L) during 5–6 h of stirring. The extract was evaporated under vacuum, yielding an oily residue (9 g), which was redissolved in methylene chloride–methanol (19:1, 20 mL) and fractionated on a column (200 mL) of E.M. silica gel 60 (230–400 mesh) packed in hexane–methylene chloride (1:1). Elution of the column was carried out by a step gradient of methylene chloride in hexane yielding partially purified compound in the 80% methylene chloride fractions. After evaporation of the solvent under reduced pressure, the residue (600 mg) was taken up in methanol (4 mL). Upon refrigeration, a precipitate formed which was removed by filtration. The filtrate was further fractionated in two identical runs (2 mL each) on a Rainin Dynamax 60A C_{18} column (1 in. × 25 cm), eluted with a 15 mL/min gradient of acetonitrile–water (7:13 to 9:1) over 40 min. Appropriate fractions were evaporated to dryness, yielding variecolin (95 mg); the homogeneity was verified by HPLC (Whatman Partisil ODS-3) eluted with acetonitrile–H_2O (3:1, k' 5.2) and by TLC on silica gel 60 F_{254} (E. Merck) (R_f 0.40 in CH_2Cl_2 and R_f 0.52 in hexane–acetone, 4:1) and Whatman KC18 plates (R_f 0.43 in ACN–H_2O, 90:10, and R_f 0.50 in MeOH–H_2O, 95:5).

Variecolin (1): $[\alpha]_D$ −11.5° (c 0.50, ACN); EI-MS m/z 360 (M^+); IR 1735, 1687, 1626, 1455, 1404, 1382, 1228, 1208, 1192, 1141, 933, 884, 839, 808, 768, 735, 711 cm^{-1}; UV (MeOH) λ_{max} (ϵ) 241 (12 130) and 203 (9020); ^1H NMR (CD_3CN and C_6D_6) see Table I; ^{13}C NMR (CD_3CN) see Table I. Anal. Calcd for $C_{25}H_{36}O_2$: C, 18.46; H, 9.85. Found: C, 81.53; H, 9.77.

Novel Sponge-Derived Amino Acids. 12. Tryptophan-Derived Pigments and Accompanying Sesterterpenes from *Fascaplysinopis reticulata*†

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Received June 21, 1990

This paper reports the bioactive constituents of *Fascaplysinopsis reticulata* collected from the Benga Lagoon of the Fiji islands. The previous literature of this genus includes two aplysinopsins (monomeric tryptophans) from *F. reticulata*, as well as fascaplysin (**5a**) (an apparent tryptophan dimer) and luffariellolide (**3**) (a sesterterpene) from *Fascaplysinopsis* sp. Our investigation of *F. reticulata* has revealed new sesterterpenes isodehydrouffariellolide (**1**) and dehydroluffariellolide diacid (**2**); unique alkaloid-sesterterpene salts fascaplysin A (**5b**) [fascaplysin cation/dehydroluffariellolide diacid anion] and homofascaplysin A cation/dehydroluffariellolide diacid anion (**6**); and novel neutral alkaloids homofascaplysin C (**7**), homofascaplysin B (**8**), and secofascaplysin A (**9**). These substances were accompanied by fascaplysin (**5a**) and the known alkaloid (+)-octopamine **4**. The most important findings in this study are (a) fascaplysin derivative **5b** is the first known salt comprised of a complex alkaloid cation and a terpene carboxylate anion, and (b) secofascaplysin A (**9**) is the first naturally occurring β-carbolinone. An amino acid biogenesis pathway is outlined for each of the above alkaloids. The biological activity profile against the HIV reverse transcriptase is reported for selected metabolites.

Nitrogen-containing metabolites are rarely observed from Dictyoceratid sponges as this group is an excellent source of di- or sesterterpenes.[1,2] A few atypical members of the Dictyoceratid family Thorectidae are sources of both sesterterpenes and amino acid derivatives.[2,3] In 1985 we collected *Fascaplysinopsis reticulata* (Thorectidae family, Dictyoceratida order) which were eye-catching because of their massive, globular, and shiny red-brown appearance.[4] This sponge was targeted for further study when the crude extracts of a 1987 collection exhibited significant bioactivity against bacteria [(inhibition zone diameter size in millimeters at 100 μg/disk) including *Staphylococcus aureus* (18), *Streptococcus pyrogenes* (11), *Candida albicans* (24), and *Trichophyton mentagrophytes* (7)] and virus [100% inhibition against reverse transcriptase at 1 mg/mL; IC_{50}'s (μg/mL), HIV on ALEX cells = 0.4, ALEX cell control = 6.2].[5]

The natural products of the genus *Fascaplysinopsis* have been the subject of prior publications. Some time ago the Roche group isolated two aplysinopsins, monomeric tryptophans, from *F. reticulata*.[6] Significantly, these were unaccompanied by terpenoids and there was no mention of biological activity properties. More recently, luffariellolide (**3**), a known sesterterpene, and fascaplysin (**5a**), a 12*H*-pyrido[1,2-*a*:3,4-*b'*]diindole, were reported by Ireland and Clardy from a Fijian collection of *Fascaplysinopsis* sp.[7a] A total synthesis of this alkaloid has just been completed by Gribble.[7b] Our comprehensive study of *F. reticulata* involved four separate Fijian collections, and its vary complex mixtures consisted of sesterterpenes,

†Presented at the 199th National Meeting of the American Chemical Society, April 1990, Boston, MA, Abstr no. 356.

(1) For reviews see: (a) Crews, P.; Naylor, S. *Prog. Chem. Org. Nat. Prod.* **1985**, *48*, 203. (b) Hanson, J. R. *Nat. Prod. Rep.* **1986**, *3*, 87.

(2) For examples see: Bergquist, P. R.; Wells, R. J. In *Marine Natural Products*; Scheuer, P. J., Ed.; Academic Press: New York, 1983; Vol. V pp 35–42.

(3) Consult Table 3 in ref 1a and Tables 3 and 4 in ref 2.

(4) Our voucher collection (no. 89051) was identified by C. Diaz, UCSC Institute of Marine Sciences and Prof. R. W. M. van Soest, Institute of Taxonomic Zoology, University of Amsterdam. An underwater photo can be supplied by P.C. Particularly distinctive traits are: the thick outer ectosome layer which embeds and grains, the very sharp conules (2 mm high and 5 mm apart), and the absences of spicules. These properties and its appearance are similar to those (including the photograph) reported for *F. reticulata*: Bergquist, P. R. *New Zeal. J. Zool.* **1980**, *7*, 443.

(5) These results were provided by Dr. Tom Matthews (Syntex Research, Palo Alto, CA) and his staff. Purified cloned HIV-1 reverse transcriptase was assayed by a previously described procedure: Chen, M. S.; Oshana, S. C. *Biochem. Pharm.* **1987**, *36*, 4361.

(6) Kazlauskas, R.; Murphy, P. T.; Quinn, R. J.; Wells, R. J. *Tetrahedron Lett.* **1977**, 61 (see ref 2, Table 4, for the correct taxonomy of this sponge).

(7) (a) Roll, D. M.; Ireland, C. M.; Lu, H. S. M.; Clardy, J. *J. Org. Chem.* **1988**, *53*, 3276. (b) Pelcman, B.; Gribble, G. W. *Tetrahedron Lett.* **1990**, *31*, 2381.

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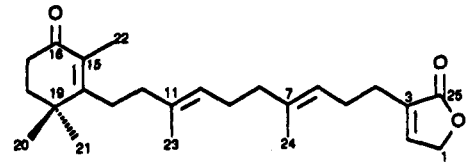

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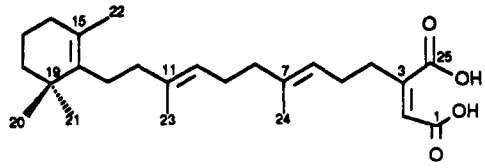

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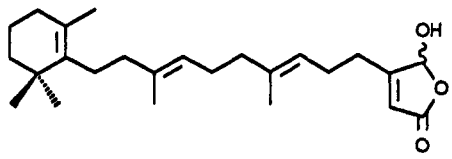

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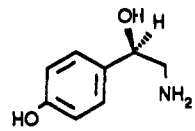

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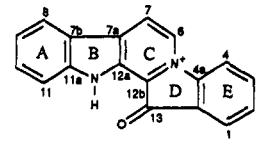

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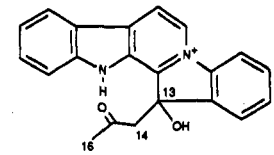
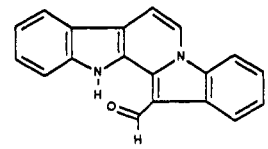
b anion = 2^-



