Full Papers

β-Carboline Alkaloids from a Korean Tunicate *Eudistoma* sp.

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Seven new β-carboline-based metabolites, designated as eudistomins Y_1-Y_7 (**1**−**7**), were isolated from a tunicate of the genus *Eudistoma* collected near Tong-Yeong City, South Sea, Korea. These new metabolites differ from previously isolated marine metabolites due to the presence of a benzoyl group attached to the β-carboline nucleus at C-1. Eudistomins Y_1-Y_7 were evaluated for their antibacterial activity, and eudistomin Y_6 exhibited moderate antibacterial activity against Gram-positive bacteria *Staphylococcus epidermis* and *Bacillus subtilis* without cytotoxicity in the MTT assay at 100 μM.

Marine tunicates belonging to the genus *Eudistoma* (order Aplousobranchia, family Polycitoridae) have been the subject of extensive chemical and biological investigations. A large number of alkaloids containing the β-carboline skeleton have been isolated from various *Eudistoma*species since Rinehart and co-workers first reported the structures of eudistomins A−Q.[1,2] Examples of this class include eudistomins R−T,[3] W, and X,[4] eudistomidins A−F,[5–7] eudistalbins A and B,[8] isoeudistomins,[9] and two trypargine derivatives.[10] Also, metabolites structurally related to the eudistomins were reported from other tunicate genera such as *Ritterella*, *Pseudodistoma*, and *Lissoclinum*.[4] Many of them exhibited interesting biological activity, such as antiviral,[2,11] antimicrobial,[2,12] cytotoxic,[6,8,13] and calmodulin antagonistic activity.[5] The biosynthesis of these β-carbolines is generally believed to involve the coupling of tryptophan with a second amino acid, as demonstrated by *in vivo* studies with *E. olivaceum*.[14]

As part of our interest in marine natural products with biological activity, we investigated the organic extract of a tunicate *Eudistoma* sp., which yielded an antibacterial EtOAc fraction. The EtOAc fraction was subjected to solvent partitioning and chromatographic separation, as described in the Extraction and Isolation section, to provide seven new eudistomin congeners designated as eudistomins Y_1-Y_7 (**1**–**7**). The structures of the compounds were elucidated on the basis of NMR spectroscopic and MS spectrometric data. Eudistomins Y_1-Y_7 (**1**–**7**) were further evaluated for their antibacterial activity, and some of them were found to exhibit moderate antibacterial activity against Gram-positive pathogenic bacteria. Herein, we report the isolation and structure elucidation of these new β-carboline metabolites, as well as their antibacterial activity.

Results and Discussion

Eudistomin Y_1 (**1**) had the molecular formula $C_{18}H_{12}O_2N_2$, as determined by high-resolution FABMS. The ^{1}H NMR spectrum of **1** was comparable to those of eudistomin T,[3,15] which points out the presence of both a β-carboline moiety and a phenyl ring. The four contiguous aromatic proton signals at δ 8.32 (br d, J = 7.88 Hz), 7.35 (t, J = 7.88 Hz), 7.64 (t, J = 7.88 Hz), and 7.89 (br d, J = 7.88 Hz), respectively, were indicative of a 1,2-disubstituted phenyl ring. The two mutually coupled doublet signals at δ 8.56 and 8.39, respectively, could be assigned to α and β pyridine protons, according to their chemical shifts, coupling constant (J = 4.93 Hz), and the chemical shifts of carbons to which they are attached (C-2, δ 119.2; C-3, δ 138.5). These spectroscopic characteristics suggested the presence of a C-1-substituted β-carboline, which was supported by HMBC correlations. In the HMBC spectrum, all possible $^3J_{CH}$ and some $^2J_{CH}$ were displayed, from the six protons to the 11 carbons in the β-carboline system. In particular, in DMSO-d_6, one could observe the HMBC correlations of the exchangeable indolic NH proton to the four quaternary sp^2 carbons of the indole ring. In addition to the above-mentioned proton signals, the ^{1}H NMR spectrum showed two two-proton doublets at δ 8.47 (d, J = 8.69 Hz) and 7.00 (d, J = 8.69 Hz), respectively, which come from two pairs of adjacent aromatic protons on a *para*-disubstituted phenyl ring (H-12/16 and H-13/15). The HMBC correlations of the protons H-13/15 to the carbons resonating at δ 130.2 and 162.7 allowed the assignment of the carbon C-11 and the oxygenated carbon (C-14).

