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CHEMISTRY OF SPONGES, VII.[1] 11,19-DIDEOXYFISTULARIN 3 AND 11-HYDROXYAEROTHIONIN, BROMOTYROSINE DERIVATIVES FROM *PSEUDOCERATINA DURISSIMA*

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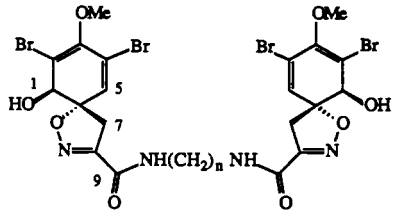
ABSTRACT.—11,19-Dideoxyfistularin 3 [**3**] and 11-hydroxyaerothionin [**6**], two new bromotyrosine-derived metabolites, have been isolated from the verongid sponge *Pseudoceratina durissima*, along with the known compounds aerothionin [**1**] and homoaerothionin [**2**].

Marine sponges in the order Verongida are distinct both chemically and biologically from those in other orders of the Porifera (1,2). All genera of the Verongida chemically examined so far contain secondary metabolites that are derived from bromotyrosine or from chlorotyrosine (3,4). In such metabolites the side chain has been converted into a variety of nitrogenous groups while the aromatic ring has either been retained or has undergone rearrangement or partial reduction.

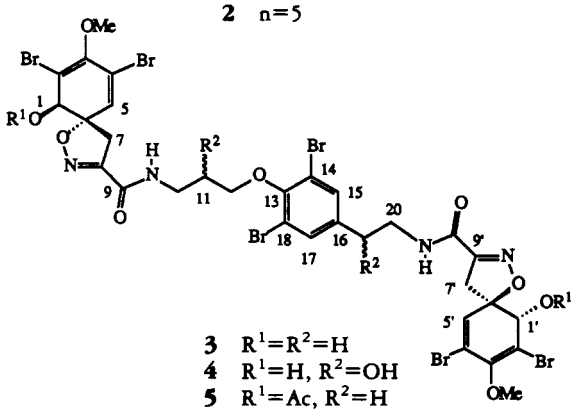
Interest in the sponge *Pseudoceratina durissima* Carter (Family Aplysinellidae) collected from the Great Barrier Reef was stimulated by the potent in vitro antimicrobial activity of its CH_2Cl_2 extract. Separation of the crude extract on Sephadex LH-20 gave a single antimicrobial fraction that was purified by cc on Si gel to give the known metabolites aerothionin [**1**] (5) (0.34%) and homoaerothionin [**2**] (6) (0.11%) and two new metabolites, 11,19-dideoxyfistularin 3 [**3**] (0.33%) and 11-hydroxyaerothionin [**6**] (0.09%). Aerothionin [**1**], whose absolute configuration has been established (7), and homoaerothionin [**2**] were identified from spectral data, including optical rotations, that were identical with literature values (5,6). The structures of 11,19-dideoxyfistularin 3 [**3**] and 11-hydroxyaerothionin [**6**] were determined by spectroscopic methods.

11,19-Dideoxyfistularin 3 [**3**] was obtained as an optically active and unstable yellow powder. The compound failed to show a molecular ion in either the eims, the desorption ms, or the fabms. The molecular formula $C_{31}H_{30}Br_6N_4O_9$ was therefore determined from the ^{13}C-nmr spectrum (31 carbons, 26 attached protons), the ^1H-nmr spectrum (4 D_2O exchangeable protons in Me_2CO-d_6), and the elemental analysis. The ir spectrum contained bands characteristic of alcohol, amine, and secondary amide groups (3500, 3350, 1645 cm^{-1}), while the uv spectrum had absorptions at λ max 284 (ϵ 10,400) (cisoid diene), 257 (16,000), and 224 nm (26,000), indicative of a cyclohexadienyl moiety. Like aerothionin [**1**], 11,19-dideoxyfistularin 3 [**3**] had a ^1H-nmr spectrum that indicated two dibromospirocyclohexadienylisoxazole ring systems (5,8). A ^1H-^1H COSY of **3** allowed assignment of the ^1H-nmr signals associated with these systems. A two-proton signal at δ 6.29 (br s, 2H), due to overlapping signals of two distinct olefinic protons (H-5, H-5′), was coupled to signals at δ 3.92 (br d, 1 H, J = 19 Hz) and 3.89 (br d, 1 H, J = 19 Hz). In turn, the signals at δ 3.92 and 3.89 were coupled to signals at δ 3.00 (d, 1H, J = 19 Hz) and 2.98 (d, 1H, J = 19 Hz) respectively. The signal at δ 6.29 also showed coupling to a signal at δ 4.34 (br s, 2H) that was assigned to the two methine protons (H-1, H-1′). The signals at δ 3.92 and 3.00 were assigned to the methylene protons (H-7) of one of the isoxazole rings, and the sig-