6

anion = 2^-



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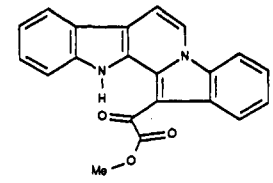

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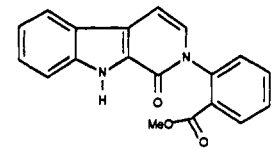

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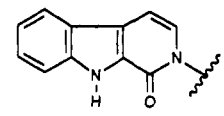

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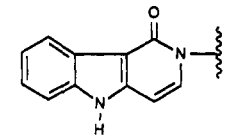

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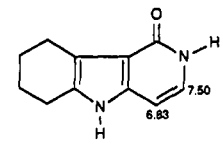


12

alkaloids, and alkaloid-sesterterpene salts. Several new compounds are described below, including the following: two sesterterpenes, isodehydroluffariellolide (1) and dehydroluffariellolide diacid (2); two alkaloid-sesterterpene salts, fascaplysin A (**5b**) [fascaplysin cation/dehydroluffariellolide diacid anion] and homofascaplysin A cation/dehydroluffariellolide diacid anion (**6**); and three neutral alkaloids, homofascaplysin C (**7**), homofascaplysin B (**8**), and secofascaplysin A (**9**). These were accompanied by **5a** and (+)-octopamine (**4**).[8]

Results and Discussion

There are striking morphological features of *F. reticulata*. This includes a thick red-brown dense outer layer which covers a less dense drab inner derma. Such an organization is reminiscent of examples in which photosynthetic symbionts (especially cyanobacteria) are limited to the outer layer of dense sponges due to insufficient light in the inner tissues.[9] It seemed worthwhile to investigate whether the chemical composition of *F. reticulata* depended on the tissue layer analyzed. Consequently, the 1989 collection was carefully cut to separate the outer ectosome from the endosome prior to the extraction of this sponge. Formalin-preserved specimens from each of these preparations were subsequently cut into thick sections and examined by fluorescence microscopy (blue light excitation). Disappointingly, both the ectosome and endosome were equally devoid of a cyanobacteria population. Furthermore, as will be shown below, both the ectosome and endosome exhibited a parallel pattern in the relative compositions of the different types of metabolites.

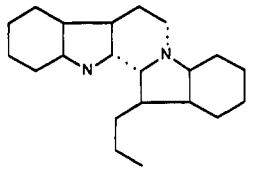
Four separate collections of this sponge were gathered by SCUBA during July–August between 1985 and 1989 (none obtained in 1986) from the Benga Lagoon, Fiji Islands, at 10–20 m. Specimens were either extracted immediately after collection or preserved for a brief period before extraction. Representative results are as follows. A viscous crude oil (7.2 g) was obtained from the CH_3OH extract of the 1987 material (2.5 kg wet weight), and then submitted to solvent partitioning. The compositions of the 1987–1989 collections were studied in detail. The CCl_4 partition fraction of all three contained neutral compounds **1**, **7**, and **8**. Alternatively, salt **5b** was present in all three collections in the CH_2Cl_2 fractions. Individual differences in the composition of the extracts was as follows: **2** (endosome, CCl_4, 1989 collection), **4** (BuOH, 1988 collection), **5a** (ectosome and endosome, CH_2Cl_2, 1989 collection), **6** (CCl_4, 1987 collection), **9** (CCl_4, 1987, 1988, and ectosome 1989 collections). The ectosome and endosome preparations of the 1989 collection were studied simultaneously and did not show any significant differences in their chemical composition (**1**, **2**, **5a**, **5b**, **7**, **8**, and **9**), with the singular exception that no **2** was found in the former and no **9** was found in the latter.

The characterization of isodehydroluffariellolide (**1**), an oil of molecular formula $C_{25}H_{36}O_3$ (HREIMS 384.2653 = M^+, Δ 0.2 mmu of calcd) and major component in the CCl_4 partition fractions, was completed first. Two α,β-unsaturated carbonyl functionalities, as a substituted trimethylcyclohexenone **i** and a butenolide array of **iii** or **iv**

(8) (a) See Merck Index no. 6599. (b) Octopamine has been recently investigated in skeletal muscle receptor binding assays: Evans, P. D.; Thonor, C. Mohan; Midgley, J. M. *J. Pharm. Pharmacol.* **1988**, *40*, 855.

(9) For an overview see: Wilkinson, C. R. In *Endosymbiosis and Cell Biology*; Schwemmler, W., Schenk, H. E. A., Eds.; Walter de Gruyter & Co.: New York, 1980; Vol. I, pp 553–63.

(Chart I), were intimated by a characteristic UV absorption



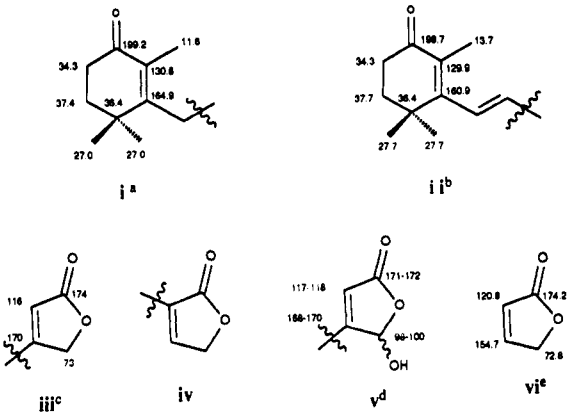
A

(249 nm), IR bands (1753 and 1652 cm^{-1}), MS fragmentation, [**i**: m/z = 233.1537, ($M^+ - C_{10}H_{15}O$), Δ 0.1 mmu of calcd; m/z = 137.0969 ($C_9H_{13}O$), Δ 0.6 mmu of calcd], and ^{13}C NMR resonances [**i**: δ 199 (s), 131 (s), 165 (s); **iii** or **iv**: δ 175 (s), 144 (d), 135 (s), 70 (t)]. Moiety **i** has similar ^{13}C shifts as compared to array **ii** in the carotenoid canthaxanthin[10] (Chart I). Substructure **iv** was favored over **iii** by the relatively low-field shift of H-2[11] (^1H–^1H COSY NMR correlation observed between δ 7.10 [H-2] and 4.76 [H-1]), by comparison to the butenolide ring proton chemical shifts of the mokupalides[12] (see **iii**, Chart I), and by the similarity of the observed shifts of **1** (see above) to those calculated from a model butenolide ring with R at C-3 of δ 135 (C-3) and 146 (C-4) by adding standard ^{13}C substituent shift increments to the data of **vi**[13] (Chart I). There are other relevant examples of this substructure from sesterterpenes which have an extra OH (as in **v**) and include luffariellolide[14] (**3**), manoalides,[15] luffariellins,[16] and cacospongionolide.[17] Appropriate data for substructure **iv** could only be obtained by adding standard as shown (Chart I) for **iii'** and **iv'**. The structure of **1** was completed by comparing the NMR data of the unsaturated chain from C-4 to C-13 to that in several related sesterterpenes.[1,15]

The above structure and data of **1** along with those in the literature for luffariellolide (**3**)[14] provided a straightforward way to elucidate the structure of dehydroluffariellolide diacid (**2**). Its molecular formula of $C_{25}H_{38}O_4$ was established from MS data (negative ion FABMS: 401, $M^- - H$; LREIMS: 384, $M^+ - H_2O$) and ^{13}C APT NMR data. Both the ^{13}C and ^1H NMR spectra of **2** were completely assigned by comparison to almost identical resonance patterns and position of luffariellolide (**3**).[14]

Mass spectrometry provided the most definitive indication that fascaplysin A (**5b**), an amorphous reddish solid, was a salt comprised of sesterterpene and alkaloid subunits. The molecular formula of the sesterterpene anion was deduced as $C_{25}H_{37}O_4$ by negative ion FABRMS (401 = M^-) and HREIMS (384.2665 = $M^- - OH$, Δ 1.0 mmu of calcd); while the alkaloid cation was assigned as $C_{18}H_{11}N_2O$ by positive ion FABMS (271 = M^+). The ^1H and ^{13}C NMR spectra unveiled the overall framework for each member of this pair. The sesterterpene was assigned structure **2**$^-$ because ^{13}C signals nearly identical with those of luffariellolide (**3**)[14] were observed with the exception of the two C=O resonances at δ 169.8 (s) and 170.0 (s) and the shifted vinyl resonances at δ 148.3 (s) and 131.4 (d).[18] The overall

Chart I.[a] **A. Experimental ^{13}C NMR Shifts**



[a] (a) Data from **1**. (b) Data from canthaxanthin in ref 10. (c) Data from mokupalides in ref 12. (d) Data from various sesterpenes in refs 12–17. (e) Data from ref 13.

pattern of ^{13}C and ^1H signals of the alkaloid cation paralleled those reported for fascaplysin (**5a**).[7a] Thus, the complete structure of this salt could be assigned as **5b** and a 1:1 ratio was evident between the constituent ions as equivalent ^1H NMR integrations were measured for H-2/6/10 (anion) versus three of the aromatic ring protons (cation) as shown in Figure 1a. Chemical confirmation was provided when **5b** was transformed into **5a** with basic anion exchange resin chromatography (Dowex-1, chloride form). Moreover, treatment of **5b** with aqueous HCl afforded **5a** and dehydroluffariellolide dicarboxylic acid (**2**) in an approximate ratio of 1:1. The isolation of this salt was reproducible as it was isolated from the CH_2Cl_2 solvent partition fractions of both the 1987 and 1989 collections.