Since the two separate moieties, the β-carboline and the phenyl ring, were fully defined by the NMR data, they must be linked between the C-1 and C-11 positions via the C-10 carbonyl group. The presence of the carbonyl group was indicated by the IR absorption at 1678 cm^{-1} and the ^{13}C NMR signal at δ 192.9. The intense MS fragment ion peak at m/z 121 (arising from the benzoyl moiety [HO−C_6H_4−CO−]$^+$) together with the clear HMBC cross-peaks from H-12/16 to the carbonyl carbon (δ 192.9) fixed the connection of C-10 to C-11. The assignment of the 1-benzoyl-β-carboline was further corroborated by comparison of the NMR data with those of 1-acyl-β-carbolines (e.g., xestomanzamine A[16] and hyrtiomanzamine[17]) and 1-aroyl-β-carbolines.[18,19] The latter are often produced as reaction intermediates during the synthesis of the biologically active natural product fascaplysin[19,20] and also are prepared as a reactant during the synthesis of yohimbanone.[18] Thus, eudistomin Y_1 (**1**) differs from eudistomin T by the presence of a benzoyl functionality at C-1 instead of a 2-phenylacetyl substituent. To the best of our knowledge, there has been only one naturally occurring 1-benzoyl-β-carboline reported, named alstonilidine, which was found from the root bark of *Alstonia constricta*.[21]

Eudistomin Y_2 (**2**) yielded an isotopic cluster of [M + H]$^+$ ion peaks at m/z 367/369 with intensities in a 1:1 ratio in the FAB mass spectrum, which is a characteristic of a monobrominated compound. The molecular formula of **2** was deduced as

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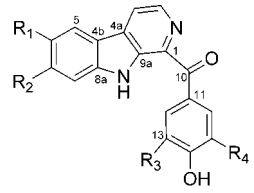

1	R_1=H	R_2=H	R_3=H	R_4=H
2	R_1=Br	R_2=H	R_3=H	R_4=H
3	R_1=H	R_2=H	R_3=Br	R_4=H
4	R_1=Br	R_2=H	R_3=Br	R_4=H
5	R_1=H	R_2=H	R_3=Br	R_4=Br
6	R_1=Br	R_2=H	R_3=Br	R_4=Br
7	R_1=H	R_2=Br	R_3=Br	R_4=Br

Figure 1. Structures of compounds **1**–**7**.

$C_{18}H_{11}O_2N_2Br$ on the basis of the HRFABMS data. The examination of the ^1H NMR spectrum revealed that it is comparable to that of eudistomin Y_1 (**1**). However, changes were observed in the A ring of the β-carboline nucleus. The spin system of four aromatic proton signals in **1** was replaced by a three-proton aromatic coupling pattern in which there was a downfield shift of H-5 (+ 0.21 ppm) and H-7 (+0.12 ppm) signals. In comparison with **1**, significant differences were also observed in the ^{13}C chemical shifts of C-5 (+2.8 ppm), C-6 (−7.8 ppm), and C-7 (+2.6 ppm). These spectroscopic features immediately restrict the substituent at C-6 to only a few possibilities such as bromine, cyano, or an ethynyl group. Careful comparison of its ^1H and ^{13}C NMR data with those of 6-brominated β-carbolines (e.g., eudistomins H[2] and S[3]), and other calculated values, established the presence of a bromine atom in **2**, which is consistent with the pseudomolecular ion peak at *m/z* 367/369 $[M + H]^+$ and the fragment ion peak at *m/z* 245/247 due to the loss of a benzoyl function. Thus, eudistomin Y_2 (**2**) is characterized as a 6-brominated congener of eudistomin Y_1 (**1**).