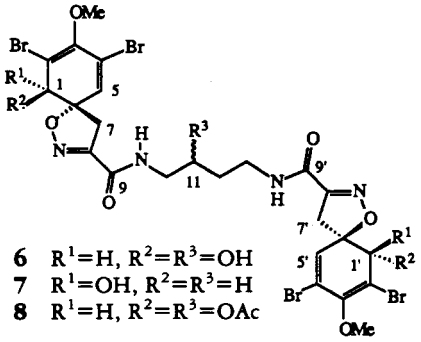
[1]For Part VI, see Karuso *et al.* (15).



1 n=4
2 n=5



3 $R^1=R^2=H$
4 $R^1=H$, $R^2=OH$
5 $R^1=Ac$, $R^2=H$



6 $R^1=H$, $R^2=R^3=OH$
7 $R^1=OH$, $R^2=R^3=H$
8 $R^1=H$, $R^2=R^3=OAc$

nals at δ 3.89 and 2.98 were assigned to the methylene protons (H-7′) of the second isoxazole ring system. In addition, the ^{1}H-nmr spectrum of **3** contained signals arising from two methoxyl groups [δ 3.72 (s, 3H), 3.71 (s, 3H)], and a signal at δ 7.37 (br s, 2H) of an isolated aryl proton that indicated a symmetrically tetrasubstituted aromatic ring. The COSY of **3** showed a long range coupling from the signal at δ 7.37 to a signal at δ 2.76 (br t, 2H, $J=8$ Hz) that was assigned to a benzylic methylene group; the signal at δ 2.76 showed further coupling to a signal at δ 3.54 (t, 2H, $J=8$ Hz). These ^{1}H-nmr signals suggested the presence of an Ar-CH_2-CH_2-X system. The remainder of the spectrum contained signals due to an NH-CH_2-CH_2-CH_2-X system: δ 7.24 (br t, 1H, $J=6$ Hz, NH), 3.67 (m, 2H), 2.09 (m, 2H), and 4.06 (t, 2H, $J=6$ Hz), the connectivities of which were determined from the COSY.

The complete structure of 11,19-dideoxyfistularin 3 [**3**] was established from comparison of its ^{13}C-nmr spectrum with those of aerothionin [**1**], diacetylhexadellin A [**10**] (9), diacetylhexadellin B [**11**] (9), and psammaplysin A [**9**] (10,11) (Table 1). A two-dimensional ^{13}C-, ^{1}H-nmr chemical shift correlation experiment (Table 2) allowed complete assignment of all protonated carbons of 11,19-dideoxyfistularin 3.

11,19-Dideoxyfistularin 3 [**3**] is related to fistularin 3 [**4**] from the verongid sponge *Aplysina fistularis* forma *fulva* (8) and differs from **4** in that the two secondary al-

TABLE 1. ^{13}C-nmr Spectra of Aerothionin [**1**][a], 11,19-Dideoxyfistularin 3 [**3**][b], 11-Hydroxyaerothionin [**6**][b], Psammaplysin A [**9**][b,c], Diacetylhexadellin A [**10**][b,c], and Diacetylhexadellin B [**11**].[b,c]

