FULL PAPER

Pseudonocardides A – G, New γ-Butyrolactones from Marine-derived *Pseudonocardia* sp. YIM M13669

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Seven new γ-butyrolactones, named pseudonocardides A – G (**1** – **7**), were isolated from the marine-derived actinomycete strain *Pseudonocardia* sp. YIM M13669. Their structures were elucidated on the basis of spectroscopic data including 1D- and 2D-NMR, and HR-ESI-MS. The absolute configuration of **1** was determined by X-ray crystallographic analysis of **1a** (4-bromobenzoate derivative of **1**). The antibacterial activity against *Mycobacterium smegmatis* MC^2155 and cytotoxicities of compounds **1** – **7** were evaluated in this study.

Keywords: γ-Butyrolactones, Pseudonocardides A – G, *Pseudonocardia*, X-Ray crystallography, Antibacterial activity

Introduction

Pseudonocardia is the type genus of the bacteria family Pseudonocardiaceae, and it was first described by *Henssen* [1]. Although *Pseudonocardia* does not represent a major group within the phylum Actinobacteria, the ubiquitous distribution of *Pseudonocardia* strains has been reported from various environments including rhizospheric soil [2], ants-associated *Pseudonocardia* sp. [3], plant tissue [4], coastal sediment [5], and deep sea sediment [6]. This genus is also associated with the production of many bioactive secondary metabolites, for example, pseudonocardians A – C [6], phenazostatin D [7], pseudonocardones A – C, dentigerumycin [3][8], and the nystatin-like polyen NPP [9].

In continuation of our search for bioactive molecules, strain YIM M13669 exhibiting antibacterial activity against *Mycobacterium smegmatis* MC^2155 was selected for further study. The strain was isolated from a marine sediment sample collected from the South China Sea (2448 m depth). It is moderately halophilic (*i.e.*, requires 2.5% *w*/*v* NaCl in growth medium) and shows optimal growth at 28 °C and pH 8.0. The strain YIM M13669 was found to produce seven new γ-butyrolactone derivatives **1** – **7**, named as pseudonocardides A – G (*Fig. 1*).

This article describes the isolation, structure elucidation, and antimicrobial and cytotoxic activities of pseudonocardides A – G.

Results and Discussion

The molecular formula of pseudonocardide A (**1**) was deduced as $C_{14}H_{24}O_4$ from the HR-ESI-MS data (*m/z* 535.3236 ($[2M + Na]^+$)), indicating three degrees of unsaturation. The IR spectrum of **1** showed absorption bands due to OH (3345 cm^{-1}) and carbonyl group of γ-lactone (1748 cm^{-1}) [10]. The ^{1}H-NMR data of **1** displayed the presence of two olefinic H-atoms at δ(H) 5.74 (H–C(8)) and 5.50 (H–C(7)), three O-bearing CH (δ(H) 4.61 (H–C(4)), 3.87 (H–C(5)), and 4.58 (H–C(6))), and one Me group at δ(H) 0.89 (*t*, *J* = 6.8 Hz) (*Table 1*).

The ^{13}C-NMR and DEPT data (*Table 1*) exhibited resonances for 14 C-atoms: one lactone C=O (δ(C) 176.9), one 1,2-disubstituted C=C bond (δ(C) 135.9, 126.0), three O-bearing CH group (δ(C) 79.4, 73.6, and 67.5), seven CH_2 groups (δ(C) 31.0, 28.8, 28.3, 27.8, 27.3, 22.1, and 21.9), and one Me group (δ(C) 13.4). All protonated carbons were assigned by HMQC correlations. The key HMBCs (*Table 1*) of H–C(2) and H–C(3) with C(1) and C(4), and of H–C(4) with C(1) allowed the lactone cyclic ring to be defined, and the chemical shifts of the two CH_2 groups (CH_2(2), CH_2(3)) as well as that of H–C(4) agreed well with the values reported for γ-lactone derivatives [11][12]. The dihydroxy monounsaturated alkyl chain was determined by the HMBCs of H–C(6)/C(5), C(7) and C(8), CH_2(9)/C(7), C(8) and C(11), CH_2(10)/C(8), C(9), and Me(14)/C(12) and C(13). The key HMBCs of H–C(5)/C(3), C(4), and H–C(4)/C(6) established the