High-resolution FABMS of eudistomin Y_3 (**3**) provided the molecular formula $C_{18}H_{11}O_2N_2Br$. The ^1H NMR spectroscopic data of eudistomin Y_3 (**3**) are analogous to those of eudistomin Y_1 (**1**) and contain three spin systems as verified by COSY correlations. Two of them are in close agreement with the chemical shifts observed in eudistomin Y_1 (**1**), which established the presence of a C-1-substituted β-carboline nucleus. The third one, an ABX spin system, constituted a 1,2,4-trisubstituted benzene ring, as demonstrated by signals at δ 8.55 (d, J = 1.97 Hz), 7.03 (d, J = 8.10 Hz), and 8.21 (dd, J = 8.10, 1.97 Hz) for H-12, H-15, and H-16, respectively. The absence of the H-13 signal and the corresponding upfield shift of C-13 (−6.1 ppm) compared to that of **1** required that a bromine atom should be substituted at C-13. Therefore, the structure of eudistomin Y_3 (**3**) was assigned as a 13-brominated derivative of eudistomin Y_1 (**1**). The 3-bromo-4-hydroxybenzene ring is also encountered in other marine natural products such as psammaplins A_1[22] and K.[23]

Eudistomin Y_4 (**4**) showed a $[M + H]^+$ ion cluster at *m/z* 445/447/449 with intensities in a 1:2:1 ratio in the FABMS spectrum, suggesting the presence of two bromine atoms in the molecule. As in the case of eudistomin Y_2 (**2**), the MS fragment ion peaks at *m/z* 245/247 due to the loss of a benzoyl moiety were also observed, which implies the location of one bromine atom on the β-carboline unit and the other one attached on the benzoyl part. Inspection of the ^1H and ^{13}C NMR spectra of eudistomin Y_4 (**4**) revealed that it shares the same 6-brominated β-carboline nucleus with eudistomin Y_2 (**2**) and the same 3-bromo-4-hydroxybenzoyl function with eudistomin Y_3 (**3**). This structural assignment was compatible with the high-resolution FABMS data and is further corroborated by a combination of COSY, HSQC, and HMBC experiments.

Eudistomin Y_5 (**5**) again showed a $[M + H]^+$ ion cluster at *m/z* 445/447/449 with intensities in a 1:2:1 ratio in the FABMS spectrum, which is ascribed to the occurrence of a dibrominated substitution pattern in the molecule as in eudistomin Y_4 (**4**). However, the MS fragment ion peak at *m/z* 121 attributed to a nonbrominated β-carboline unit, different from that of **4**, was observed, indicating both of the bromine atoms were present in the benzoyl moiety. A singlet at δ 8.55 integrating for two protons was observed in the ^1H NMR spectrum, reminiscent of a symmetrically tetrasubstituted phenyl ring. The HMBC correlations from this proton signal to C-13/15 at δ 111.0, C-14 at δ 154.8, and the carbonyl carbon at δ 188.8 (C-10) proved the presence of a 3,5-dibromo-4-hydroxy benzoyl ring. Thus, the structure of eudistomin Y_5 (**5**) is established as a 13,15-dibrominated congener of eudistomin Y_1 (**1**).

The FABMS of eudistomin Y_6 (**6**) showed an isotopic cluster of $[M + H]^+$ ion peaks at *m/z* 523/525/527/529 in an approximate ratio of 1:3:3:1, indicating the presence of three bromine atoms in the molecule. A formula of $C_{18}H_9O_2N_2Br_3$ was deduced from the interpretation of its HRFABMS data. The NMR features of **6** closely resemble those of eudistomins Y_4 (**4**) and Y_5 (**5**). Comparison of its ^1H and ^{13}C NMR data with those of **4** and **5** and examination of the 2D NMR experiments revealed that eudistomin Y_6 (**6**) possesses the same β-carboline nucleus as in **4** and the same benzoyl function as in **5**. Hence, eudistomin Y_6 (**6**) was elucidated as a 6,13,15-tribrominated derivative of eudistomin Y_1 (**1**).

