ_C, type	δ_H, mult (J in Hz)	δ_C, type	δ_H, mult (J in Hz)	δ_C, type	δ_H, mult (J in Hz)
1	175.2, C		172.6, C		168.1, C	
2	54.3, CH	3.56, ddd (8.9, 6.7, 2.1)	55.4, CH	2.66, dd (10.5, 10.5)	129.8, CH	6.34, m[c]
3	124.9, CH	5.47, m[c]	129.8, CH	5.13, dd (15.0, 10.5)	141.4, CH	7.42, dd (15.0, 8.5)
4	132.4, CH	6.66, ddd (10.4, 9.7, 2.1)	134.6, CH	5.85, m[c]	125.5, CH	6.22, m[c]
5	130.8, CH	5.42, d (9.7)	134.1, CH	5.63, m[c]	143.9, CH	6.36, m[c]
6	130.6, C		132.9, C		132.5, C	
7	53.1, CH	1.99, dd (10.5, 10.5)	55.0, CH	2.31, dd (11.5, 10.5)	140.0, CH	5.87, d (8.0)
8	69.7, CH	3.87, dd (10.5, 4.2)	70.3, CH	3.85, dd (11.5, 4.0)	73.4, CH	5.33, dd (8.0, 2.8)
9	66.2, CH	4.23, dd (4.2, 4.1)	66.4, CH	4.25, m	71.4, CH	5.03, m[c]
10	127.0, CH	5.91, m[c]	127.1, CH	5.86, m[c]	132.4, CH	6.18, m[c]
11	133.4, CH	5.93, m[c]	134.2, CH	5.85, m[c]	124.4, CH	6.33, m[c]
12	42.4, CH	2.76, dd (10.8, 10.5)	42.0, CH	3.13, dd (10.5, 10.5)	124.8, CH	6.24, m[c]
13	131.7, CH	4.96, d (10.8)	132.8, CH	4.84, d (10.5)	137.4, CH	6.21, m[c]
14	136.9, C		136.4, C		133.7, C	
15	57.8, CH	3.08, dd (8.9, 8.9)	141.2, CH	5.66, m[c]	131.2, CH	6.11, brd (11.0)
16	67.1, CH	3.90, m[c]	125.8, CH	4.88, dd (15.0, 9.5)	131.3, CH	6.50, dd (15.0, 11.0)
17	75.6, CH	3.93, m[c]	46.5, CH	2.39, m	132.2, CH	5.90, m
18a	41.5, CH_2	1.80, m	34.7, CH_2	1.57, m	41.8, CH_2	2.53, m
18b		2.48, m		2.21, m		2.07, m
19	52.2, CH	3.95, m	52.5, CH	3.51, m	50.4, CH	4.58, m
20a	37.5, CH_2	2.39, m	40.4, CH_2	2.15, m	38.9, CH_2	2.44, m
20b		2.55, m		2.34, m		2.37, m
21	125.5, CH	5.47, m[c]	124.8, CH	5.44, ddd (15.0, 8.5, 6.5)	128.6, CH	5.75, ddd (15.0, 7.5, 7.5)
22	133.9, CH	6.10, dd (14.5, 10.5)	134.9, CH	6.12, dd (15.0, 10.5)	131.2, CH	6.19, m[c]
23	131.2, CH	6.08, ddd (14.5, 10.5, 1.5)	130.8, CH	6.02, ddd (15.0, 10.5, 1.5)	127.6, CH	6.02, ddd (15.0, 10.5, 1.5)
24	128.4, CH	5.65, d (14.3)	129.5, CH	5.65, m[c]	133.0, CH	5.59, dq (15.0, 6.5)
25	18.0, CH_3	1.74, d (7.3)	18.0, CH_3	1.75, d (6.5)	12.6, CH_3	1.63, d (6.5)
26	12.6, CH_3	1.71, s	12.9, CH_3	1.64, s	18.0, CH_3	1.78, s
27	16.6, CH_3	1.36, s	13.8, CH_3	1.61, s	12.5, CH_3	1.73, s
NH				5.94, s		7.85, s

[a]Measured in $CDCl_3$. [b]Measured in pyridine-d_5. [c]Overlapping signals.

of *Streptomyces* sp. SCSIO 03032 under cultivation in another five media, including AM1, AM2, AM3, AM4, and modified ISP3 media (Figure S1). HPLC analyses of the EtOAc extracts of these cultures revealed a distinct metabolite profile in the modified ISP3 medium from that in the modified A1BFe+C medium, in which indole alkaloids were major products. Subsequently, a 12 L fermentation of *Streptomyces* sp. SCSIO 03032 was performed in the modified ISP3 medium. Upon XAD-16 resin-assisted extraction and a variety of chromatographic steps, three new compounds were isolated and characterized to be macrolactams heronamides D−F (**1**−**3**).