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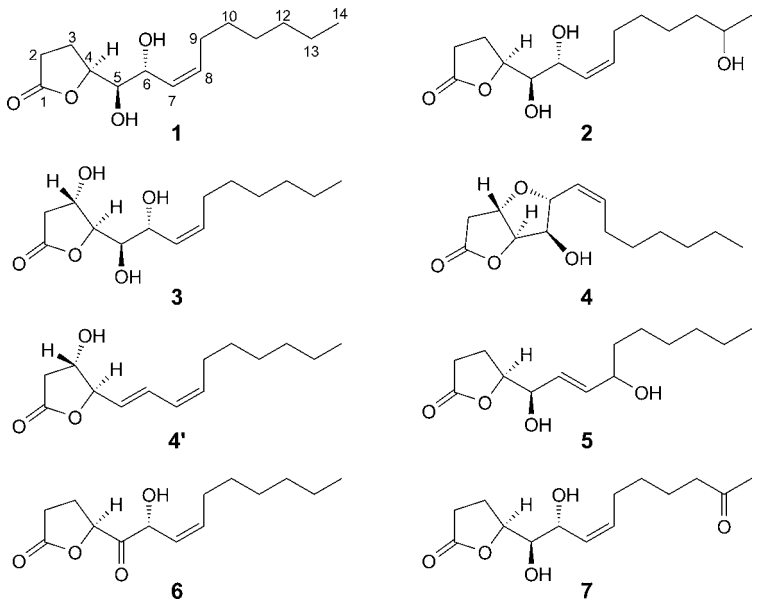

Fig. 1. The structures of compounds **1** – **7**

connectivity of C(4) and C(5), which confirmed that the lactone cyclic ring was substituted by the dihydroxy monounsaturated alkyl chain at the γ-locus. Thus, the planar structure of **1** was confirmed to be 5-[(3*Z*)-1,2-dihydroxydec-3-en-1-yl]dihydrofuran-2(3*H*)-one.

The 4-bromobenzoate derivative of **1** (**1a**) was obtained as colorless needle crystals by treatment of **1** with 4-bromobenzoyl chloride in the presence of pyridine and DMAP [13]. The absolute configuration of **1a** was determined as (4*S*,5*R*,6*R*) (*Fig. 2*) by X-ray diffraction analysis (CCDC No. 974435), which indicated that pseudonocardide A (**1**) possesses the same (4*S*,5*R*,6*R*) configuration.

The ^{1}H- and ^{13}C-NMR data of pseudonocardide B (**2**) (*Tables* 2 and *3*) were similar to those of **1**, except that the chemical shift of C(13) was downfield shifted to δ(C) 68.5 due to hydroxylation. The HMBCs of Me(14)/C(12) and C(13), and H–C(13) at δ(H) 3.70 further confirmed the existence of a OH group at C(13). Therefore, the planar structure of **2** was elucidated as 5-[(3*Z*)-1,2,9-trihydroxydec-3-en-1-yl]dihydrofuran-2(3*H*)-one.

Analysis of the HR-ESI-MS (*m/z* 295.1511 ([*M* + Na]$^+$)) of pseudonocardide C (**3**) established the molecular formula of $C_{14}H_{24}O_5$, corresponding to 16 amu more than that of **1**. The NMR data of **3** (*Tables 2* and *3*) revealed similar structural features to those of **1**, except

Table 1. NMR Data (600 MHz for ^{1}H- and 150 MHz for ^{13}C-NMR) for Pseudonocardide A (**1**) in $CDCl_3$. δ in ppm, *J* in Hz