Homofascaplysin A (**6**), a dark red/purple amorphous salt, was also comprised of sesterterpene and alkaloid subunits. The sesterterpene anionic component was assigned as **2**$^-$ owing to the identical nature of its ^{13}C and ^1H NMR shifts in comparison to those of fascaplysin A (**5b**). The cation array in **6** was of molecular formula $C_{21}H_{17}N_2O_2$ (positive HRFABMS: 329.1295 = M^+, Δ 0.9 mmu of calcd), and possessed additional C_3H_6O atoms in comparison to **5b**. Evidence of geminal hydroxyl and 2-oxopropyl groups at position 13 of the fascaplysin skeleton was as follows. Three key MS fragmentations include: loss of OH to 312.1265 ($C_{21}H_{16}N_2O$ Δ 2.8 mmu of calcd); loss of $COCH_3$ to 287 ($C_{19}H_{13}N_2$); and loss of C_3H_6O via a retroaldol cleavage to 271.0879 ($C_{18}H_{11}N_2O$ = **5a** cation, Δ 0.7 mmu of calcd). The ^{13}C NMR spectrum had shifts corresponding to those of the pentacyclic core of fascaplysin (**5a**) except the carbonyl signal (C-13) was missing and a new quaternary signal was observed at δ 78.2 (C-13). Also, there were new resonances at δ 30.5 (q, C-16), 204.6 (s, C-15), and 51.0 (t, C-14). The ^1H–^1H COSY NMR spectrum also revealed the polyaromatic ring core (one AB and two ABCD systems) and a NH group (δ 14.1) along with additional resonances (an AB pattern) at δ 4.71 and 3.98 (d, J = 18 Hz, CH_2-14) and singlet resonance at 1.94 ppm (CH_3-16). The ^{13}C and ^1H NMR differences between

(10) Moss, G. P. *Pure Appl. Chem.* **1976**, *47*, 97.
(11) Compound **vi** (see Chart I) has ^1H NMR shifts of δ 6.15 (H-α) and 7.63 (H-β) and the β-substituted butenolide ring of mokulpolide[12] has δ 5.49 (H-α) in mokupalide.
(12) Yunker, M. B.; Scheuer, P. J. *J. Am. Chem. Soc.* **1978**, *100*, 307.
(13) Spectrum 11950c In *Sadtler Standard ^{13}C NMR Indexes*; Sadtler Research Laboratories; Division of Bio-rad Laboratories, Inc.: PA, 1960.
(14) Albizati, K. F.; Holman, T.; Faulkner, D. J.; Glaser, K. B.; Jacobs, R. S. *Experientia* **1987**, *43*, 949.
(15) De Silva, E. D.; Scheuer, P. J. *Tetrahedron Lett.* **1980**, *21*, 1611.
(16) Kernan, M. R.; Faulkner, D. J. *J. Org. Chem.* **1987**, *52*, 3081.
(17) De Rosa, S.; De Stefano, S.; Zavodnik, N. *J. Org. Chem.* **1988**, *53*, 5020.

(18) Comparison of the spectral data of **2** and the anion of salt **5b** showed subtle, yet significant differences which were consistent with the variation in charge between these species. More specifically, differences of the ^{13}C NMR spectrum between **2** and the sesterterpene component of **5b** showed the resonances due to C-1, C-25, and C-2 were shifted by +4.1, +3.8, and +2.6, respectively. Dissociation of the carboxyl proton causes deshieldings from the carboxyl to the α carbon which are attributed to electronic fields. See Breitmaier, E.; Voelter, W. In *Carbon-13 NMR Spectroscopy*; VCH: New York, 1987; p 227.

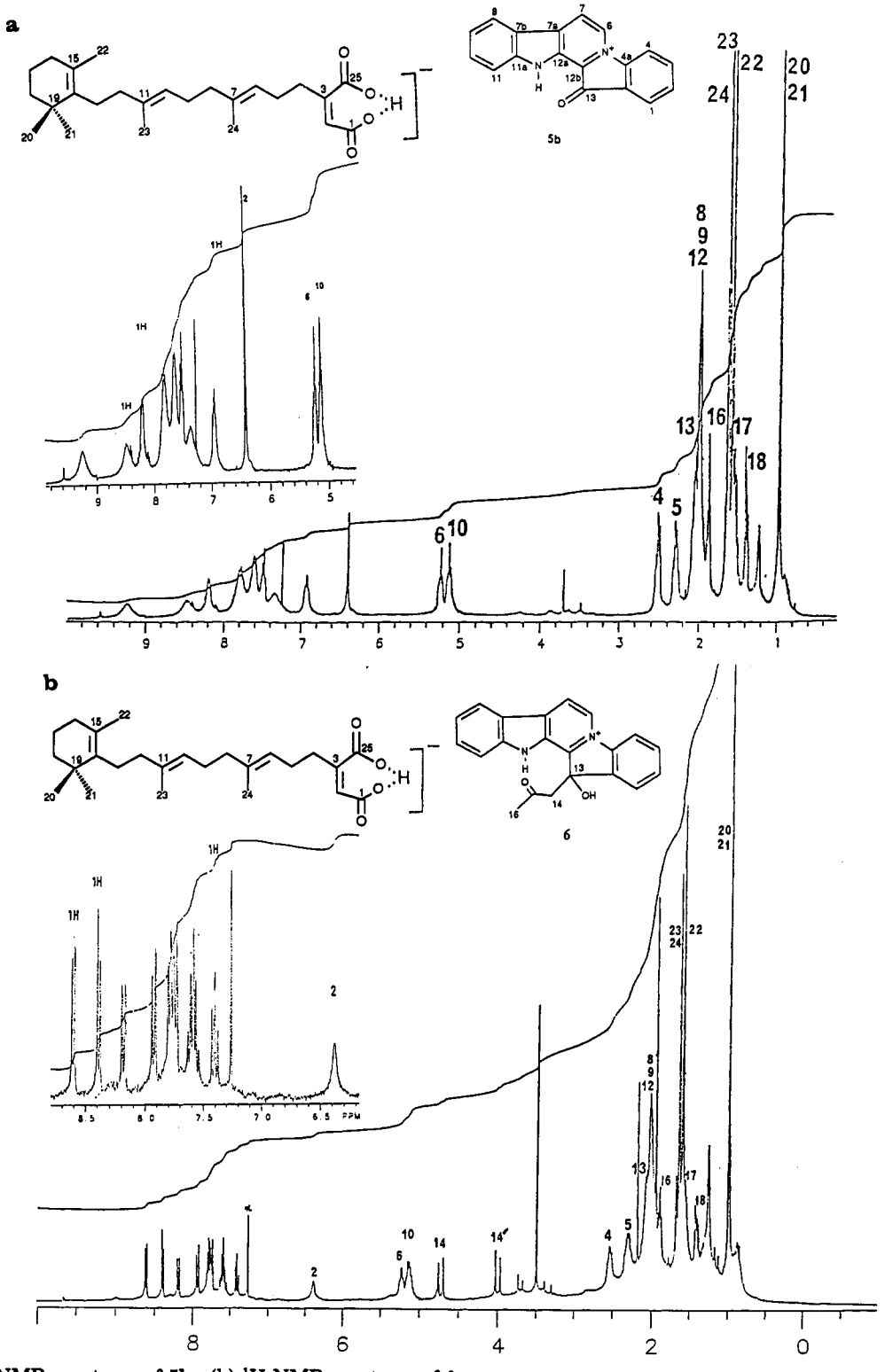
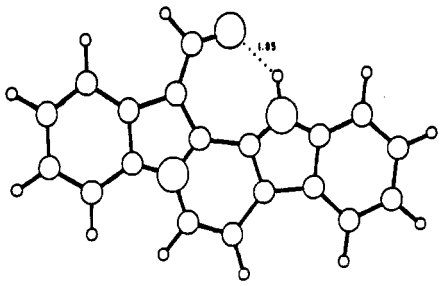


Figure 1. (a) ^{1}H NMR spectrum of **5b**. (b) ^{1}H NMR spectrum of **6**.

the cations of **5b** and **6** could be explained by the new additions from C-13 to C-16 proposed for the latter compound. A 1:1 ratio is evident between the anion and cation units in **6** as estimated by the ^{1}H NMR integration of H-2 (anion) versus each of the two aromatic protons (cation) as shown in Figure 1b. Although **6** does exhibit optical activity ($[\alpha]^{20}_D = -9.36°$, $c = 0.0064$, MeOH) and acetone was never used during this investigation, its structure intimates that it may not be a natural product. Compound **6** was only isolated from one collection, so the paucity of material on hand precluded attempts to chemically interconvert **6** to **5**.