Carbon	Compound					
	1	**3**[d]	**6**[d]	**9**	**10**	**11**
C-1, -1′	75.1	73.9, 73.8 (d)	73.8 (d, 2C)	67.4	73.2	73.1
C-2, -2′	122.0	121.4 (s, 2C)	121.4, 121.3 (s)	117.9	121.7	122.1
C-3, -3′	148.7	147.9 (s, 2C)	147.7 (s, 2C)	148.0	149.7	149.7
C-4, -4′	113.8	112.7 (s, 2C)	113.1 (s, 2C)	103.9	107.8	107.8
C-5, -5′	132.3	130.9, 130.7 (d)	130.6, 130.6 (d)	144.9	130.5	130.2
C-6, -6′	91.5	91.8, 91.6 (s)	91.9, 91.8 (s)	103.9	89.7	89.9
C-7, -7′	40.2	38.9, 38.8 (t)	38.7 (t, 2C)	36.7	40.1	39.9
C-8, -8′	155.3	154.1, 154.0 (s)	153.9 (s, 2C)	153.9	153.6	153.5
C-9, -9′	160.0	159.3 (s, 2C)	160.0, 160.0 (s)	156.6	158.6	158.6
C-10	39.4	37.2 (t)	36.2 (t)	36.7	37.3	37.7
C-11	26.7	29.2 (t)	68.0 (d)	28.8	29.3	29.4
C-12		71.1 (t)	45.0 (t)	70.5	71.2	72.1
C-13		151.3 (s)	33.6 (t)	151.5	151.2	151.5
C-14, -18		118.1 (s, 2C)		119.4	118.1	118.2
C-15, -17		132.9 (d, 2C)		132.6	132.8	132.8
C-16		137.4 (s)		139.5	137.9	137.2
C-19		34.2 (t)		31.5	34.5	34.4
C-20		40.4 (t)		40.1	60.2	40.1
OMe	60.2	60.1, 60.1 (q)	60.0 (q, 2C)	58.4	60.2	60.2

[a]In Me_2CO-d_6.

[b]In $CDCl_3$.

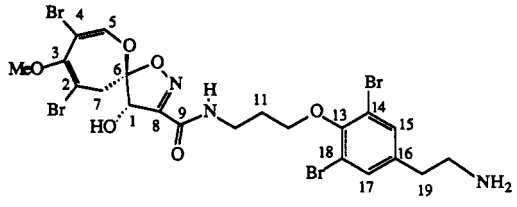
[c]Psammaplysin A, diacetylhexadellin A, and diacetylhexadellin B were renumbered for nmr comparisons.

[d]Proton attachments determined via DEPT.

cohol groups in **4** at C-11 and C-17 are replaced by methylene groups in **3**. Acetylation of **3** gave a diacetate **5** whose ^{1}H-nmr spectrum showed a close correspondance to that recorded for fistularin 3 tetraacetate (8). Like fistularin 3 and fistularin 3 tetraacetate (12), 11,19-dideoxyfistularin 3 did not give a fabms. However, the deims of **3** showed a pentet centered at *m/z* 725 that was attributed to the major fragment after cleavage of **3** at the C-8, -9 or C-8′, -9′ bond, followed by loss of H_2O. Subsequent loss of a bromine atom from this species gave the peaks at *m/z* 647 that were observed in the cims of **3**. A positive ion fabms of the diacetate **5** showed two clusters of seven ions centered at *m/z*

TABLE 2. ^{13}C-^{1}H-nmr Chemical Shift Assignments for Protonated Carbons of 11,19-Dideoxyfistularin 3 [**3**] ($CDCl_3$).

Assignment	δ ^{13}C	δ ^{1}H
C-15	132.9 (d, 2C)	7.37 (br s, 4H)
C-5, C-5′	130.9, 130.7 (d)	6.29 (br s, 1H), 6.28 (br s, 1H)
C-1, C-1′	73.9, 73.8 (d)	4.34 (br s, 2H)
C-12	71.1 (t)	4.06 (t, 2H, 6 Hz)
OMe	60.1, 60.0 (q)	3.73 (s, 3H), 3.72 (s, 3H)
C-20	40.4 (t)	3.54 (t, 2H, 8 Hz)
C-7, C-7′	38.9, 38.8 (t)	3.92 (d, 1H, 19 Hz), 3.89 (d, 1H, 19 Hz) 3.00 (d, 1H, 19 Hz), 2.98 (d, 1H, 19 Hz)
C-10	37.2 (t)	3.67 (m, 2H)
C-19	34.2 (t)	2.76 (br t, 2H, 8 Hz)
C-11	29.2 (t)	2.09 (m, 2H)



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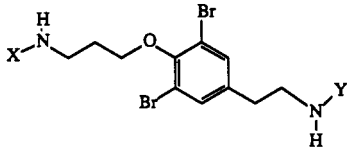
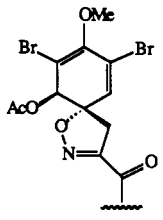
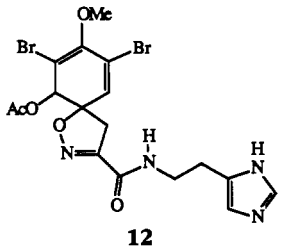