Position	δ(H)	δ(C)	HMBC
1		176.9	
2α	2.53 (*dd*, *J* = 8.8, 17.8)	27.8	C(1), C(4)
2β	2.62 – 2.56 (*m*)		C(1), C(3), C(4)
3α	2.31 – 2.26 (*m*)	22.1	C(1), C(2), C(4), C(5)
3β	2.38 – 2.33 (*m*)		C(1), C(2), C(4), C(5)
4	4.61 (*q*, *J* = 6.9)	79.4	C(1), C(2), C(3), C(6)
5	3.87 (*t*, *J* = 5.2)	73.6	C(3), C(4), C(6), C(7)
6	4.58 (*dd*, *J* = 9.0, 5.3)	67.5	C(4), C(5), C(7), C(8)
7	5.50 (*t*, *J* = 10.1)	126.0	C(5), C(6), C(9)
8	5.74 (*dt*, *J* = 10.8, 7.5)	135.9	C(6), C(9), C(10)
9α	2.12 (*dd*, *J* = 6.9, 14.4)	27.3	C(7), C(8), C(11)
9β	2.19 (*q*, *J* = 7.2)		C(7), C(8), C(11)
10	1.38 – 1.35 (*m*)	28.3	C(8), C9)
11	1.25 – 1.32 (*m*, 3×CH_2)	28.8	C(12), C(13), C(14)
12		31.0	
13		21.9	
14	0.89 (*t*, *J* = 6.8)	13.4	C(12), C(13)

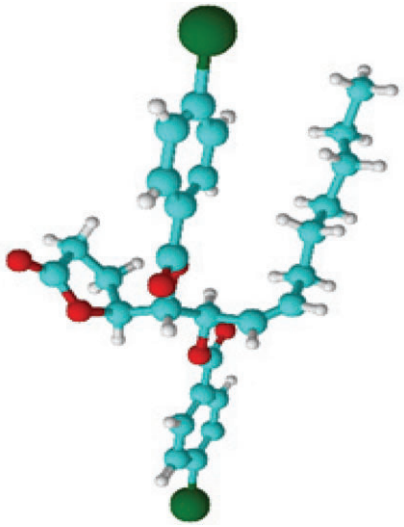

Fig. 2. X-Ray analysis of **1a**

Table 2. ^{1}H-NMR Spectral Data (600 MHz) for Pseudonocardides B – G (**2** – **7**) (δ in ppm, J in Hz)