Three additional zoochromic compounds **7**, **8**, and **9** were isolated. Their dark color suggested a polyaromatic system analogous to that in **5** and **6**, but these new compounds showed the lack of both ionic character and a terpenoid constituent. The first of these neutral compounds, homofascaplysin C (**7**), $C_{19}H_{12}N_2O$ ($M^+ = 284.0940$, Δ 0.7 mmu of calcd) displayed ^{13}C and APT NMR spectra showing 11 sp^2 CH and 8 sp^2 quaternary carbons. The ^{13}C and ^{1}H NMR spectra (two aromatic ABCD systems, an isolated AB system, and a NH δ 12.2) again indicated a pyridodiindole framework. In comparison to **5** there were key major differences in the NMR data including the lack



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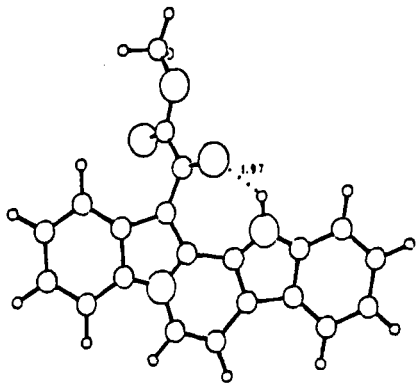

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Figure 2. ORTEP plots of lowest energy conformations of 7 and 8.

of a C=O at δ 186 (C-13) of **5a**) and the presence of new resonances at δ 118.6 (s, C-13) and 181.0 (d, C-14) in the ^{13}C NMR and at δ 10.3 (s, H-14) in the ^{1}H NMR. These signals are characteristic of a 3-formylindole.[19] The IR spectrum contained a strong aldehyde band at 1651 cm^{-1}, suggestive of intramolecular H bonding. A lowest energy structure calculated by molecular mechanics (using MACROMODEL[20]) had an almost perfectly flat ring system (Figure 2) with the carbonyl being slightly out of the plane by 13.9° and pointing toward the NH. The calculated distance of 1.853 Å between the O and the H is consistent with the 1651-cm^{-1} IR band and is compatible with delocalization between the aldehyde and the indole N.

At this point, we realized that the relative shift of the AB system corresponding to H-6/H-7 of the *Fascaplysinopsis* pigments (**5**, **6**, and **7**) was diagnostic of the charged or uncharged nature of these alkaloids. For example, the AB system of the cation counterparts of **5** and **6** exhibited shifts at δ 9.01/8.69[7a] and 8.60/8.38 respectively, whereas the corresponding protons in the neutral system **7** were shielded to δ 8.30/7.67. Similarly, the J values of the AB system varied from 5.93 Hz (**5**)[7a] or 6.3 Hz (**6**) to 7.2 Hz (**7**). These correlations could be directly applied to homofascaplysin B (**8**) of molecular formula $C_{21}H_{14}N_2O_3$ (342.1002, Δ 0.3 mmu of calcd) which possessed the neutral pyridodiindole system as indicated by resonances for H-6/H-7 at δ 8.39/7.79 and J_{AB} = 7.2 Hz. Other NMR chemical shifts of **8** at δ 4.12, 52.9 (OMe), 167.5 (CO-15), and 177.8 (CO-14) indicated that the aldehyde functional group in **7** was replaced by an α-keto methyl ester.[21] This was supported by MS fragments at m/z 283.0871 (M^+ – COOMe) and 255.0925 (M^+ – COCOOMe), and by IR (1600–1568 cm^{-1}, br band). The low field shift of the NH at δ 12.51 also signified an interaction between the C=O and NH. Molecular mechanics calculations predict a minimum energy conformer (Figure 2) with the ketone carbonyl forming dihedral angles of 47.2° with the rings and 163.1° with the carbonyl of the ester. The ketone C=O was also pointing toward the NH providing an O···H distance of 1.969 Å, suggesting the possibility of H bonding.

The last of the neutral pigments, secofascaplysin A (**9**) was tetracyclic rather than pentacyclic. This was established by comparison of the unsaturations deduced from the molecular formula of $C_{19}H_{14}N_2O_3$ (HREIMS m/z 318.1007, M^+, Δ 0.3 mmu of calcd) to those revealed from the NMR data. The presence of eight C–C double bonds and two carbonyls was deduced as follows. All 19 carbons (8 s, 10 d, and 1 q) were observed in the ^{13}C NMR spectrum (125 MHz, $CDCl_3$) while a ^{1}H–^{1}H COSY NMR spectrum (300 MHz, $CDCl_3$) clearly showed the two aromatic ABCD systems (end resonances of the separate systems at δ 8.15, H-1 and 7.97, H-8) and an isolated AB system (δ 7.06 and 7.12). Two carbonyls were assigned with the aid of IR absorptions and ^{13}C NMR shifts as a pyridone (1656 cm^{-1} and δ 156.0 s, C-12b) and a methyl ester (1726 cm^{-1}, δ 165.0 s, C-13, and δ 52.3 q, OMe). An extra tetrasubstituted double bond was envisioned to rationalize the two remaining ^{13}C singlet carbons (between δ 123 and 141). Finally, an indole type NH (δ 10.4, bs) was also observed. Additional firm evidence for the COOMe group was provided by its loss in the HREIMS spectrum (m/z = 287.0793, $C_{18}H_{11}N_2O_2$, M^+ – OMe, Δ 2.8 mmu of calcd; and 259.0861 $C_{17}H_{11}N_2O$, M^+ – COOMe, Δ 1.1 mmu of calcd). Most importantly, the relatively high-field shift of the isolated AB pattern in secofascaplysin A (**9**) indicated an entirely different chromophore in comparison to that in **7** or **8**. A carboline[22] framework was consistent with this data, and four isomers were possible. The J_{C-H} = 178 Hz at δ 128.3 assigned to the C-6 ring-C carbon was inconsistent with an α- or δ-carbolinone derivative. The choice between the remaining β- or γ-carbolinone structures, **10** or **11**, was made with the aid of IR C=O stretch frequencies, ^{1}H NMR δ's, and biogenetic considerations. The observed lactam C=O stretch (1656 cm^{-1}) of secofascaplysin A is closer to that of compounds with general structure **10** (1645–1670 cm^{-1})[23] versus general structure **11** (1629–1648 cm^{-1}).[24] The ^{1}H NMR shifts of the C-ring H's of B- or γ-carbolinones **10** and **11** ought to be different because of the variation in the cross-conjugated environment of the diene. Even though both compound families are known, exact shifts have only been published for the latter, as illustrated by the data of **12** [DMSO-d_6 δ 7.5 (CHN) and 6.83].[25] These C-ring proton shifts are quite different as compared to those of **9** [DMSO-d_6 δ 7.31 (CHN) and 7.13] and suggests a β-carbolinone assignment for this compound. An additional argument for the carbolinone regiochemistry assigned in secofascaplysin A (**9**) is that this framework provides the closest biogenetic link between **9** and **5**. A majority of the NMR resonances in **9** could be unambiguously assigned by ^{1}H–^{1}H and ^{1}H–^{13}C

(19) Shamma, M.; Hindenlang, D. M. *Carbon-13 NMR Shift Assignments of Amines and Alkaloids*; Plenum Press: New York, 1979; no. 477.
(20) MACROMODEL program (version 1.5) on a Vax 11/750 computer with an Evans and Sutherland (PS 300) picture system.
(21) For model compounds with an indole ring having a C-3 α-keto ester or α-keto amide see ref 17, compounds 4847C and 10263C.
(22) For ^{1}H NMR data of the four isomeric carbolines see: Balkau, F.; Heffernan, M. L. *Aust. J. Chem.* **1973**, *26*, 1501.
(23) (a) Chatterjea, J. N.; Swaroop, B. B.; Singh, R. P.; Ojha, N. *J. Ind. Chem. Soc.*, **1987**, *64*, 28. (b) Ohmoto, T.; Nikaido, T.; Koike, K.; Kohda, K.; Sankawa, U. *Chem. Pharm. Bull.* **1988**, *36*, 4588. (c) Mashelkar, U. C.; Usgaonkar, R. N. *Chem. Ind.* **1978**, 35.
(24) (a) Clark, B. A. J.; Parrick, J. *J. Chem. Soc., Perkin Trans. 1* **1974**, 2270. (b) Bisagni, E.; Bourzat, J.-D.; Louisfert, J. A. *Tetrahedron* **1970**, *26*, 2087.
(25) The C-ring proton shift range for the four γ-carbolinones are: CH–N δ 7.0–7.5 and CH δ 6.26–6.83 (DMSO-d_6): see refs 24a and 24b.