Heronamide D (**1**) was found to have the molecular formula $C_{27}H_{35}NO_4$ by HRESIMS (Figure S2), indicating 11 degrees of unsaturation. The ^{1}H and ^{13}C NMR spectroscopic data of **1** (Table 1, Figure S2) were similar to those of heronamide A (**4**).[12] Detailed comparison of 1D and 2D NMR data revealed that **1** differed from **4** only by having a side chain two methylenes shorter attached at C-19. In heronamide D (**1**), a methyl group (δ_H 1.74 d, J = 7.3, H_3-25; δ_C 18.0 CH_3, C-25; Table 1) was linked to C-24, evident from the COSY correlation of H-25/H-24 and the HMBC correlations from H_3-25 to C-24/C-23 (Figure S2), while a propyl group was linked to C-24 in **4**.[12]

Recently, the configurations of **4** at C-2, C-7, C-8 C-9, and C-12 were reassigned.[13] The relative configuration of **1** was assigned the same as **4** upon a careful comparison of the ^{1}H−^{1}H coupling constants and NOESY correlations of **1** (Table S1, Figure S2) with those reported for **4**.[12,13] The *cis* orientation of H-2 and H-15 in **1** and **4** was indicated from the NOESY correlations from H-5 to H-2/H-7/H-13/H-15 (Table S1).[13] The H-2/H-15 *trans* orientation was originally proposed in **4** on the basis of a large coupling constant of $^3J_{H\text{-}2/H\text{-}15}$ (8.9 Hz).[12] However, caution has been recommended when determining the configuration of five-membered rings by a spin−spin coupling constant approach:[14] a large coupling constant can be present in both *cis* (e.g., $^3J_{H\text{-}2/H\text{-}6}$ 10.8 Hz in roussoellol A[15]) and *trans* (e.g., $^3J_{H\text{-}1/H\text{-}5}$ 10.8 Hz in boscartols A[16]) orientations. Although the ^{1}H NMR resonances for the three methines H-16, H-17, and H-19 overlapped for **1** in $CDCl_3$ (Figure S2), they were well separated in the *R*-Mosher ester (**1b**) of acetonide **1a** (Figures S3−S5). NOESY correlations (**1b**) from H-17 to H-15/H-18b and from H-18b to H-19 suggested that the protons H-15, H-18b, and H-19 were on the same side as H-17 (Table S2, Figure S5), whereas NOESY correlations (**1b**) from H_3-27 to H-12/H-16 supported the *cis* orientation of H-16 and H-12 (Table S2), thus clearly determining the *trans* orientation of H-15 and H-16 ($^3J_{H\text{-}15/H\text{-}16}$ 9.0 Hz) in **1b**. Finally, the absolute configuration of **1** was determined as 2*S*, 7*S*, 8*S*, 9*R*, 12*R*, 15*S*, 16*R*, 17*S*, and 19*R*, the same as that of **4**,[13] following the same approach of Mosher's esterification of **1** and its derivatives **1b**−**e** (Tables S3 and S4, Figures S6 and S7).

The molecular formula of heronamide E (**2**) was determined as $C_{27}H_{35}NO_3$ (11 degrees of unsaturation) on the basis of

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HRESIMS (Figure S8). The ^{1}H and ^{13}C NMR data of **2** (Table 1, Figure S8) were similar to those of heronamide B (**5**).[12] The only difference was that the propyl group at C-24 in **5** was replaced by a methyl group (δ_H 1.75 d, J = 6.5 Hz, H-25; δ_C 18.0 CH_3, C-25; Table 1) in **2**,[12] which was supported by the COSY correlation between H-24 and H-25 and HMBC correlations from H-25 to C-23/C-24 (Figure S8). The relative configuration of **2** was assigned by carefully analyzing $^{1}H-^{1}H$ coupling constants and NOESY correlations (Table S5, Figure S8). The *s-trans* Δ^{13}, Δ^{15} diene conformation in **2** was supported by NOESY correlations between H-16 and H_3-27 and from H-15 to H-13/H-17 (Table S5, Figure S8).[17] Analysis of the differences between the ^{1}H NMR chemical shifts of the 8-mono-MTPA derivatives **2a** and **2b** confirmed the 8*S* configuration of **2** (Table S6, Figure S9). Thus the absolute configuration of **2** was determined as 2*R*, 7*S*, 8*S*, 9*R*, 12*R*, 17*R*, and 19*R*, which differs from that of **5** (2*R*, 7*R*, 8*R*, 9*S*, 12*S*, 17*R*, and 19*R*).[12,18]