10 X=A, Y=Ac
11 X=Ac, Y=A



A



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1198 and *m/z* 1182 that were assigned to glycerol adducts of the protonated molecule: $[M-60+G]^+$, $[M-60-16+G]^+$. The same types of species have been reported in the fabms of aerophobin 1 acetate [**12**] (12). Treatment of 11,19-dideoxyfistularin 3 with methanolic KOH afforded the diphenol **13**; the fabms of **13** had a pentet centered at *m/z* 718 and a quartet at *m/z* 638, 640 that were assigned to the major fragment after cleavage of the molecule at the C-9 or C-9′ amide bond followed by loss of a bromine atom.

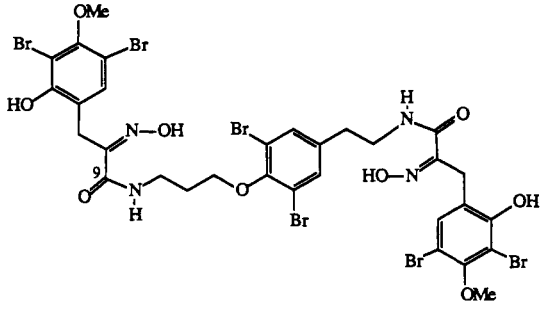
The hydroxyl groups at C-1 and C-1′ in 11,19-dideoxyfistularin 3 [**3**] were determined to be cis to the methylene carbons at C-7 and C-7′, respectively, from comparison of the observed ^{1}H-nmr chemical shifts with those of aerothionin [**1**], *cis,cis*-aerothionin [**7**], amide **14**, and amide **15** (Table 3). Me_2CO-d_6 was used as nmr solvent for this comparison because the ^{1}H-nmr spectra of **1**, **7**, **14**, and **15** are reported in this

TABLE 3. Comparison of ^{1}H-nmr Chemical Shifts of Aerothionin [**1**], 11,19-Dideoxyfistularin 3 [**3**], 11-Hydroxyaerothionin [**6**], *cis,cis*-Aerothionin [**7**], **14**, and **15** in Me_2CO-d_6.[a]

Proton	Compound					
	1	**3**	**6**	**7**	**14**	**15**
H-1	4.18(7.5)	4.16(6)	4.16(7)	4.52(7.5)	4.22(7.5)	4.53(7.5)
H-5	6.53	6.51	6.51	6.60	6.52	6.58
H-7	3.15(18)	3.18(18)	3.12(18)	3.2–3.5	3.17(18)	3.42
	3.85(18)	3.83(18)	3.83(18)		3.85(18)	
1-OH	5.40(7.5)	5.46(6)	5.46(7)	4.97(7.5)	5.38(7.5)	4.98(7.5)

[a]Values in parentheses are coupling constants (Hz).

solvent (5,13); the chemical shifts of the appropriate signals in Me_2CO-d_6 differ considerably from those in $CDCl_3$ [e.g., H-1, -1′ δ 4.06 ($CDCl_3$), 4.16 (Me_2CO-d_6) for **3**]. The signals at δ 4.16 and 6.51, assigned to H-1, H-1′ and H-5, H-5′, respectively, are more similar to those in aerothionin [**1**] and **14** (13) than to those in *cis,cis*-aerothionin [**7**] (13) or in **15** (13). Furthermore, the spectra of both **7** and **15** have broad ^{1}H-nmr signals attributed to the geminal C-7 protons, while the corresponding nmr signals in **1**, **14**, and 11,19-dideoxyfistularin 3 [**3**] are observed as two doublets because of the interaction of the hydroxyl group with one of the geminal protons. Finally, the long range correlations observed in the COSY experiment ($CDCl_3$) between the signals at δ 6.29 and 4.34 and the signals at δ 6.29 and 3.90 require a relatively planar orientation for **3** between C-5, C-6, C-7, and H-7 (14), which is only possible in the trans, trans configuration found in **1** and **14**. The structure of **3** is thus as pictured. Only relative stereochemistry is implied, and the particular enantiomer drawn was arbitrarily picked to accord to the stereochemistry found in aerothionin.