High-resolution FABMS of eudistomin Y_7 (**7**) established its molecular formula as $C_{18}H_9O_2N_2Br_3$, which is isomeric with that of eudistomin Y_6 (**6**). The primary difference observed in the ^1H NMR spectrum was that the signals assigned to the 1,2,4-trisubstituted benzene ring in **6** were replaced by another set of ABX spin system. The long-range HMBC correlation of the methine signal at δ 7.48 with the quaternary carbon at δ 119.3 (4b) suggested the C-6 position of the methine signal. The lack of an H-7 signal and the upfield shift of the C-7 signal placed a bromine atom on C-7. The attachment of the bromine atom at C-7 was also confirmed by comparison of its NMR data with those of 7-brominated β-carbolines, as exemplified by eudistomin R.[14,15] Thus, the structure of eudistomin Y_7 (**7**) was determined as a 7,13,15-tribrominated congener of eudistomin Y_1 (**1**).

Up to now, all complex β-carboline metabolites isolated from tunicates have been considered to be biosynthetically related, being derived from tryptophan and a second amino acid. For example, eudistomin X is apparently derived from the condensation of tryptophan and phenylalanine.[4] In the case of eudistomin E and eudistomidin B, the unusual amino acids *S*-methylcysteine and *p*-methylphenylalanine are involved in the biosynthetic pathway in addition to tryptophan.[1,8] The biosynthetic studies of eudistomins H and I with *E. olivaceum* by Shen and co-workers confirmed that tryptophan and proline serve as primary biosynthetic precursors of these two β-carbolines.[14,24] In comparison to the previously described β-carboline metabolites from tunicates, eudistomins Y_1−Y_7 exhibit a different carbon skeleton and represent the first examples of β-carbolines of marine origin substituted at the C-1 position by a benzoyl group. The coupling of tryptophan and *p*-hydroxyphenylglycine might provide a plausible pathway to these new β-carboline metabolites. Taking into account the recent reports that tyrosine is a biosynthetic precursor of *p*-hydroxyphenylglycine in actinomycetes,[25,26] and the important roles that microbes play in the biosynthesis of secondary metabolites from hosts and their symbiotic organisms, it may be proposed that the common amino acid tyrosine is involved in the biosynthetic pathway.

The antibacterial activity of the β-carboline congeners was determined by the 2-fold microtiter broth dilution method.[27] Eudistomin Y_6 (**6**) exhibited moderate antibacterial activity against Gram-positive bacteria *Staphylococcus epidermidis* ATCC12228 and *Bacillus subtilis* ATCC 6633 with MICs of 12.5 and 25 μg/mL, respectively, but showed no inhibitory activity toward the other two strains of Gram-positive bacteria, *S. aureus* ATCC 6538 and *Micrococcus lutes* ATCC 9341, and the Gram-negative bacteria including *Escherichia coli* ATCC 11775, *Salmonella typhimurium*

Table 1. ^{1}H and ^{13}C NMR Data for Compounds **1–4**[a]

	1[b]		**2**[b]		**3**[c]		**4**[c]	
position	δ_H (J in Hz)	δ_C	δ_H (J in Hz)	δ_C	δ_H (J in Hz)	δ_C	δ_H (J in Hz)	δ_C
1		139.0		139.0		136.0		137.1
3	8.56 (d, 4.93)	138.5	8.60 (d, 4.93)	138.5	8.54 (d, 4.94)	136.9	8.58 (d, 4.73)	137.3
4	8.39 (d, 4.93)	119.2	8.43 (d, 4.93)	119.4	8.43 (d, 4.94)	118.4	8.50 (d, 4.73)	119.1
4a		131.0		131.1		130.8		129.9
4b		122.1		123.7		120.1		122.0
5	8.32 (br d, 7.88)	122.6	8.53 (d, 1.49)	125.4	8.32 (br d, 7.85)	121.8	8.62 (br s)	124.5
6	7.35 (t, 7.88)	121.3		113.5	7.31 (br t, 7.85)	120.1		112.2
7	7.64 (t, 7.88)	129.9	7.76 (dd, 8.65, 1.82)	132.5	7.60 (td, 7.85, 1.00)	128.8	7.74 (dd, 8.73, 1.59)	131.4
8	7.89 (br d, 7.88)	113.6	7.86 (d, 8.65)	115.5	7.79 (br d, 7.85)	112.9	7.77 (d, 8.73)	114.9
8a		142.5		141.2		141.5		140.3
9a		137.5		137.6		135.7		135.9
10		192.9		192.4		189.7		189.9
11		130.2		130.2		130.5		129.4
12	8.47 (d, 8.69)	135.0	8.47 (d, 8.76)	135.1	8.55 (d, 1.97)	136.4	8.57 (br s)	136.4
13	7.00 (d, 8.69)	115.6	7.00 (d, 8.76)	115.5		109.5		108.8
14		162.7		162.7		158.8		158.3
15	7.00 (d, 8.69)	115.6	7.00 (d, 8.76)	115.5	7.03 (d, 8.10)	116.0	7.15 (d, 8.52)	115.7
16	8.47 (d, 8.69)	135.0	8.47 (d, 8.76)	135.1	8.21 (dd, 8.10, 1.97)	132.6	8.25 (dd, 8.52, 1.93)	132.5
N*H*	11.32 (s)		11.46 (s)		11.95 (s)		12.13 (s)	
O*H*	not observed		9.33 (br s)		not observed		11.33 (br s)	