COSY results, and our results are superimposable on assignments recently published for other β-carbolines such as keramine-A.[26] These assignments were used to assign the proton and carbon spectra of **7** and **8**.

Fascaplysin A (**5b**) and secofascaplysin A (**9**) are especially unique. In comparison to previous sponge natural products, the former represents the first example of a salt comprised of a complex alkaloid accompanied by a terpenoid carboxylate anion[27] and the latter is the first naturally occurring β-carbolinone to be reported.[28] The union of two tryptophan type precursors[29] as depicted by **A**, provides a plausible most convenient pathway to fascaplysins (**5**), homofascaplysin C (**7**), and homofascaplysin B (**8**).[30] This is consistent with the prior isolation of monotryptophan and monotryptamine derivatives from other Thorectid sponges including *Thorectandra*, *Thorectranda*, and *Smenospongia*.[2] An opening of the D ring of **5** could provide a direct pathway to secofascaplysin A (**9**), and this compound is always accompanied by **5b** along with **7**–**8**.[31] Several of the compounds above were tested for their antiviral (but not antimicrobial) properties. Assay against reverse transcriptase (at 1 mg/mL) revealed inhibitions: **1** = 81%, **2** = not active, **5a** = 58%, and **6** = 94%.

Experimental Section

Multiplicities of ^{13}C NMR resonances were determined from APT or DEPT data and COSY experiments (300 MHz for ^{1}H, 75 MHz for ^{13}C). High-performance liquid chromatography (HPLC) was done using reversed-phase 10-μM columns. Standard pulse sequences[32] were used for the homo COSY (ref 32b) and the hetero COSY (ref 32b) experiments.

Isolation Procedures. The workup of the 1987 collection is representative of how the additional collections made in 1988 and 1989 were processed. The preserved sponge (2.5 kg wet weight) was soaked in CH_2Cl_2 for 24 h. Next, the sponge was soaked in MeOH three successive times for 24 h. The organics were combined and concentrated to yield 7.2 g of a crude viscous oil. As detected by the ^{1}H and ^{13}C NMR spectra, alkaloids and sesterterpenes were the major constituents of the extract. The crude oil was then successively partitioned between equal volumes (500 mL of aqueous MeOH, percent adjusted to produce a biphase solution) and a solvent series (yield in g of oil) of hexanes (2.5 g), CCl_4 (1.3 g), CH_2Cl_2 (1.2 g), and 1-butanol (0.7 g). The partition fractions were then separately chromatographed on Sephadex LH20 (methanol). The chromatography fractions were purified by repeated reversed-phase HPLC (ODS, MeOH–H_2O, 85:15). The CCl_4 partition fraction afforded **1** (34 mg), **6** (12 mg), **7** (9 mg), **8** (11 mg), and **9** (≈3 mg), while the CH_2Cl_2 afforded **5b** (42 mg).

Parallel results from other collections are as follows. The 1988 collection was partitioned into CCl_4 (2.8 g), CH_2Cl_2 (2.6 g), and BuOH (9.2 g). The CCl_4 and BuOH partition fractions were subjected to chromatography and respectively afforded [in order of elution from Sephadex] **1** (450 mg), **9** (5 mg), **8** (1 mg), and **7** (4 mg) from the former and **4** (26 mg) from the latter. The 1989 collection was subdivided (see text) into the ectosome (0.142 kg) and endosome (3.2 kg). Each was separately extracted with MeOH [7.16 and 60.8 g of oil was obtained respectively from the ectosome and endosome], and a portion of each oil was partitioned to yield fractions from the ectosome [7.16 g of oil afforded CCl_4 (1.0 g), CH_2Cl_2 (2.6 g), and BuOH (0.02 g)] and the endosome [30 g of oil afforded CCl_4 (2.5 g), CH_2Cl_2 (1.1 g), and BuOH (1.4 g)]. Analogous mixtures of metabolites were separately isolated [the order is according to elution from Sephadex] and the patterns are as follows: ectosome, CCl_4 partition fraction, **1** (160 mg), **9** (7 mg), **8** (3 mg), **7** (7 mg); CH_2Cl_2 partition fraction, **5a** (20 mg), **5b** (35 mg) plus other uncharacterized sesterterpene alkaloid salts; endosome, CCl_4 partition fraction, **1** (289 mg), **2** (654 mg), **8** (4 mg), **7** (6 mg); CH_2Cl_2 partition fraction, **5a** (10 mg), **5b** (5 mg) plus other uncharacterized sesterterpene alkaloid salts.

Cyanobacteria Analysis. Thick sections of preserved ectosome and endosome samples from 1989 collection were observed for the presence of cyanobacteria at 100× and 250× magnification on a Leitz Diaplan epi-fluorescence microscope with blue light excitation (100-W mercury bulb, I 2/3 filter combination). The ectosome sample and the endosome sample showed only occasional clusters of cyanobacteria and diatoms. Observation of *Jaspis stellifera*, under the same conditions, provided a positive control as it showed a dense layer of cyanobacteria in the outermost layer of sponge tissue and very few in the inside tissues of the sponge.

Isodehydroluffariellolide (1). A colorless oil. IR (neat): 3019, 2932, 1753, 1652, 1450, 1210, 1074 cm^{-1}. UV (MeOH) λ_{max} (ε) 406 (1920), 334 (5069), 249 (28224) nm. NMR ($CDCl_3$) shifts in ppm from Me_4Si with assignments based on assessing the number of attached protons and the COSY data [atom number], ^{13}C δ's at 75 MHz and ^{1}H δ's at 300 MHz: [1] 70.2, 4.76 (d, J = 1.5 Hz, 2 H); [2] 144.4, 7.10 (t, J = 1.5 Hz, 1 H); [3] 134.8; [4] 30.2, 2.27 (m, 2 H); [5] 25.6, 2.27 (m, 2 H); [6] 122.8, 5.11 (m, 1 H); [7] 136.6; [8] 39.6, 2.05 (m, 2 H); [9] 25.8, 2.30 (m, 2 H); [10] 124.7, 5.16 (m, 2 H); [11] 134.0; [12] 38.6, 2.05 (m, 2 H); [13] 26.7, 2.05 (m, 2 H); [14] 164.9; [15] 130.8; [16] 199.2; [17] 34.3, 2.45 (t, J = 7.2 Hz, 2 H); [18] 37.4, 1.80 (t, J = 6.9 Hz, 2 H); [19] 36.4; [20] 27.0, 1.16 (s, Me); [21] 27.0, 1.16 (s, Me); [22] 11.6, 1.77 (s, Me); [23] 16.1, 1.61 (s, Me); [24] 16.2, 1.66 (s, Me); [25] 174.5. FABMS (positive ion) m/z (%): 407 [$C_{25}H_{36}O_3$ + Na (44)], 385 [$C_{25}H_{36}O_3$ + H (100)], 233 [$C_{15}H_{21}O_2$ (15)], 152 [$C_{10}H_{16}O$ (90)], 137 [$C_9H_{13}O$ (95)]. HREIMS m/z (%): 384.2653 [$C_{25}H_{36}O_3$ (10), Δ 0.2 mmu of calcd]; 369.2416 [$C_{24}H_{33}O_3$ (1), Δ 0.5 mmu of calcd]; 233.1537 [$C_{15}H_{21}O_2$ (1), Δ 0.1 mmu of calcd]; 152.1190 [$C_{10}H_{16}O$ (100), Δ 0.7 mmu of calcd]; 137.0969 [$C_9H_{13}O$ (8), Δ 0.6 mmu of calcd]. LREIMS m/z (%): 384 [M^+ (10)], 233 [$C_{15}H_{21}O_2$ (18)], 152 [$C_{10}H_{16}O$ (100)], 137 [$C_9H_{13}O$ (95)]. LRCIMS (isobutane) m/z (%): 385 [M^+ + H (100)].