Pos.	**2** (CD_3OD)	**3** ($CDCl_3$)	**4** ($CDCl_3$)	**4′** ($CDCl_3$)	**5** ($CDCl_3$)	**6** (CD_3OD)	**7** (CD_3OD)
2α	2.53 (*dt*, *J* = 17.3, 5.8)	2.67 (*dd*, *J* = 17.6, 8.1)	2.66 (*dd*, *J* = 17.5, 7.9)	2.55 (*dd*, *J* = 17.8, 3.9)	2.54 – 2.47 (*m*)	2.80 (*dd*, *J* = 18.9, 6.2)	2.57 – 2.5 (*m*)
2β	2.61 (*dt*, *J* = 17.3, 7.9)	2.91 (*dd*, *J* = 17.6, 7.8)	2.91 (*dd*, *J* = 17.6, 7.8)	2.86 (*dd*, *J* = 17.8, 6.6)	2.62 – 2.57 (*m*)	2.95 (*dd*, *J* = 18.9, 6.5)	
3α	2.27 – 2.24 (*m*)	4.75 (*q*, *J* = 6.7)	4.75 (*q*, *J* = 7.5)	4.37 (*f*, *J* = 2.7)	2.17 – 2.12 (*m*)	2.48 (*t*, *J* = 6.4)	2.30 – 2.25 (*m*)
3β	2.35 – 2.32 (*m*)				2.26 – 2.21 (*m*)	2.54 – 2.52 (*m*)	
4	4.69 (*q*, *J* = 6.7)	4.35 (br. *s*)	4.78 (*dd*, *J* = 8.3, 3.4)	4.87 (*dd*, *J* = 7.7, 2.6)	4.53 – 4.52 (*m*)	4.08 (*d*, *J* = 2.5)	4.69 (br. *s*)
5	3.73 (*t*, *J* = 5.6)	3.94 (*d*, *J* = 2.2)	3.93 (*t*, *J* = 6.0)	5.60 (*t*, *J* = 6.7)	4.51 – 4.49 (*m*)		3.71 (br. *s*)
6	4.39 (*t*, *J* = 6.8)	4.79 – 4.78 (*m*)	4.35 (*t*, *J* = 4.9)	6.67 (*dd*, *J* = 15.0, 10.3)	5.66 (*dd*, *J* = 15.5, 5.2)	4.58 (*dd*, *J* = 6.7, 4.8)	4.39 – 4.34 (*m*)
7	5.47 (*t*, *J* = 10.0)	5.46 (*t*, *J* = 10.0)	5.47 (*t*, *J* = 9.8)	5.99 (*t*, *J* = 11.0)	5.93 (*dt*, *J* = 15.5, 6.1)	5.46 (*t*, *J* = 10.5)	5.50 – 5.49 (*m*)
8	5.64 (*dt*, *J* = 10.8, 7.6)	5.71 (*q*, *J* = 8.4)	5.72 (*dt*, *J* = 10.8, 7.7)	5.58 (*dd*, *J* = 6.9, 11.3)	4.17 – 4.14 (*m*)	5.52 – 5.51 (*m*)	5.63 (br. *s*)
9α	2.16 (*q*, *J* = 6.7)	2.16 – 2.12 (*m*)	2.16 (*q*, *J* = 7.2)	2.20 (*q*, *J* = 7.1)	1.53 (br. *s*)	2.02 (*q*, *J* = 7.3)	2.18 – 2.15 (*m*)
9β		2.10 – 2.06 (*m*)	2.10 (*q*, *J* = 7.1)			2.08 (*q*, *J* = 7.3)	
10	1.33 – 1.45 (*m*) (3×CH_2)	1.39 – 1.36 (*m*)	1.39 – 1.36 (*m*)	1.38 – 1.35 (*m*)	1.29 – 1.37 (*m*) (4×CH_2)	1.36 – 1.29 (*m*)	1.59 – 1.58 (*m*)
11		1.24 – 1.30 (*m*) (3×CH_2)	1.25 – 1.32 (*m*) (3×CH_2)	1.25 – 1.32 (*m*) (3×CH_2)		1.25 – 1.31 (*m*) (3×CH_2)	1.41 – 1.39 (*m*)
12							2.50 – 2.49 (*m*)
13	3.70 – 3.69 (*m*)						
14	1.14 (*d*, *J* = 6.1)	0.89 (*t*, *J* = 6.8)	0.89 (*t*, *J* = 5.6)	0.88 (*t*, *J* = 6.5)	0.88 (*t*, *J* = 6.9)	0.88 (*t*, *J* = 6.8)	2.13 (*m*)

for the downfield shift of H–C(3) (δ(H) 4.75 in **3**; δ(H) 2.38 in **1**) and C(3) (δ(C) 67.0 in **3**; δ(C) 22.1 in **1**), which indicated that C(3) was hydroxylated. Thus, **3** was a derivative of **1** with a OH group at C(3) and the structure was shown to be 5-[(3*Z*)-1,2-dihydroxydec-3-en-1-yl]-4-hydroxydihydrofuran-2(3*H*)-one.

Pseudonocardide D (**4**) was obtained as colorless oil. The ^{1}H-NMR spectrum of **4** indicated a pure compound. However, further 1D- and 2D-NMR experiments revealed the formation of another compound **4′** during the measurements. Comparison of the NMR data of **4′** with those of **3** (*Tables 2* and *3*) showed that they share similar structures, except that C(5) and C(6) hydroxylated CH carbons of **3** were replaced by a (*E*) C=C bond in **4′**. Therefore, **4′** was elucidated as 5-[(1*E*,3*Z*)-deca-1,3-dien-1-yl]-4-hydroxydihydrofuran-2(3*H*)-one. The structure of **4** was deduced as in *Fig. 1* according to its ^{1}H-NMR data.