The molecular formula of heronamide F (**3**) was established as $C_{27}H_{35}NO_3$ (11 degrees of unsaturation) by HRESIMS (Figure S10). The difference of **3** from heronamide C (**6**)[12] was that the propyl group at C-24 in **6** was substituted by a methyl group (δ_H 1.75 d, J = 6.5 Hz, H-25; δ_C 18.0 CH, C-25) in **3** (Table 1, Figure S10). The *E* configurations of double bonds $\Delta^{16,17}$, $\Delta^{21,22}$, and $\Delta^{23,24}$ in **3** were assigned by large $^{3}J_{HH}$ values of 15.0 Hz (Table S7). However, the weak and overlapped NMR signals of **3** in pyridine-d_5 impeded further assignment of the relative configuration (Figure S11). By analogy to assigning the relative configuration of **6**,[12] the diacetate derivative **3a** was made to improve solubility, and the more resolved 1D and 2D NMR signals of **3a** in $CDCl_3$ (Figure S11) allowed for the assignment of the relative configuration of **3** by the analysis of coupling constants and NOESY correlations (Tables S7 and S8). Although a *cis*-8,9-diol functionality was suggested by a $J_{8,9}$ value of 2.8 Hz (Table S7), the flexibility of the macrolactam prevented a direct determination of the relative configuration. On the consideration of **3** as a biosynthetic precursor of **1** and **2**, similar to the biosynthetic proposal of **6** en route to **4** and **5**,[12] the absolute configuration of **3** was tentatively assigned as 8*S*, 9*R*, and 19*R*.

Heronamides D–F (**1**–**3**) and the diacetate derivative **3a** showed no antimicrobial activities, with MIC values above 128 μg/mL against four bacteria (*Escherichia coli* ATCC 25922, *Staphylococcus aureus* ATCC 29213, *Bacillus thuringensis* SCSIO BT01, *Bacillus subtilis* SCSIO BS01) and the fungus *Candida albicans* ATCC 10231. Compounds **1**–**3** exhibited no growth inhibitions to the cancer cell lines SF-268, MCF-7, and NCI-460, with IC_{50} values of above 100 μM. The IC_{50} values of compound **3a** against the cancer cell lines MCF-7, SF-268, and NCI-H460 were 15.4, 23.3, and 56.4 μM, respectively. Compounds **1**–**3** displayed no antioxidant activities compared with the reference antioxidant Trolox evaluated by DPPH radical scavenging activity assays.[19] No inhibitory effects of heronamides D (**1**) and F (**3**) on the migration of HUVEC cells were observed in scratch wound healing assays (Figure S12).[20]

EXPERIMENTAL SECTION

General Experimental Procedures. Optical rotations were measured on a 341 polarimeter (Perkin-Elmer, Inc.). UV spectra were recorded on a U-2900 spectrophotometer (Hitachi). CD spectra were measured with a Chirascan circular dichroism spectrometer (Applied Photophysics), and IR spectra were measured on a Nicolet*6700 FT-IR spectrometer (Thermo Scientific). NMR spectra were recorded on a Bruker AV-500 MHz NMR spectrometer (Bruker Biospin GmbH) with TMS as an internal standard. ESIMS data were measured with an LCQDECA XP HPLC/MS spectrometer. HRESIMS data were measured using a MaXis 4G UHR-TOFMS spectrometer (Bruker Daltonics Inc.).

Bacterial Material. The source of the strain SCSIO 03032 was previously described.[5]

Screening and Fermentation. The strain *Streptomyces* sp. SCSIO 03032 was grown and maintained on ISP4-agar containing 3% sea salt. A few loops of cells of strain SCSIO 03032 were inoculated into 50 mL of AM1, AM2, AM3, AM4, modified-ISP3, and modified A1BFe + C media, respectively, in 250 mL Erlenmeyer flasks. The cultivation was carried out on a rotary shaker (200 rpm) at 28 °C for 7 days. The fermentation broths were extracted with EtOAc and were monitored by HPLC-DAD (Figure S1). A total fermentation of 12 L of *Streptomyces* sp. SCSIO 03032 was performed by inoculating 40 mL of the seed culture into 2000 mL Erlenmeyer flasks (30 in total) containing 400 mL of modified ISP3 medium and was incubated on a rotary shaker (200 rpm) at 28 °C for 4 days. A 20 mL (5 vol %) amount of sterilized polystyrene resin (Amberlite XAD-16) was added into the production medium (400 mL) to absorb metabolites, and the fermentation was prolonged for another day.