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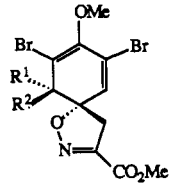

14 R^1=H, R^2=OH
15 R^1=OH, R^2=H

11-Hydroxyaerothionin [**6**] was obtained as an optically active colorless glass for which the molecular formula $C_{24}H_{26}Br_4N_4O_9$ was determined from the elemental analysis and ^{13}C-nmr spectrum (24 carbons). The positive ion fabms showed a cluster of five ions centered at *m/z* 857 that was attributed to $[M+Na]^+$. The ir spectrum of **6** showed bands due to alcohol, amide, and amine groups (3400, 3350, 1645 cm^{-1}) while the uv spectrum had absorptions due to a *cis*-dienoid group [λ max 284 (ε 11,100), 233 (19,750)]. The ^{1}H-nmr spectrum of **6** was very similar to that of aerothionin [**1**]. Signals at δ 6.16 (br s, 2H), 4.10 (d, 2H, J = 6 Hz), 3.72 (d, 1H, J = 18 Hz), 3.70 (d, 1H, J = 18 Hz), 3.59 (s, 6H), and 2.85 (d, 2H, J = 18 Hz) were assigned to two dibromospirocyclohexadienyl ring systems. In addition, the ^{1}H-nmr spectrum of **6** contained signals that required an -NH-CH_2-CH_2-CH(OH)-CH_2-NH- group. Signals assigned to a methylene group [δ 3.46 (dd, 1H, J = 14, 3 Hz), 3.32 (dd, 1H, J = 14, 7 Hz)] showed a correlation in the COSY to a signal at δ 3.60 (m, 1H) that in turn was coupled to signals assigned to a second methylene group [δ 1.73 (m,

1H), 1.61 (m, 1H)]. The signals at δ 1.73 and 1.61 showed additional coupling to signals due to the third methylene group [δ 3.37 (m, 1H), 3.55 (m, 1H)]. These data suggested that 11-hydroxyaerothionin had the structure **6**. Comparison of the ^{13}C-nmr spectrum of 11-hydroxyaerothionin with the ^{13}C-nmr spectra of aerothionin [**1**] and homoaerothionin [**2**] revealed that the methylene group at C-11 in aerothionin [**1**] was replaced by a secondary alcoholic group in 11-hydroxyaerothionin. Like aerothionin and 11,19-dideoxyfistularin 3, the two dibromospirocyclohexadienyl rings in **6** have the trans,trans configuration (Table 3). Acetylation of 11-hydroxyaerothionin [**6**] gave the expected triacetate **8**. The structure of **6** is thus as pictured. Only relative stereochemistry is implied, and the enantiomer drawn was arbitrarily picked to accord with that of aerothionin; the relative stereochemistry of the 11-OH group could not be determined from the spectral data.

Although aerothionin [**1**], homoaerothionin [**2**], 11,19-dideoxyfistularin 3 [**3**], and 11-hydroxyaerothionin [**6**] all had in vitro antimicrobial activity, 11,19-dideoxyfistularin 3 was the most active of the metabolites tested. Aerothionin [**1**], homoaerothionin [**2**], and 11-hydroxyaerothionin [**6**] all inhibited the growth of *Staphylococcus aureus* at 100 μg/disk, *Bacillus subtilis* at 50 μg/disk, and *Candida albicans* at 50 μg/disk, whereas 11,19-dideoxyfistularin 3 inhibited the growth of *S. aureus* at 25 μg/disk, *B. subtilis* at 10μg/disk, and *C. albicans* at 25 μg/disk.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—These were as in Karuso *et al.* (15), except as follows. Ir spectra were obtained either on a Shimadzu IR-27G spectrometer or a Bio-Rad FTir spectrometer. All solvents were distilled prior to use. Hplc was carried out with a Shimadzu LC-6A solvent delivery system equipped with a Waters R401 r.i. detector, using a Merck LiChrosorb Si gel column (25 × 1 cm). 2D nmr experiments were performed on a Bruker 400 MHz nmr spectrometer following literature procedures (16,17).

ISOLATION OF NATURAL PRODUCTS.—The freeze-dried sponge (96.12 g) (Ref. No. AUZ-BWJ; British Museum Register No. 83-12-4-28) collected from Bowl Reef, Great Barrier Reef, Australia (at 10 m depth), was extracted exhaustively with CH_2Cl_2 to give a brown oil (3.75 g) that showed in vitro antimicrobial activity against *S. aureus* (Oxford strain), *B. subtilis* (PB 2576), and *C. albicans* (ATCE 12061). A portion (1.25 g) of the extract was purified by cc on Sephadex LH-20 CH_2Cl_2-MeOH (1:1) followed by cc on silica (0–10% MeOH gradient in CH_2Cl_2) to give 11,19-dideoxyfistularin 3 (105 mg), homoaerothionin (34 mg), aerothionin (110 mg), and 11-hydroxyaerothionin (29 mg). Aerothionin was crystallized from CH_2Cl_2, mp 130–132°, [lit. (5) 134–137°]; $[\alpha]_D$ +250° [lit. (5) +252°].