Dehydroluffariellolide Diacid (2). Oil. IR (neat): 2930, 1716, 1646, 1456, 1381, 1194 cm^{-1}. UV (MeOH) λ_{max} (ε) 314 (838), 256 (2010) nm. NMR ($CDCl_3$) shifts in ppm from Me_4Si with assignments based on assessing the number of attached protons and the COSY data [atom number], ^{13}C δ's at 75 MHz and ^{1}H δ's at 300 MHz [1] 166.0; [2] 128.8, 6.58 (t, J = 1.2 Hz, 1 H); [3] 153.3; [4] 39.7, 2.56 (t, J = 6.9 Hz, 2 H); [5] 28.0, 2.34 (q, J = 7.2 Hz, 2 H); [6] 123.2, 5.10 (m, 1 H); [7] 136.5; [8] 40.4, 2.00 (m, 2 H); [9] 26.2, 2.05 (m, 2 H); [10] 121.2, 5.10 (m, 1 H); [11] 136.5; [12] 39.9, 2.00 (m, 2 H); [13] 25.3, 2.05 (m, 2 H); [14] 138.4; [15] 127.0; [16] 32.8, 1.89 (t, J = 6.0 Hz, 2 H); [17] 19.6, 1.59 (m, 2 H); [18] 39.7, 1.40 (m, 2 H); [19] 35.0; [20] 28.7, 0.98 (s, Me); [21] 28.7, 0.98 (s, Me); [22] 19.8, 1.59 (s, Me); [23] 16.3, 1.63 (s, Me); [24] 16.1, 1.61 (s, Me); [25] 166.0. (CD_3OD) [1] 170.0; [2] 129.6, 6.11 (s); [3] 148.8; [4] 36.5, 2.36 (t, J = 7.3 Hz); [5] 27.3, 2.17 (q, J =

(26) (a) Kobayashi, J.; Harbour, G. C.; Gilmore, J.; Rinehart, K. L. *J. Am. Chem. Soc.* **1984**, *106*, 1526. (b) Nakamura, H.; Deng, S.; Kobayashi, J.; Ohizumi, Y.; Tomotake, Y.; Matsuzaki, T. *Tetrahedron Lett.* **1987**, *28*, 621.

(27) The closest examples of such organic salts include guanidinium sulfates such as suvanine [Manes, L. V.; Crews, P.; Kernan, M. R.; Faulkner, D. J.; Fronczek, F. R.; Gandour, R. D. *J. Org. Chem.* **1988**, *53*, 570] and sulfircin [Wright, A. E.; McCarthy, P. J.; Schulte, G. K. *J. Org. Chem.* **1989**, *54*, 3472].

(28) The manzamines (keramamines) are the only β-carbolines known from sponges see: (a) Reference 26b. (b) Ichiba, T.; Sakai, R.; Kohmoto, S.; Saucy, G.; Higa, T. *Tetrahedron Lett.* **1988**, *29*, 3083 and references within.

(29) For a review of the many marine alkaloids that appear to be of tryptophan origin see: Christophersen, C. In *The Alkaloids*; Brossi, A., Ed.; Academic Press: New York, 1985; Vol. 24, pp 39–51 and references within.

(30) The condensation oftryptamine with tryptophan or itself would respectively generate **6** or homofascaplysin B (**8**). Additionally, the observation of octapamine **4** from the crude extracts suggest a possibility in which both tryptophan and octopamine might be involved in the biogenesis of **5** by analogy to this type of genesis that can be envisioned for clionamide [Andersen, R. *J. Tetrahedron Lett.* **1978**, 2541; Andersen, R. J.; Stonard, R. J. *Can. J. Chem.* **1979**, *57*, 2325].

(31) The alternative known biogenesis of β-carbolines via union of a tryptamine and an aldehyde precursor does not easily rationalize the additional *N*-aryl substituent in **9**. For a review of β-carboline biosynthesis see: *Biosynthesis of indole alkaloids*; Atta-Ur-Rahman, Basha, A., Eds.; Clarendon Press: Oxford, 1983; p 1–5 and 153–159.

(32) For reviews see: (a) Shoolery, J. N. *J. Nat. Prod.* **1984**, *47*, 226. (b) Benn, R.; Günther, H. *Angew. Chem.* **1983**, *22*, 350. (c) Kessler, H.; Gehrke, M.; Griesinger, C. *Angew. Chem., Int. Ed. Engl.* **1988**, *27*, 490.

7.5 Hz); [6] 123.0, 5.12 (dd, J = 12.6, 6.3 Hz); [7] 136.0; [8] 40.2, 2.06 (m); [9] 26.3, 2.02 (m); [10] 123.5, 5.12 (dd, J = 12.6, 6.3 Hz); [11] 135.6; [12] 39.7, 2.06 (m); [13] 26.4, 2.02 (m); [14] 136.9; [15] 126.7; [16] 32.4, 1.89 (t, J = 6.3 Hz); [17] 19.3, 1.58 (m); [18] 39.5, 1.40 (m); [19] 34.6; [20] 27.8, 0.97 (s); [21] 27.8, 0.97 (s); [22] 18.8, 1.62 (s, Me); [23] 14.8, 1.58 (s, Me); [24] 14.8, 1.58 (s, Me); [25] 166.0. FABMS (negative ion) (MF = $C_{25}H_{38}O_4$) m/z (%): 401 [$C_{25}H_{38}O_4$ - H (100)], 197 [$C_{10}H_{13}O_4$ (20)], 151 [$C_{11}H_{19}$ (22)], 128 [$C_5H_5O_4$ - H (45)]. LREIMS: 384 [$C_{25}H_{38}O_4$ - H_2O (30)], 137 [$C_{10}H_{17}$ (100)]. LRCIMS (isobutane): 385 [$C_{25}H_{38}O_4$ - H_2O + H (100)].

Fascaplysin A (Fascaplysin Cation/Dehydroluffariellolide Diacid Anion, 5b). A red oil. IR (neat): 2927, 1723, 1623, 1579, 1514, 1467, 1187, 1084, 755 cm^{-1}. UV (MeOH) λ_{max} (ϵ) 334 (5614), 296 (11228), 260 (14637) nm. NMR ($CDCl_3$) shifts in ppm from Me_4Si with assignments based on assessing the number of attached proton [atom number], ^{13}C δ's at 75 MHz and ^1H δ's 300 MHz: Anion 2^- [1] 169.8; [2] 131.4, 6.38 (s, 1 H); [3] 148.3; [4] 36.8, 2.51 (t, J = 6.9 Hz, 2 H); [5] 27.9, 2.29 (q, J = 7.2 Hz, 2 H); [6] 123.4, 5.22 (t, J = 6.9 Hz, 1 H); [7] 136.1; [8] 40.4, 2.00 (m, 2 H); [9] 28.0, 2.05 (m, 2 H); [10] 123.7, 5.12 (t, J = 6.9 Hz, 1 H); [11] 136.1; [12] 39.9, 2.00 (m, 2 H); [13] 26.9, 2.05 (m, 2 H); [14] 137.2; [15] 126.9; [16] 32.8, 1.88 (t, J = 6.3 Hz, 2 H); [17] 19.6, 1.57 (m, 2 H); [18] 39.9, 1.39 (m, 2 H); [19] 35.0; [20] 28.7, 0.97 (s, Me); [21] 28.7, 0.97 (s, Me); [22] 19.9, 1.58 (s, Me); [23] 16.3, 1.62 (s, Me); [24] 16.1, 1.63 (s, Me); [25] 170.0. Cation **5b** 9.25 (br s, 1 H), 8.50 (br s, 1 H), 8.20 (m, 1 H), 7.81 (m, 2 H), 7.64 (m, 3 H), 7.52 (m, 2 H). FABMS (negative ion): 401 [$C_{25}H_{37}O_4$ (100)], 197 [$C_{10}H_{13}O_4$ (10)], 128 [$C_5H_5O_4$ (9)]. FABMS (positive): 447 [$C_{25}H_{37}O_4Na_2$ (8)], 425 [$C_{25}H_{37}O_4Na$ + H^+ (25)], 271 [$C_{18}H_{11}N_2O$ (100)], 137 [$C_{10}H_{17}$ (27)]. HREIMS: 384.2665 [$C_{25}H_{36}O_3$, Δ 1.0 mmu of calcd]. LREIMS: 385 [$C_{25}H_{36}O_3$ - H_2O + 2H (5)], 271 [$C_{18}H_{11}N_2O$ (10)], 205 [$C_{15}H_{25}$ (5)], 137 [$C_{10}H_{17}$ (100)]. LRCIMS (isobutane): 385 [$C_{25}H_{36}O_3$ - H_2O + 2H (30)], 272 [$C_{18}H_{11}N_2O$ + H^+ (100)], 205 [$C_{15}H_{25}$ (30)], 137 $C_{10}H_{17}$ (100)].