Extraction and Isolation. The mycelia and polystyrene resin were separated from the culture media by filtration through a metal sieve (40 mesh). The mycelia were extracted three times, each with 2 L of acetone, and the acetone was removed under vacuum. The resins were washed twice with water followed by washing with 2 L of acetone. The acetone fractions were concentrated under vacuum to afford an aqueous residue, which was extracted four times with 1.5 L of EtOAc to yield a dry extract (7.0 g) under vacuum. The EtOAc extract was subjected to column chromatography over silica gel (300–400 mesh), eluting with a gradient of $CHCl_3$/MeOH (100:0 → 0:100) to give three fractions (Fr.1–Fr.4). Fr.4 (2.4 g) was purified by C_{18} reversed-phase MPLC (40 × 2.5 cm i.d.), eluting with a linear gradient of H_2O/MeOH (0–100%, 20 mL/min, 200 min) to give four fractions (Fr.4-1, Fr.4-2, Fr.4-3, Fr.4-4). Heronamide D (**1**, 19.2 mg) was obtained from fraction Fr.4-1 by preparative TLC. Heronamide E (**2**, 11.1 mg) was obtained from fraction Fr.4-2 by PTLC. Heronamide F (**3**, 21.0 mg) was obtained from Fr.4-3 by reversed-phase semipreparative HPLC.

*Heronamide D (**1**):* yellow powder; $[\alpha]_D^{20}$ −25.8 (*c* 0.23, MeOH); UV (MeOH) λ_{max} (log ε) 222 nm (3.88); ECD (*c* 8.8 × 10^{-5} M, MeOH) λ_{max} ($\Delta\varepsilon$) 259 (−1.09), 226 (−1.61) nm; IR (KBr) ν_{max} 3395, 2925, 1671, 1604 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) and ^{13}C NMR (125 MHz, $CDCl_3$) data, see Table 1; ESIMS *m/z* 460 $[M + Na]^+$ and 897 $[2\ M + Na]^+$; HRESIMS *m/z* 438.2610 $[M + H]^+$ (calcd for $C_{27}H_{36}NO_4$, 438.2639).

*Heronamide E (**2**):* yellow powder; $[\alpha]_D^{20}$ +66.1 (*c* 0.19, MeOH); UV (MeOH) λ_{max} (log ε) 229 nm (4.28); ECD (*c* 7.1 × 10^{-5} M, MeOH) λ_{max} ($\Delta\varepsilon$) 257 (−0.89), 227 (2.41) nm; IR (KBr) ν_{max} 3347, 2920, 1643, 1074 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) and ^{13}C NMR (125 MHz, $CDCl_3$) data, see Table 1; ESIMS *m/z* 444 $[M + Na]^+$, 865 $[2\ M + Na]^+$; HRESIMS *m/z* 422.2695 $[M + H]^+$ (calcd for $C_{27}H_{36}NO_3$, 422.2690).

*Heronamide F (**3**):* yellow powder; $[\alpha]_D^{20}$ +130 (*c* 0.06, pyridine); UV (MeOH) λ_{max} (log ε) 289 (3.99); 279 (3.91); 225 (3.67); 207 nm (3.62); ECD (*c* 1.3 × 10^{-4} M, MeOH) λ_{max} ($\Delta\varepsilon$) 320 (1.66), 226 (−0.63) nm; IR (KBr) ν_{max} 3413, 2922, 1647, 1348 cm^{-1}; ^{1}H NMR (500 MHz, pyridine-d_5) and ^{13}C NMR (125 MHz, pyridine-d_5) data, see Table 1; ESIMS *m/z* 422 $[M + H]^+$; HRESIMS *m/z* 422.2708 $[M + H]^+$ (calcd for $C_{27}H_{36}NO_3$, 422.2690).

*Heronamide D Acetonide (**1a**).* Compound **1** (2.5 mg) and pyridinium-*p*-toluenesulfonate (4.0 mg) were dissolved in 2,2-dimethoxypropane (1 mL) and MeOH (1 mL). The solution was stirred at room temperature (rt) for 1 h. The reaction was stopped with aqueous $NaHCO_3$ (5%, 1 mL) and extracted three times with CH_2Cl_2. The combined organic layers were reduced to dryness under vacuum, and the residue was purified by PTLC to yield the isopropylidene derivative **1a** as a yellow powder. **1a**: ESIMS *m/z* 478 $[M + H]^+$; ^{1}H NMR (Table S2, Figure S4).