11,19-DIDEOXYFISTULARIN 3 [**3**].—The compound was obtained as an unstable yellow powder: $[\alpha]_D$ +98.5° (*c* = 0.10). Found N 4.9, Br 43.2; $C_{31}H_{30}Br_6N_4O_9$·MeOH requires C 34.5, H 3.1, N 5.0, Br 43.1. Correct elemental analysis for C and H not observed. Uv λ max (MeOH) 284 (ε 10,400), 257 (16,000), 224 (26,000); ir ν max (film) 3450, 3350 (OH, NH_2), 1645 cm^{-1} (CONH); ^{1}H nmr (Me_2CO-d_6) δ 7.78 (t, *J* = 6 Hz, NH), 7.73 (t, *J* = 6 Hz, NH), 7.50 (s, 2H, H-15, H-17), 6.51 (d, *J* = 1 Hz, H-5 or H-5′), 6.50 (d, *J* = 1 Hz, H-5′ or H-5), 5.46 (d, *J* = 6 Hz, OH), 5.45 (d, *J* = 6 Hz, OH), 4.16 (d, *J* = 6 Hz, H-1 or H-1′), 4.15 (d, *J* = 6 Hz, H-1′ or H-1), 4.07 (t, *J* = 6 Hz, H-12), 3.83 (d, *J* = 18 Hz, H-7a or H-7′a), 3.81 (d, *J* = 18 Hz, H-7′a or H-7a), 3.71 (s, 6H, OMe), 3.60 (td, *J* = 7, 6 Hz, H-20), 3.53 (ddd, *J* = 6, 6, 6 Hz, H-10), 3.18 (d, *J* = 18 Hz, H-7b or H-7′b), 3.15 (d, *J* = 18 Hz, H-7′b or H-7b), 2.86 (t, *J* = 7 Hz, H-19), 2.11 (m, H-11); ^{13}C nmr ($CDCl_3$) see Table 1; deims *m/z* 729 (15% base peak), 727 (35), 725 (45), 723 (25), 721 (10), 666 (20), 664 (40), 662 (40), 660 (20), 646 (25), 644 (40), 642 (40), 640 (20), 323 (10), 321 (20), 319 (10), 293 (40), 191 (80), 295 (40), 267 (50), 265 (100), 263 (50), 212 (80), 210 (80); dcims (NH_3^+) *m/z* 649 (2), 647 (4), 645 (2), 633 (1.5), 631 (2), 629 (1.5), 569 (2), 567 (4), 565 (2), 451 (16), 449 (25), 447 (14), 381 (10), 379 (20), 377 (10), 298 (30), 296 (60), 294 (30), 267 (50), 265 (100), 263 (50).

ACETYLATION OF 11,19-DIDEOXYFISTULARIN 3 [**3**].—A solution of 11,19-dideoxyfistularin 3 (10 mg) in pyridine (0.5 ml) and Ac_2O (0.5 ml) was stirred at room temperature for 3 h. Toluene was added, and the resulting azeotrope was evaporated in vacuo. The product was purified on Si gel (50% EtOAc/hexane) to give the diacetate **5** (8.0 mg, 70%). Found N 4.3, Br 40.2; $C_{35}H_{34}Br_6N_4O_{11}$·MeOH requires H 3.2, N 4.7, Br 40.1. Correct elemental analysis for C and H not observed. Uv λ max (MeOH) 280