[a] The ^{1}H NMR spectra were recorded at 500 MHz, while the ^{13}C NMR spectra were recorded at 125 MHz. [b] Spectra were recorded in acetone-d_6. [c] Spectra were recorded in DMSO-d_6.

Table 2. ^{1}H and ^{13}C NMR Data for Compounds **5–7** in DMSO-d_6[a]

	5		**6**		**7**	
positions	δ_H (J in Hz)	δ_C	δ_H (J in Hz)	δ_C	δ_H (J in Hz)	δ_C
1		136.1		136.6		136.8
3	8.57 (d, 4.67)	137.1	8.59 (d, 4.94)	137.4	8.60 (d, 4.93)	137.7
4	8.47 (d, 4.67)	119.0	8.52 (d, 4.94)	119.5	8.49 (d, 4.93)	119.2
4a		131.1		130.1		130.5
4b		120.0		122.0		119.3
5	8.33 (br d, 7.87)	121.8	8.62 (br s)	124.5	8.31 (d, 8.39)	123.8
6	7.32 (t, 7.87)	120.2		112.3	7.48 (dd, 8.39, 1.69)	123.2
7	7.61 (t, 7.87)	129.0	7.73 (dd, 8.67, 1.66)	131.5		121.8
8	7.82 (br d, 7.87)	113.0	7.76 (d, 8.67)	115.0	7.98 (d, 1.69)	115.6
8a		141.6		140.3		142.5
9a		135.8		136.0		135.9
10		188.8		188.7		188.9
11		130.7		130.3		130.4
12	8.55 (s)	135.3	8.52 (s)	135.4	8.53 (s)	135.4
13		111.0		111.0		111.1
14		154.8		155.1		155.2
15		111.0		111.0		111.1
16	8.55 (s)	135.3	8.52 (s)	135.4	8.53 (s)	135.4
N*H*	12.02 (s)		12.16 (s)		12.12 (s)	
O*H*	9.15 (br s)		not observed		not observed	

[a] The ^{1}H NMR spectra were recorded at 500 MHz, while the ^{13}C NMR spectra were recorded at 125 MHz.

ATCC 14028, and *Klebsiella pneumoniae* ATCC 4352. Eudistomins Y_1 (**1**) and Y_4 (**4**) also displayed the same selectivity as **6**, but demonstrated weak antibacterial activity against the two strains of bacteria *S. epidermidis* ATCC12228 and *B. subtilis* ATCC 6633 with MICs of 50 and 200 μg/mL, respectively. There was no obvious structure–activity relationship observed in this data set. All the compounds showed no cytotoxicity in the MTT assay at the concentration of 100 μM. Therefore, these compounds may serve as lead compounds for development of new antibiotics targeting some pathogenic strains of Gram-positive bacteria.