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Mosher's MTPA Esters ***1b/1c***. A solution of **1a** (1.0 mg, 0.002 mmol), (*R*)-MTPACl (10 μL, 0.055 mmol), DCC (1.0 mg), and DMAP (1.0 mg) in pyridine (600 μL) was stirred at rt for 24 h to afford (*S*)-MTPA ester **1b**. A second reaction with (*S*)-MTPACl was performed similarly to yield (*R*)-MTPA ester **1c**. The products were purified by PTLC and analyzed by ESIMS (**1b** *m/z* 694 $[M + H]^+$; **1c** *m/z* 694 $[M + H]^+$) and ^1H NMR (Table S2, Figures S5 and S6).

Mosher's MTPA Esters ***1d/1e, 2a/2b***. Compound **1** or **2** (6 mg) was divided into two equal portions, and each was dissolved in 300 μL of pyridine in separate NMR tubes. To each, 1 mg of DMAP, DCC, and 10 μL of (*R*)-MTPA-Cl or 10 μL of (*S*)-MTPA-Cl were added. After 3 h, LC-MS or HPLC analysis indicated that equal amounts of mono-, di-, and tri-Mosher's or mono- and di-Mosher's ester products were formed. The reactions were terminated and the products were purified by PTLC. ESIMS: bis-9,17-(*S*)-MTPA ester **1d** *m/z* 870 $[M + H]^+$; bis-9,17-(*R*)-MTPA ester **1e** *m/z* 870 $[M + H]^+$; 8-(*S*)-MTPA ester **2a** *m/z* 638 $[M + H]^+$; 8-(*R*)-MTPA ester **2b** *m/z* 638 $[M + H]^+$. The ^1H NMR and COSY spectra for Mosher esters **1d**/**1e** (Figure S7) and **2a**/**2b** (Figure S9) were recorded.

*Heronamide F Diacetate (**3a**)*. A solution of **3** (10.0 mg) in pyridine (1 mL) and acetic anhydride (1 mL) was stirred overnight. The reaction was stopped, and the solution was reduced to dryness *in vacuo*. The residues were purified by PTLC to yield the **3** diacetate derivative (**3a**, 7.0 mg) as a yellow powder. **3a**: yellow powder; $[\alpha]_D^{20}$ +52 (*c* 0.4, MeOH); UV (MeOH) λ_{max} (log ε) 278 (3.68), 226 nm (3.85); ECD (*c* 9.8 × 10^{-5} M, MeOH) λ_{max} ($\Delta\varepsilon$) 320 (2.18), 277 (−0.88), 225 (−0.80) nm; IR (KBr) ν_{max} 3410, 2931, 1741, 1246 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) and ^{13}C NMR (125 MHz, $CDCl_3$) data, see Table S8; ESIMS *m/z* 506 $[M + H]^+$; HRESIMS *m/z* $[M + H]^+$ 506.2898 (calcd for $C_{31}H_{40}NO_5$, 506.2901).

Biological Assays. Antimicrobial activities were measured against the five indicator strains *Escherichia coli* ATCC 25922, *Staphylococcus aureus* ATCC 29213, *Bacillus thuringensis* SCSIO BT01, *Bacillus subtilis* SCSIO BS01, and *Candida albicans* ATCC10231 by the broth microdilution method. *In vitro* cytotoxic activities were evaluated against three tumor cell lines, MCF7 (human breast adenocarcinoma cell line), NCI-H460 (human non-small-cell lung cancer cell line), and SF268 (human glioma cell line) by the SRB assay according to a previously described protocol.[21] DPPH (1,1-diphenyl-2-picrylhydrazyl) radical scavenging activity was assessed according to Kang et al.[19] The effect of compounds **1** and **3** on migration potential of HUVEC cells was detected by the scratch wound healing assay.[20]

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

Supplementary methods, 1D and 2D NMR spectroscopic data of compounds **1**−**3**, **3a**, **1a**−**e**, **2a**, and **2b** are available free of charge via the Internet at http://pubs.acs.org.

■ AUTHOR INFORMATION

Corresponding Author

*Tel/Fax: +86 20 89023038. E-mail: czhang2006@gmail.com.

Notes

The authors declare no competing financial interest.

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