Pseudonocardide E (**5**) has the same molecular formula $C_{14}H_{24}O_4$ as that of **1** according to HR-ESI-MS. The NMR data of **5** (*Tables 2* and *3*) revealed the presence of a γ-lactone framework similar to that of **1**, which was substituted with a C_{10} paraffin chain including two OH groups. The key HMBCs of H–C(5)/C(3), C(4), C(6), and C(7), H–C(8)/C(6), C(7), C(9), and C(10) suggested that the C=C bond is located between the two hydroxylated CH groups. Therefore, the structure of **5** was assigned as 5-[(3*E*)-1,4-dihydroxydec-2-en-1-yl]dihydrofuran-2(3*H*)-one.

The ^{1}H- and ^{13}C-NMR data of pseudonocardide F (**6**) showed similar pattern as **1** except for a ketone signal at δ(C) 212.3 (C(5)). The HMBCs of CH_2(2), CH_2(3), and H–C(4) with C(5) further confirm the ketone group at C(5). Thus, **6** was a derivative of **1** with C(5)=O group and therefore identified as 5-[(3*Z*)-2-hydroxydec-3-enoyl]-dihydrofuran-2(3*H*)-one.

Comparison of the NMR data of **7** (*Tables 2* and *3*) with those of **2** showed the presence of a γ-butyrolactone derivative substituted with a C_{10} paraffin chain having one C=O group, two OH groups, and a (*Z*)-configured C=C bond. Furthermore, the HMBCs of Me(14)/C(12), and C(13), combined with the downfield chemical shifts of CH_2(12) and Me(14) revealed the position of the C=O group was at C(13). Thus, **7** was a derivative of **1** with C(13)=O group and was elucidated as 5-[(3*Z*)-1,2-dihydroxy-9-oxodec-3-en-1-yl]dihydrofuran-2(3*H*)-one.

The configurations of **2** – **7** were established through analysis of 1D- and 2D-NMR data and comparison with those of **1**. The coupling constants ($J \approx 10.0$ Hz) between H–C(7) and H–C(8) led to assignment of an (*Z*) configuration of C(7)=C(8) bond of compounds **1** – **4** and **6** – **7**. The (*E*) configuration of the C(6)=C(7) bond of **5** was deduced by the large coupling constants (J = 15.5 Hz). The relative configurations at C(4), C(5), and C(6) for compounds **2** – **7** were tentatively assigned as (4*S*), (5*R*), and (6*R*) by comparison with the configuration of **1a** which was determined by single crystal X-ray diffraction and on the basis of the obvious biogenetic relationship. In

Table 3. ^{13}C-NMR Spectral Data (150 MHz) for Pseudonocardides B – G (**2** – **7**)

Position	**2** (CD_3OD)	**3** ($CDCl_3$)	**4′** ($CDCl_3$)	**5** ($CDCl_3$)	**6** (CD_3OD)	**7** (CD_3OD)
1	180.5	174.6	174.8	177.8	176.5	180.7
2	29.4	37.1	36.9	28.7	35.8	29.6
3	23.1	67.0	72.3	21.2	28.3	23.3
4	82.3	85.7	87.3	82.1	81.6	82.4
5	75.9	72.8	126.1	72.2	212.3	76.1
6	68.7	69.5	129.1	126.3	69.8	68.9
7	129.9	126.0	126.6	137.1	128.5	130.3
8	135.1	136.3	135.8	72.0	135.0	134.9
9	28.9	28.2	27.9	37.3	28.9	28.7
10	26.6	29.6	29.4	25.5	30.1	24.6
11	30.8	29.1	28.9	29.3	30.7	30.2
12	40.0	31.8	31.7	31.9	32.9	44.2
13	68.5	22.7	22.6	22.7	23.7	212.3
14	23.5	14.2	14.1	14.3	14.4	30.0

addition, the configuration at C(3) for compounds **3**, **4**, and **4′** was determined as (3*S*) by the NOE correlations of H–C(3)/H–C(5) and H–C(3)/H_β–C(2) in **3** and **4′**, and the formation of the tetrahydrofuran ring in **4**, which was further supported by the coupling constants, 3.4 Hz and 2.6 Hz, between H_β–C(3) and H–C(4) in **4** and **4′**, respectively.