Experimental Section

General Experimental Procedures. UV spectra were obtained in MeOH using a Scinco UVS-2100 spectrophotometer. IR spectra were measured by a Thermo Electron Corp. Nicolet 570 spectrometer. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker Avance DPX-500 instrument. Chemical shifts were reported with reference to the respective solvent peaks and residual solvent peaks [δ_H 2.05, δ_C 29.9 and 206.7 for acetone-d_6, δ_H 2.50 and δ_C 39.5 for DMSO-d_6]. FABMS spectra were measured on a JEOL JMS-AX505WA mass spectrometer. HPLC was performed with a Synergi Fusion-RP column (250 × 10 mm, 4 μm, 80 Å) using a Younglin M 720 UV detector.

Animal Material. The tunicate *Eudistoma* sp. was collected with the aid of scuba at a depth of 10−15 m off the shore of Tong-Yeong City, South Sea, Korea, in July 2005. The colonies are characterized by a massive round shape and brick red color. The maximum dimension of a colony is 15 × 12 × 8 cm. The fresh specimen was immediately frozen and kept at −24 °C until chemically investigated. A voucher specimen (BYD-25) was deposited in the Center for Marine Natural Products and Drug Discovery, Seoul National University, Seoul, Korea.

Bioassay Procedures. The following seven microorganisms, obtained from the stock culture collection at the American Type Culture Collection (Maryland), were used in this bioassay study: *Staphylococcus aureus* ATCC 6538, *Micrococcus lutes* ATCC 9341, *Staphylococcus epidermidis* ATCC12228, *Bacillus subtilis* ATCC 6633, *Escherichia coli* ATCC 11775, *Salmonella typhimurium* ATCC 14028, and *Klebsiella pneumoniae* ATCC 4352. The antibacterial activity was determined by the 2-fold microtiter broth dilution method.[27] Dilutions of test compounds dissolved in dimethyl sulfoxide (DMSO) were added to each well of a 96-well microtiter plate containing a fixed volume of standard methods broth (SM broth, Difco) (final 0.5% DMSO). Each

well was inoculated with an overnight culture of bacteria (10 CFU/mL) and incubated at 37 °C for 24 h. The minimum inhibitory concentration (MIC) was taken as the concentration at which no growth was observed.

Extraction and Isolation. The lyophilized specimens (dry wt 336 g) were cut into small pieces and extracted three times with 50% MeOH in CH_2Cl_2 at room temperature. These extracts were combined and partitioned three times between *n*-hexane and MeOH. Then, the MeOH-soluble layer was further partitioned between EtOAc and H_2O three times. The EtOAc layer (1.15 g) was active in the antibacterial assay and was subjected to silica flash column chromatography, eluting with a step gradient solvent system of EtOAc and *n*-hexane, to afford 10 fractions. Among them, active fractions were further separated by reversed-phase HPLC (Synersi Fusion-RP, 250 × 10 mm, 4 μm, 80 Å, 210 nm), eluting with 65% CH_3CN in H_2O (0.0025% TFA) to afford compounds **1**−**7** (Figure 1) as yellow, amorphous solids. The overall purified metabolites were isolated in the following amounts: 2.5, 2.0, 6.4, 5.3, 14.0, 13.9, and 17.1 mg for **1**–**7**, respectively.

Eudistomin Y_1 (1): yellow, amorphous solid; IR (film) υ_{max} 3335, 1678, 1599, 1246, 1214, 1164 cm^{-1}; UV (MeOH) λ_{max} (log ϵ) 223 (3.46), 301 (3.25), 382 (2.87) nm; ^1H and ^{13}C NMR data, see Table 1; LRFABMS *m/z* 121, 167, 289 $[M + H]^+$, 311 $[M + Na]^+$; HRFABMS *m/z* 289.0977 (calcd for $C_{18}H_{13}O_2N_2$, 289.0977).

Eudistomin Y_2 (2): yellow, amorphous solid; IR (film) υ_{max} 3338, 1598, 1470, 1270, 1213, 1164, 975, 852 cm^{-1}; UV (MeOH) λ_{max} (log ϵ) 233 (3.74), 307 (3.75), 387 (3.27) nm; ^1H and ^{13}C NMR data, see Table 1; LRFABMS *m/z* 245/247, 367/369 $[M + H]^+$, 389/391 $[M + Na]^+$; HRFABMS *m/z* 367.0083 (calcd for $C_{18}H_{12}O_2N_2{}^{79}Br$, 367.0082).