Homofascaplysin A Cation/Dehydroluffariellolide Diacid Anion (6). A red viscous oil, $[\alpha]^{20}_D$ = -9.36° (c = 6.4 × 10^{-3}, MeOH). IR (neat): 3071, 2927, 1714, 1644, 1625, 1557, 1382, 868, 505 cm^{-1}. UV (MeOH) λ_{max} (ϵ) 334 (5250), 264 (8100), 220 (11200), 202 (11283) nm. NMR ($CDCl_3$) shifts in ppm from Me_4Si, ^{13}C δ's at 75 MHz and ^1H δ's at 300 MHz and ^1H NMR assignments are based on the ^1H-^1H COSY NMR data. Cation: ABCD system 8.18 (d, J = 8.1 Hz, 1 H), 7.40 (t, J = 7.5 Hz, 1 H), 7.73 (t, J = 7.5 Hz, 1 H), 7.92 (d, J = 8.4 Hz, 1 H); AB system 8.60 (d, J = 6.3 Hz, 1 H), 8.38 (d, J = 6.3 Hz, 1 H); ABCD system 7.78 (d, J = 7.2 Hz, 1 H), 7.60 (t, J = 7.0 Hz, 1 H), 7.58 (t, J = 6.9 Hz, 1 H), 7.73 (d, J = 7.5 Hz, 1 H); AB system 4.71 (d, J = 18.6 Hz, 1 H), 3.98 (d, J = 18.3 Hz, 1 H); 1.94 (s, Me), N-H [14.1 (bs, 1H)]. ^{13}C NMR: 204.6 (s), 144.4 (s), 140.7 (s), 136.3 (s), 135.0 (s), 133.1 (d), 130.8 (d), 124.9 (s), 124.5 (d), 124.3 (d), 123.7 (d), 123.2 (d), 122.5 (d), 120.9 (s, 2×), 119.8 (s), 116.4 (d), 114.4 (d), 112.9 (d), 78.2 (s), 51.0 (t), 30.5 (q). Anion: Same as the anion of the compound **5b**. HRFABMS (positive ion) m/z (%): 329.1295 [$C_{21}H_{17}N_2O_2$ (50), Δ 0.9 mmu of calcd]; 287 [$C_{21}H_{17}N_2O_2$ - 42 + H (10)]; 271.0897 [$C_{18}H_{11}N_2O$ (100), Δ 2.5 mmu of calcd]. HREIMS m/z: 312.1265 [$C_{21}H_{16}N_2O$, Δ 2.8 mmu of calcd]. LRCIMS m/z (%): 385 [$C_{25}H_{37}O_3$ (30)], 329 [$C_{21}H_{17}N_2O_2$ (8)], 313 [$C_{21}H_{17}N_2O_2$ - OH + H (20)]; 287 [$C_{21}H_{17}N_2O_2$ - 42 + H (8)]; 272 [$C_{18}H_{11}N_2O$ (80)]. LREIMS m/z (%): 384 [$C_{25}H_{36}O_3$ (4)], 328 [$C_{21}H_{17}N_2O_2$ - H (4)]; 312 [$C_{21}H_{16}N_2O$ (20)]; 286 [$C_{21}H_{17}N_2O_2$ - 42 (7)]; 271 [$C_{18}H_{11}N_2O$ (100)].

Homofascaplysin C (7). A yellow oil. IR (neat): 2921, 1651, 1595, 1505, 1488, 1393, 1331, 1209, 737 cm^{-1}. UV (MeOH) λ_{max} (ϵ) 292 (2982), 266 (7810) nm. NMR ($CDCl_3$) shifts in ppm from Me_4Si and ^1H δ's at 300 MHz. Assignments of the proton systems are based on the ^1H-^1H COSY NMR data and arguments in the text. ABCD system 8.14 (1 H, d, J = 8.1 Hz, H-1), 7.54 (1 H, t, j = 7.8 Hz, H-2), 7.34 (1 H, t, J = 7.8 Hz, H-3), 8.07 (1 H, d, J = 8.1 Hz, H-4); AB system 8.30 (1 H, d, J = 6.9 Hz, H-6) and 7.67 (1 H, d, J = 7.2 Hz, H-7); ABCD system: 7.94 (1 H, d, J = 8.4 Hz, H-8), 7.43 (1 H, t, J = 7.9 Hz, H-9), 7.54 (1 H, t, J = 7.8 Hz, H-10), 7.72 (1 H, d, J = 8.1 Hz, H-11); 12.2 (NH, bs); 10.3 (COH, s). ^{13}C δ's at 125 MHz [groups can be switched noted by a or b]: 181.0 (d, C-14), 138.1 (s, C-11a), 132.5[a] (s, C-12a), 132.2[a] (s, C-4a), 132.1[a] (s, C-13a), 129.5 (s, C-7a), 126.9 (d, C-6), 125.6 (d, C-10), 122.7 (d, C-1), 122.2 (s, C-7b), 120.6 (d, C-8), 120.3 (d, C-9), 118.6 (s, C-13), 117.7[b] (d, C-2), 116.7[b] (s, C-3), 112.7 (d, C-11), 111.2 (d, C-4), 108.0 (d, C-7), 105.5 (s, C-12b). HREIMS m/z (%): 284.0940 [$C_{19}H_{12}N_2O$ (94), Δ 0.7 mmu of calcd]; 255.0931 [$C_{18}H_{11}N_2$ (44), Δ 1.1 mmu of calcd]. LREIMS m/z (%): 284 [M^+ (100)], 255 (28). LRCIMS m/z (%): 285 [M^+ + H (100)].

Homofascaplysin B (8). A red oil. IR (neat): 1568, 1520, 1379, 1278, 1027 cm^{-1}. UV (MeOH) λ_{max} (ϵ) 330 (7692), 294 (5983), 268 (12141) nm. NMR ($CDCl_3$) shifts in ppm from Me_4Si and ^1H δ's at 300 MHz. Assignments of the proton systems are based on the ^1H-^1H COSY NMR data and arguments in the text. ABCD system 8.11 (1 H, d, J = 8.1 Hz, H-1), 7.36 (1 H, t, J = 7.4 Hz, H-2), 7.57 (1 H, t, J = 7.6 Hz, H-3), 7.70 (1 H, d, J = 8.4 Hz, H-4); AB system 8.39 (1 H, d, J = 6.9 Hz, H-6) and 7.79 (1 H, d, J = 7.2 Hz, H-7); ABCD system 7.95 (1 H, d, J = 8.1 Hz, H-8), 7.44 (1 H, dd, J = 7.5, 1.2 Hz, H-9), 7.51 (1 H, t, J = 7.2 Hz, H-10), 7.75 (1 H, d, J = 8.1 Hz, H-11); 12.51 (NH, bs); 4.12 (OMe, s). ^{13}C δ's at 125 MHz [groups can be switched noted by a or b]: 177.9 (s, C-14), 167.5 (s, C-15), 138.3 (s, C-11a), 134.8[a] (s, C-4a), 133.2[a] (s, C-12a), 129.3[a] (s, C-13a), 127.5 (d, C-6), 127.1 (s, C-7a), 126.2 (d, C-10), 122.9 (d, C-1), 121.8 (s, C-7b), 120.7 (d, C-8), 120.4 (d, C-9), 120.3 (s, C-13), 119.6[b] (d, C-2), 117.0[b] (d, C-3), 112.7 (d, C-11), 111.2 (d, C-4), 109.0 (d, C-7), 101.8 (s, C-12b), 52.9 (q, OMe). HREIMS 342.1002 [$C_{21}H_{14}N_2O_3$ (43), Δ 0.3 mmu of calcd]; 283.0871 [$C_{19}H_{11}N_2O$; M^+ - $C_2H_3O_2$ (100), Δ 0.2 mmu of calcd]; 255.0925 [$C_{19}H_{11}N_2O$ - CO (54), Δ 0.5 mmu of calcd].