The antimicrobial activity of compounds **1** – **7** against *M. smegmatis* MC^2155 was investigated by paper disc diffusion assay. But none of them showed inhibitory activity at 30 μg/disc, indicating that other compounds should be responsible for activity and worthy of mining in further study.

The tumor cell lines MDA-MB-231 and A549 were used to evaluate the cytotoxic activities of all isolates. Compounds **1** – **7** were all inactive against the two cell lines at the concentration of 50 μM.

The isolation of γ-butyrolactones is mainly reported from *Streptomyces* spp. [14 – 16] and plants [17 – 19]. γ-Butyrolactones are reported as chemical signaling molecules or microbial hormones that regulate secondary metabolism and/or morphological differentiation in Actinobacteria especially in *Streptomyces* [20 – 22]. The present study is the first report of isolation of γ-butyrolactones from a marine *Pseudonocardia* sp.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

NMR spectra of compounds **1** – **7**.

Experimental Part

General

TLC: Precoated silica gel GF_{254} plates (*Qingdao Marine Chemical Factory*, Qingdao, P. R. China). Column chromatography (CC): SiO_2 (200 – 300 and 80 – 100 mesh; *Qingdao Marine Chemical Factory*), SiO_2 GF_{254} (*Merck*, Darmstadt, Germany), *RP-18* gel (40 – 63 μm; *Merck*), and *Sephadex LH-20* (25 – 100 μm; *Pharmacia Biotek*, California, USA). Optical rotations: *GYROMAT-HP* Polarimeter with MeOH as solvent. UV Spectra: *UV-2450* spectrophotometer (*Shimadzu*, Kyoto, Japan, in MeOH). IR: *Thermo Nicolet NEXUS 470 FT-IR* (*Thermo Fisher Scientific*, Waltham, MA, USA) spectrophotometer in KBr pellets; $\tilde{\nu}$ *in* cm^{-1}. ^{1}H- and ^{13}C-NMR Spectra: *Bruker DRX 600* spectrometer (*Bruker*, Karlsruhe, Germany), at 600 and 150 MHz, resp., δ in ppm rel. to Me_4Si, *J* in Hz. HR-ESI-MS: *LTQ-Orbitrap XL* (*Thermo Fisher Scientific*, Waltham, Massachusetts, USA); in *m/z* (rel. %). X-ray diffraction intensity data: *a MAC DIP-2030K* diffractometer using graphite-monochromater MoK_α radiation (λ = 0.71073 Å) by the ω-scan technique (scan width 0 – 180°, $2\theta \leq 50°$).

Isolation, Fermentation and Extraction

The strain YIM M13669 was isolated from a marine sediment sample from the South China Sea, and identified by 16S rRNA sequence analysis as *Pseudonocardia* sp. A BLAST query of the Ezbiocloud database yielded *Pseudonocardia antimicrobica* YIM 63235^T as the closest match to the 16S rRNA of YIM M13669 (99%). The protocols for genomic DNA extraction, PCR amplification, and sequencing of the gene were carried out by *Li et al.* [23]. Seed culture was prepared on YMG agar media. The seed culture was then inoculated onto *Petri* dishes containing MP agar (mannitol 20 g, peptone 20 g, sea salt 25 g, agar 15 g, tap water 1 l, pH 8.0; 20 ml/dish) as the antibiotic producing medium. The fermentation was

carried out at 28 °C for 14 days in darkness. The fermented media was extracted using AcOEt/MeOH/AcOH (80:15:5); the organic solvent was evaporated under vacuum to afford the crude extract. Then, the extract was partitioned between H_2O and AcOEt to give the AcOEt-soluble fraction. The AcOEt-soluble fraction was dissolved in 80 ml of 95% MeOH and extracted with an equal volume of petroleum ether (PE) to afford a MeOH-soluble fraction (4.0 g) and a PE-soluble fraction (0.3 g), respectively.