Eudistomin Y_3 (3): yellow, amorphous solid; IR (film) υ_{max} 3411, 1683, 1592, 1315, 1208, 1138, 1063, 842, 802 cm^{-1}; UV (MeOH) λ_{max} (log ϵ) 223 (3.81), 297 (3.56), 387 (3.29) nm; ^1H and ^{13}C NMR data, see Table 1; LRFABMS *m/z* 167, 199/201, 367/369 $[M + H]^+$, 389/391 $[M + Na]^+$; HRFABMS *m/z* 367.0081 (calcd for $C_{18}H_{12}O_2N_2{}^{79}Br$, 367.0082).

Eudistomin Y_4 (4): yellow, amorphous solid; IR (film) υ_{max} 3421, 1654, 1637, 1617, 1591, 1560, 1471, 1318, 1269, 1246, 1208, 980, 840 cm^{-1}; UV (MeOH) λ_{max} (log ϵ) 240 (3.88), 306 (3.91), 389 (3.57) nm; ^1H and ^{13}C NMR data, see Table 1; LRFABMS *m/z* 245/247, 445/447/449 $[M + H]^+$, 467/469/471 $[M + Na]^+$; HRFABMS *m/z* 444.9188 (calcd for $C_{18}H_{11}O_2N_2{}^{79}Br_2$, 444.9187).

Eudistomin Y_5 (5): yellow, amorphous solid; IR (film) υ_{max} 3429, 1705, 1621, 1577, 1492, 1465, 1427, 1312, 1283, 1241, 1207, 988, 849 cm^{-1}; UV (MeOH) λ_{max} (log ϵ) 224 (3.79), 296 (3.61), 388 (3.50) nm; ^1H and ^{13}C NMR data, see Table 2; LRFABMS *m/z* 121, 445/447/449 $[M + H]^+$, 467/469/471 $[M + Na]^+$; HRFABMS *m/z* 444.9184 (calcd for $C_{18}H_{11}O_2N_2{}^{79}Br_2$, 444.9187).

Eudistomin Y_6 (6): yellow, amorphous solid; IR (film) υ_{max} 3435, 1670, 1611, 1563, 1469, 1409, 1344, 1269, 1245, 1205, 1048, 993, 803 cm^{-1}; UV (MeOH) λ_{max} (log ϵ) 239 (3.97), 304 (3.95), 392 (3.75) nm; ^1H and ^{13}C NMR data, see Table 2; LRFABMS *m/z* 245/247, 277/279/281, 523/525/527/529 $[M + H]^+$; HRFABMS *m/z* 522.8293 (calcd for $C_{18}H_{10}O_2N_2{}^{79}Br_3$, 522.8292).

Eudistomin Y_7 (7): yellow, amorphous solid; IR (film) υ_{max} 3420, 1668, 1606, 1558, 1464, 1416, 1362, 1278, 1242, 1203, 1052, 803 cm^{-1}; UV (MeOH) λ_{max} (log ϵ) 225 (3.68), 299 (3.42), 392 (3.28) nm; ^1H and ^{13}C NMR data, see Table 2; LRFABMS *m/z* 245/247, 277/279/281, 523/525/527/529 $[M + H]^+$, 545/547/549/551 $[M + Na]^+$, 567/569/571/573 $[M - H + 2Na]^+$; HRFABMS *m/z* 522.8293 (calcd for $C_{18}H_{10}O_2N_2{}^{79}Br_3$, 522.8292).