(ε 10,400), 225 (27,100), 204 (27,400); ir ν max ($CHCl_3$) 1740 (br, OAc), 1670, 1665 cm^{-1} (CONH); ^1H nmr ($CDCl_3$) δ 7.35 (br s, 2H), 7.18 (br t, 1H, J = 6 Hz), 6.77 (br t, 1H, J = 6 Hz), 6.31 (br s, 1H), 6.31 (br s, 1H), 5.82 (br s, 1H), 5.81 (br s, 1H), 4.07 (t, 2H, J = 5.5 Hz), 3.75 (s, 3H), 3.74 (s, 3H), 3.67 (q, 2H, J = 6.3 Hz), 3.55 (m, 2H), 3.44 (d, 1H, J = 18 Hz), 3.42 (d, 1H, J = 18 Hz), 3.07 (d, 1H, J = 18 Hz), 3.06 (d, 1H, J = 18 Hz), 2.78 (t, 2H, J = 7.1 Hz), 2.13 (s, 3H), 2.13 (s, 3H), 2.07 (m, 4H); ^{13}C nmr ($CDCl_3$) δ 169.5 (s), 158.7 (s), 158.6 (s), 153.7 (s), 153.6 (s), 151.4 (s), 149.7 (s), 137.3 (d), 132.8 (d, 2C), 130.6 (d), 130.3 (d), 121.9 (s), 121.6 (s), 118.2 (d, 2C), 107.9 (s), 107.8 (s), 89.9 (s), 89.7 (s), 73.1 (d), 73.0 (d), 70.9 (t), 60.2 (q), 60.1 (q), 40.3 (t), 39.9 (t), 39.8 (t), 37.1 (t), 34.3 (t), 29.2 (t), 20.7 (q, 2C); fabms (glycerol, EtOH, H^+ matrix) *m/z* 1204 (3.5% base peak), 1202 (7), 1200 (14), 1198 (21), 1196 (13), 1194 (7), 1192 (3.5), 1188 (13), 1186 (25), 1184 (50), 1182 (100), 1180 (50), 1178 (25), 1176 (13), 1142 (4), 1140 (8), 1138 (16), 1136 (32), 1134 (16), 1132 (8), 1130 (4), 1126 (7), 1124 (14), 1122 (28), 1120 (55), 1118 (28), 1116 (14), 1114 (7).

ALKALINE TREATMENT OF 11,19-DIDEOXYFISTULARIN 3 [**3**].—A solution of 11,19-dideoxyfistularin 3 (20 mg) in 3% methanolic KOH (20 ml) and H_2O (5 ml) was refluxed for 3 h. After evaporation of the MeOH the residue was extracted with CH_2Cl_2 (3 × 5 ml). The combined CH_2Cl_2 extracts were evaporated and recrystallized from MeOH to give the oximinophenol **13** (14 mg, 70%) as white needles, mp 90–93°; ν max (film) 3360 (br, OH, NH), 1652 (CONH), 1620, 1540, 1464, 1418, 1055, 757 cm^{-1}; ^1H nmr ($CDCl_3$) δ 7.60 (t, J = 6 Hz, NH), 7.55 (s, H-5 or H-5′), 7.53 (s, H-5′ or H-5), 7.24 (s, H-15 and H-17), 7.07 (t, J = 6 Hz, NH), 3.98 (t, J = 7 Hz, H-12), 3.85 (s, OMe), 3.84 (s, OMe), 3.82 (s, H-7 or H-7′), 3.81 (s, H-7′ or H-7), 3.65 (td, J = 7, 6 Hz, H-10), 3.50 (td, J = 7, 6 Hz, H-20), 2.73 (t, J = 7 Hz, H-19), 2.04 (m, H-11); ^{13}C nmr ($CDCl_3$) δ 165.1 (s), 164.9 (s), 152.9 (s), 150.6 (s), 140.1 (s), 137.3 (s), 134.1 (d, 2C), 132.9 (d, 2C), 120.0 (s, 2C), 118.1 (s, 2C), 109.1 (s), 107.1 (s), 71.2 (t), 60.5 (q, 2C), 40.6 (t), 37.6 (t), 34.0 (t), 30.9 (t), 29.7 (t), 28.9 (t); fabms (glycerol, MeOH, H^+ matrix) *m/z* 722 (20% base peak), 720 (70), 718 (100), 716 (70), 714 (20), 642 (20), 640 (60), 638 (60), 636 (10).