Isolation and Purification of Pseudonocardides A – G (**1** – **7**)

The MeOH-soluble fraction was chromatographed on MPLC over RP-18 (120 g, MeOH/H_2O 30:70, 50:50, 70:30, 100:0, *v*/*v*) to produce Fr.1 – 7. Fr.6 (195 mg) was purified by column chromatography (CC) (SiO_2, $CHCl_3$/MeOH 400:1, 350:1) to afford **1** (125 mg). Fr.3 (260 mg) was subjected to *Sephadex LH-20* (120 g in MeOH) to give Fr.3.1 – Fr.3.3. Fr.3.1 (48.2 mg) was subjected to MPLC (RP-18, MeOH/H_2O 15:85, 20:80, 25:75, 30:70, *v*/*v*) to produce Fr.3.1.1 – Fr. 3.1.3. Fr.3.1.1 (28 mg) was further purified by CC (SiO_2, $CHCl_3$/MeOH 80:1) to afford **2** (3 mg). Fr.5 (353 mg) was successively subjected to *Sephadex LH-20* (160 g in MeOH) to give Fr.5.1 – 5.4. Fr.5.1 (132 mg) was subjected to MPLC (RP-18, 30 g, MeOH/H_2O 30:70, 35:65, 100:0) to produce **1** (63 mg) and Fr.5.1.1 (23 mg). Fr.5.1.1 was purified by CC (SiO_2, $CHCl_3$/MeOH 400:1, 350:1) to afford **5** (17.2 mg). Fr.5.3 (14 mg) was subjected to *Sephadex LH-20* (80 g in MeOH) and followed by CC (SiO_2, $CHCl_3$/MeOH 350:1, 300:1, with 0.8% formic acid) to afford **3** (2 mg) and **6** (2.5 mg). Fr.5.4 (30 mg) was subjected to CC (SiO_2, $CHCl_3$/MeOH 300:1 with 0.5% formic acid) to afford **6** (14.5 mg). Fr.2 (1.0 g) was subjected to *Sephadex LH-20* (120 g in acetone) to give Fr.2.1 – Fr.2.4. Fr.2.1 (65 mg) was purified by MPLC (RP-18, 30 g) and CC (SiO_2; $CHCl_3$/MeOH 100:1, 80:1) to afford **7** (5 mg). Fr.7 (137 mg) was subjected to *Sephadex LH-20* (80 g in acetone) to give Fr.7.1 – Fr.7.3. Fr.7.2 (16 mg) was subjected to CC (SiO_2, PE/acetone 30:1) to yield **4** (10 mg).

4-Bromobenzoyl Derivation of Pseudonocardide A (**1**) *to Yield* **1a**

To a solution of **1** (10 mg, 39 μM) in pyridine (1 ml), 4-bromobenzoyl chloride (100 mg, 455 μM), and 4-(dimethylamino)pyridine (DMAP) (30 mg, 245 μM) were added at room temperature. The mixture was stirred overnight, then diluted with ice water (50 ml) and extracted with AcOEt (50 ml). The AcOEt phase was then washed with saturated NaCl solution (50 ml), dried with anhydrous Na_2SO_4, and concentrated under reduced pressure to give a crude residue (30 mg). Purification by *Sephadex LH-20* chromatography (80 g, MeOH) yielded compound **1a** as colorless oil. A portion of **1a** was recrystallized from a mixture of CH_2Cl_2 and MeOH to afford needle crystals suitable for X-ray crystallographic analysis.

Pseudonocardide A (= **(5*S*)-5-[(1*R*,2*R*,3*Z*)-1,2-Dihydroxydec-3-en-1-yl]dihydrofuran-2(3*H*)-one**; **1**). Colorless Oil. $[\alpha]_D^{20} = -27.8$ (*c* = 1.0, MeOH). IR (KBr): 3345, 1748. ^{1}H- and ^{13}C-NMR ($CDCl_3$): *Table 1*. HR-ESI-MS: 535.3236 ($[2M + Na]^+$, $C_{28}H_{48}NaO_8^+$; calc. 535.3247).

Pseudonocardide B (= **(5*S*)-Dihydro-5-[(1*R*,2*R*,3*Z*)-1,2,9-trihydroxydec-3-en-1-yl]furan-2(3*H*)-one**; **2**). White Powder. $[\alpha]_D^{20} = -20.0$ (*c* = 1.0, MeOH). ^{1}H- and ^{13}C-NMR (CD_3OD): *Tables 2* and *3*. HR-ESI-MS: 295.1509 ($[M + Na]^+$, $C_{14}H_{24}NaO_5^+$; calc. 295.1521).