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Supporting Information Available: NMR spectra for compounds **1**–**7**. This material is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) Rinehart, K. L.; Kobayashi, J.; Harbour, G. C.; Hughes, R. G. Jr.; Mizsak, S. A.; Scahill, T. A. *J. Am. Chem. Soc.* **1984**, *106*, 1524–1526.
(2) Kobayashi, J.; Harbour, G. C.; Gilmore, J.; Rinehart, K. L., Jr. *J. Am. Chem. Soc.* **1984**, *106*, 1526–1528.
(3) Kinzer, K. F.; Cardellina, J. H., II. *Tetrahedron Lett.* **1987**, *28*, 925–926.
(4) Schupp, P.; Poehner, T.; Edrada, R.; Ebel, R.; Berg, A.; Wray, V.; Proksch, P. *J. Nat. Prod.* **2003**, *66*, 272–275.
(5) Kobayashi, J.; Nakamura, H.; Ohizumi, Y.; Hirata, Y. *Tetrahedron Lett.* **1986**, *27*, 1191–1194.
(6) Kobayashi, J.; Cheng, J. F.; Ohta, T.; Nozoe, S.; Ohizumi, Y.; Sasaki, T. *J. Org. Chem.* **1990**, *55*, 3666–3670.
(7) Murata, O.; Shigemori, H.; Ishibashi, M.; Sugama, K.; Hayashi, K.; Kobayashi, J. *Tetrahedron Lett.* **1991**, *32*, 3539–3542.
(8) Adesanya, S. A.; Chbani, M.; Pais, M.; Debitus, C. *J. Nat. Prod.* **1992**, *55*, 525–527.
(9) Kang, H.; Fenical, W. *Nat. Prod. Lett.* **1996**, *9*, 7–12.
(10) Van Wagoner, R. M.; Jompa, J.; Tahir, A.; Ireland, C. M. *J. Nat. Prod.* **1999**, *62*, 794–797.
(11) Hudson, J. B. *Antiviral Res.* **1989**, *12*, 55–74.
(12) Hudson, J. B.; Saboune, H.; Abramowski, Z.; Towers, G. H. N.; Rinehart, K. L., Jr. *Photochem. Photobiol.* **1988**, *47*, 377–381.
(13) Ishida, J.; Wang, H. K.; Bastow, K. F.; Hu, C. Q.; Lee, K. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 3319–3324.
(14) Shen, G. Q.; Baker, B. J. *Tetrahedron Lett.* **1994**, *35*, 1141–1144.
(15) VanWagenen, B. C.; Cardellina, J. H., II. *Tetrahedron Lett.* **1989**, *30*, 3605–3608.
(16) Kobayashi, M.; Chen, Y.; Aoki, S.; In, Y.; Ishida, T.; Kitagawa, I. *Tetrahedron* **1995**, *51*, 3727–3736.
(17) Bourguet-Kondracki, M. L.; Martin, M. T.; Guyot, M. *Tetrahedron Lett.* **1996**, *37*, 3457–3460.
(18) Leonard, M. S.; Carroll, P. J.; Joullie, M. M. *Synth. Commun.* **2004**, *34*, 863–869.
(19) Garcia, M. D.; Wilson, A. J.; Emmerson, D. P. G.; Jenkins, P. R. *Chem. Commun.* **2006**, *24*, 2586–2588.
(20) Radchenko, O. S.; Novikov, V. L.; Elyakov, G. B. *Tetrahedron Lett.* **1997**, *38*, 5339–5342.
(21) Crow, W. D.; Hancox, N. C.; Johns, S. R.; Lamberton, J. A. *Aust. J. Chem.* **1970**, *23*, 2489–2501.
(22) Shin, J.; Lee, H.-S.; Seo, Y.; Rho, J.-R.; Cho, K. W.; Paul, V. J. *Tetrahedron* **2000**, *56*, 9071–9077.
(23) Tabudravu, J. N.; Eijsink, V. G. H.; Gooday, G. W.; Jaspars, M.; Komander, D.; Legg, M.; Synstad, B.; Van Aalten, D. M. F. *Bioorg. Med. Chem.* **2002**, *10*, 1123–1128.
(24) Shen, G. Q.; Baker, B. J. *Tetrahedron Lett.* **1994**, *35*, 4923–4926.
(25) Hubbard, B. K.; Thomas, M. G.; Walsh, C. T. *Chem. Bio.* **2000**, *7*, 931–942.
(26) Choroba, O. W.; Williams, D. H.; Spencer, J. B. *J. Am. Chem. Soc.* **2000**, *122*, 5389–5390.
(27) Kim, S.; Oh, K.-B. *J. Microbiol. Biotechnol.* **2002**, *12*, 1006–1009.

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