Secofascaplysin A (9). A red oil. IR (neat): 3212, 2928, 1726 (s), 1656 (s), 1590, 1557, 1453, 1303 cm^{-1}. UV (MeOH) λ_{max} (ϵ) 350 (shoulder, 3000), 334 (3600), 296 (4775), 286 (5325), 238 (22525) nm. ^1H NMR ($CDCl_3$) shifts in ppm from Me_4Si and ^1H δ's at 300 MHz. Assignments are based on arguments in text. ABCD system 8.15 (dd, J = 8.1, 1.2 Hz, H-1), 7.59 (ddd, J = 0.9, 8.1, 7.8 Hz, H-2), 7.72 (ddd, J = 1.2, 8.1, 7.8 Hz, H-3), 7.46 (m, H-4); AB system 7.12 (d, J = 6.9 Hz, H-6) and 7.06 (d, J = 7.2 Hz, H-7); ABCD system: 7.97 (d, J = 8.1 Hz, H-8), 7.24 (dt, J = 2.1, 7.9 Hz, H-9), 7.46 (m, H-10), 7.46 (m, H-11); 10.38 (NH, bs); 3.60 (OMe, s). ^1H NMR (DMSO-d_6) shifts in ppm from Me_4Si and ^1H δ's at 500 MHz. ABCD system 8.08 (d, J = 8.0 Hz, H-1), 7.21 (dt, J = 1.0, 8.0 Hz, H-2), 7.43 (dt, J = 1.0, 8.0 Hz, H-3), 7.51 (d, J = 8.0 Hz, H-4); AB system 7.31 (d, J = 7.0 Hz, H-6) and 7.13 (d, J = 7.0 Hz, H-7); ABCD system 7.96 (d, J = 8.0 Hz, H-8), 7.79 (dt, J = 2.0, 8.0 Hz, H-9), 7.62 (dt, J = 2.0, 8.0 Hz, H-10), 7.53 (d, J = 8.0 Hz, H-11); 12.02 (NH, bs); 3.55 (OMe, s). ^{13}C NMR ($CDCl_3$) δ's at 125 MHz [groups can be switched noted by a or b]: 165.5 (s, C-13), 156.0 (s, C-12b), 140.8[a] (s, C-4a), 139.7[a] (s, C-11a), 133.7 (d, $^1J_{C-H}$ = 164 Hz, $^3J_{C-H}$ = 7 Hz, C-3), 131.6 (d, $^1J_{C-H}$ = 164 Hz, $^3J_{C-H}$ = 7 Hz, C-1), 129.7 (d, $^1J_{C-H}$ = 163 Hz, $^3J_{C-H}$ = 7 Hz, C-4), 128.9 (d, $^1J_{C-H}$ + 164 Hz, $^3J_{C-H}$ = 7 Hz, C-2), 128.3 (d, $^1J_{C-H}$ = 178 Hz, $^2J_{C-H}$ = 3 Hz, C-6), 128.7[b] (s, C-12b), 127.8[b] (s, C-7a), 127.0 (d, $^1J_{C-H}$ = 158 Hz, $^3J_{C-H}$ = 5 Hz, C-10), 125.1 (s, C-13a), 122.5 (s, C-7b), 121.2 (d, $^1J_{C-H}$ = 159 Hz, $^3J_{C-H}$ = 5 Hz, C-8), 120.3 (d, $^1J_{C-H}$ = 159 Hz, $^3J_{C-H}$ = 5 Hz, C-9), 112.7 (d, $^1J_{C-H}$ = 163 Hz, $^3J_{C-H}$ = 5 Hz, C-11), 101.4 (d, $^1J_{C-H}$ = 169 Hz, $^2J_{C-H}$ = 3 Hz, C-7), 52.3 (q, OMe). HREIMS m/z (%): 318.1007 [$C_{19}H_{14}N_2O_3$ (58), Δ 0.03 mmu of calcd]; 287.0793 [$C_{18}H_{11}N_2O_2$ (7), Δ 2.8 mmu of calcd], 259.0861 [$C_{17}H_{11}N_2O$ (100), Δ 1.1 mmu of calcd]; LREIMS m/z (%): 318 [M^+ (8)], 259 (6). LRCIMS m/z (%): 319 [M^+ + H (75), 259 (100)].

Treatment of 5b with HCl To Give 5a and 2. Compound **5b** (6.5 mg, 0.01 mmol) was dissolved in CH_2Cl_2 and extracted (3 × 5 mL) with 1 N HCl. The combined aqueous layers were concentrated to dryness in vacuo to afford 2 mg (0.007 mmol; 31% yield) of fascaplysin (**5a**). The organic layer was concentrated to give 3 mg (0.007 mmol; 46% yield) of the compound **2** with properties as described above.

Treatment of 5b by Anion Exchange Resin (Dowex-1) To Give 5a. Compound **5b** (10 mg, 0.015 mmol) was loaded on a basic anion exchange resin (Dowex-1, chloride form) and eluted first with MeOH followed by TFA. The TFA fraction was extracted with CH_2Cl_2 and yielded **5a** (3 mg, 0.011 mmol; 30% yield) in the organic layer.

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Supplementary Material Available: ^{13}C NMR spectra (^1H broad-band decoupled) of new compounds **1**, **2**, **5b**, **6**, **7**, **8**, and **9** (8 pages). Ordering information is given on any current masthead page.

Nucleic Acid Related Compounds. 64. Synthesis of 2',3'-Diazido-2',3'-dideoxyadenosine and 2',3'-Diamino-2',3'-dideoxyadenosine from 9-(β-D-Arabinofuranosyl)adenine[1]

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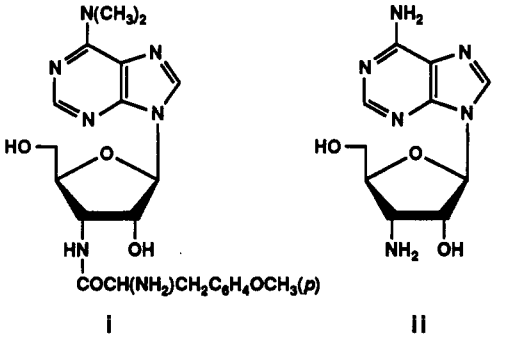
Treatment of 9-(β-D-arabinofuranosyl)adenine (**1**) with triphenylphosphine and diethyl azodicarboxylate gave 9-(2,3-anhydro-β-D-lyxofuranosyl)adenine (**2**). Treatment of **2** with lithium azide and protection of the major product gave 9-[3-azido-5-*O*-(*tert*-butyldimethylsilyl)-3-deoxy-β-D-arabinofuranosyl]adenine (**4**). Trifluoromethanesulfonylation of **4** and treatment of the resulting triflate **5** with lithium azide gave 9-(5-*O*-TBDMS-2,3-diazido-2,3-dideoxy-β-D-ribofuranosyl)adenine (**6**). Deprotection of **6** gave 2',3'-diazido-2',3'-dideoxyadenosine (**7**), which was hydrogenated to give the secondary diamino nucleoside analogue, 2',3'-diamino-2',3'-dideoxyadenosine (**8**). Biological rationale for the synthesis of nucleoside analogues **7** and **8** is discussed.

There has been a strong resurgence of interest recently in the chemistry of nucleosides.[4] Marked attention to the synthesis and properties of 2',3'-dideoxynucleosides and their sugar-substituted azido derivatives has been spurred by the efficacy of 3'-azido-3'-deoxythymidine (AZT) as a potent inhibitor of the human immunodeficiency virus (HIV) in the treatment of AIDS[5,6] and the parallel biological activity of several 2',3'-dideoxynucleosides.[6] At present, 2',3'-dideoxyadenosine,[7] 2',3'-dideoxycytidine,[8] and 2',3'-dideoxyinosine[9] are undergoing clinical trials in patients suffering from AIDS and AIDS-related complex.[10] It was recently noted that 2'-azido-2',3'-dideoxyadenosine has little inhibitory effect on HIV replication,[11] whereas 3'-azido-2',3'-dideoxyadenosine is active, but cytotoxic.[11,12]

Examples of 2'-amino-2'-deoxy- and 3'-amino-3'-deoxyribonucleosides are known to possess antibacterial, anticancer, and biosynthetic inhibitory activities.[13,14] Puromycin (i) is the well-known inhibitor of peptide biosynthesis.[13b,14b] Its core nucleoside component, 3'-amino-3'-deoxyadenosine (ii), has antitumor activity, and the 5'-triphosphate of ii has been observed to block RNA synthesis.[13c,14c] The 5'-triphosphate of 2'-amino-2'-deoxyadenosine and 2'-amino-2'-deoxyuridine are weak competitive inhibitors of DNA-dependent RNA polymerases from *E. coli*,[15] and both 2'-amino-2'-deoxy-



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