Pseudonocardide C (= **(4*S*,5*S*)-5-[(1*R*,2*R*,3*Z*)-1,2-Dihydroxydec-3-en-1-yl]dihydro-4-hydroxyfuran-2(3*H*)-one**; **3**). Colorless Oil. $[\alpha]_D^{20} = -16.7$ (*c* = 1.0, MeOH). ^{1}H- and ^{13}C-NMR ($CDCl_3$): *Tables 2* and *3*. HR-ESI-MS: 295.1511 ($[M + Na]^+$, $C_{14}H_{24}NaO_5^+$; calc. 295.1521).

Pseudonocardide D′ (= **(4*S*,5*R*)-5-[(1*E*,3*Z*)-Deca-1,3-dien-1-yl]dihydro-4-hydroxyfuran-2(3*H*)-one**; **4′**). Colorless Oil. ^{1}H- and ^{13}C-NMR ($CDCl_3$): *Tables 2* and *3*.

Pseudonocardide E (= **(5*S*)-5-[(1*R*,2*E*)-1,4-Dihydroxydec-2-en-1-yl]dihydrofuran-2(3*H*)-one**; **5**). Colorless Oil. $[\alpha]_D^{20} = -18.8$ (*c* = 1.0, MeOH). ^{1}H- and ^{13}C-NMR ($CDCl_3$): *Tables 2* and *3*. HR-ESI-MS: 279.1560 ($[M + Na]^+$, $C_{14}H_{24}NaO_4^+$; calc. 279.1572).

Pseudonocardide F (= **(5*S*)-5-[(2*R*,3*Z*)-2-Hydroxydec-3-enoyl]dihydrofuran-2(3*H*)-one**; **6**). White Powder. $[\alpha]_D^{20} = -23.5$ (*c* = 1.0, MeOH). ^{1}H- and ^{13}C-NMR (CD_3OD): *Tables 2* and *3*. HR-ESI-MS: 277.1416 ($[M + Na]^+$, $C_{12}H_{22}NaO_4^+$; calc. 277.1416).

Pseudonocardide G (= **(5*S*)-5-[(1*R*,2*R*,3*Z*)-1,2-Dihydroxy-9-oxodec-3-en-1-yl]dihydrofuran-2(3*H*)-one**; **7**). Colorless Oil. $[\alpha]_D^{20} = -24.0$ (*c* = 1.0, MeOH). ^{1}H- and ^{13}C-NMR (CD_3OD): *Tables 2* and *3*. HR-ESI-MS: 293.1355 ($[M + Na]^+$, $C_{14}H_{22}NaO_5^+$; calc. 293.1365).

Crystallographic Data for **1a**

$C_{28}H_{30}Br_2O_6$; M = 620.0; orthorhombic, a = 7.7693 (5), b = 9.6529 (6), c = 34.705 (3) Å, space group: $P2_12_12_1$, Z = 4, D_χ = 1.588 Mg/m^3, μ = 3.156 mm^{-1}, $F(000)$ = 1264; colorless needle crystals, dimension 0.4 × 0.17 × 0.12 mm^3. CCDC number 974435 contains the supplementary crystallographic data for this article. These data can be obtained free of charge *via* http://www.ccdc.cam.ac.uk/data_request/cif.131013.

Antibacterial Bioassays

Antibacterial activities of compounds **1** – **7** were tested by the paper disc diffusion method [24]. The bacterial strain *M. smegmatis* MC^{2}155 was grown on LB agar. Test compounds were absorbed onto individual paper disks (6 mm diameter) at 30 μg/disc and placed on the surface of the agar. The assay plates were incubated at 37 °C for 24 h and examined for the presence of a zone of inhibition.

Cytotoxicity Evaluation

The protein-binding sulforhodamine (SRB) and microculture tetrazolium (MTT) method were used for the cytotoxicity against MDA-MB-231 and A549 cell lines [25][26].

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