11-HYDROXYAEROTHIONIN [**6**].—The compound was obtained as a colorless glass: $[\alpha]_D$ +189° (c = 0.15). Found C 35.3, H 3.5, Br 38.1; $C_{24}H_{26}Br_4N_4O_9$ requires C 35.5, H 3.2, Br 38.4. Uv λ max (MeOH) 284 (ε 11,100), 233 (19,750), 205 (18,900); ir ν max (film) 3400, 3350 (OH, NH), 1650 cm^{-1} (CONH); ^1H nmr ($CDCl_3$ + CD_3OD, 4:1) δ 6.17 (s, H-5, H-5′), 4.11 (s, H-1 or H-1′), 4.10 (s, H-1′ or H-1), 3.71 (d, J = 18 Hz, H-7a or H-7′a), 3.70 (d, J = 18 Hz, H-7′a or H-7a), 3.60 (m, H-11), 3.59 (s, 6H, OMe), 3.55 (m, H-13a), 3.46 (dd, J = 14, 3 Hz, H-10a), 3.37 (m, H-13b), 3.32 (dd, J = 14, 7 Hz, H-10b), 2.85 (d, J = 18 Hz, H-7b, H-7′b), 1.73 (m, H-12a), 1.61 (m, H-12b); ^1H-nmr (Me_2CO-d_6) δ 7.81 (br t, J = 6 Hz, NH), 7.56 (br t, J = 6 Hz, NH), 6.51 (s, H-5, H-5′), 5.47 (d, J = 7 Hz, OH), 5.46 (d, J = 7 Hz, OH), 4.16 (d, J = 7 Hz, H-1, H-1′), 3.83 (d, J = 18 Hz, H-7a or H-7′a), 3.82 (d, J = 18 Hz, H-7′a or H-7a), 3.79 (m, 1H), 3.71 (s, 6H, OMe), 3.52 (m, 1H), 3.43 (m, 2H), 3.29 (m, 1H), 3.12 (d, J = 18 Hz, H-7b, H-7′b), 1.97 (m, H-12a), 1.60 (m, H-12b); ^{13}C-nmr ($CDCl_3$ + CD_3OD, 4:1) 160.3 (s), 154.1 (s), 147.9 (s), 130.8 (d), 121.8 (s), 121.7 (s), 113.5 (s), 91.9 (s), 91.8 (s), 73.9 (d), 68.1 (d), 60.3 (q), 45.3 (t), 39.1 (t), 36.4 (t), 33.8 (t); ^{13}C-nmr ($CDCl_3$) see Table 1; fabms (glycerol, EtOH, H^+ matrix) *m/z* 861 (25% base peak), 859 (50), 857 (100), 855 (50), 853 (25), 845 (10), 843 (20), 841 (40), 839 (20), 837 (10), 779 (25), 777 (50), 775 (25), 732 (22), 730 (45), 728 (22).

ACETYLATION OF 11-HYDROXYAEROTHIONIN [**6**].—A solution of 11-hydroxyaerothionin (8 mg) in pyridine (0.5 ml) and Ac_2O (0.5 ml) was stirred at room temperature for 3 h. Toluene was added, and the resulting azeotrope was evaporated in vacuo. The product was purified on Si gel (20% EtOAc/CH_2Cl_2) to give the triacetate **8** (7.2 mg, 82%): uv λ max (MeOH) 280 (ε 13,200), 234 (13,950), 204 (17,560); ir ν max ($CHCl_3$) 1740 (br) (OAc), 1670, 1665 (CONH) cm^{-1}; ^1H nmr ($CDCl_3$) δ 6.98 (br t, 1H, J = 6 Hz), 6.85 (br t, 1H, J = 6 Hz), 6.33 (br s, 1H), 6.32 (br s, 1H), 5.83 (s, 1H), 5.81 (s, 1H), 5.03 (m, 1H), 3.78 (s, 3H), 3.77 (s, 3H), 3.43–3.62 (m, 3H), 3.43 (d, 1H, J = 18 Hz), 3.40 (d, 1H, J = 18 Hz), 3.35 (m, 1H), 3.07 (d, 2H, J = 18 Hz), 2.15 (s, 6H), 2.12 (s, 3H), 1.85 (m, 2H); ^{13}C nmr ($CDCl_3$) δ 171.1 (s), 169.7 (s), 159.0 (s), 158.8 (s), 153.7 (s), 153.6 (s), 149.9 (s), 149.8 (s), 130.6 (d), 130.2 (d), 122.2 (s), 121.8 (s), 107.9 (s), 107.8 (s), 89.9 (s), 89.8 (s), 73.2 (d), 70.7 (d), 60.2 (q), 42.3 (t), 39.8 (t), 35.6 (t), 31.3 (t), 31.0 (t), 29.7 (t), 21.1 (q), 20.7 (q, 2C).

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