IR (film) 3450, 2920, 1660, 1465, 1380, 1160, 1100, 1050, 970, 960 cm^{-1}; HRMS: M^+, m/z obsd 304.2396, $C_{20}H_{32}O_2$ required 304.2402; low-resolution MS: m/z (relative intensity) 304 (6), 286 (47), 235 (39), 217 (9), 161 (100), 125 (40), 81 (44), 69 (18).

Asperketal B (2). The ketal **2** was obtained as a white solid after final purification by HPLC (Partisil 10 silica with 5% EtOAc in isooctane). The extract yielded 47 mg (0.14% of the crude extract) of **2**, mp 62–63 °C. Ketal **2** showed $[\alpha]^{20}_D$ +88° (*c* 0.65, C_6H_6) and exhibited the following features: IR (film) 2940, 1660, 1450, 1360, 1100, 1040, 990, 965 cm^{-1}; HRMS: M^+, m/z obsd 302.2237, $C_{20}H_{30}O_2$ required 302.2245; low-resolution MS: m/z (relative intensity) 302 (55), 273 (100), 163 (53), 161 (18), 151 (44), 109 (64), 81 (23).

Asperketal C (3). The ketal **3** was isolated as a white solid after final purification by HPLC (Partisil 10 silica with 5% EtOAc in isooctane). The extract yielded 23 mg (0.06% of the crude extract) of **3**, mp 72–73 °C. Asperketal C showed $[\alpha]^{20}_D$ +190° (*c* 1.88, C_6H_6) and exhibited the following spectral features: IR (film) 2920, 1660, 1460, 1370, 1080, 1050, 940, 920, 890, 840 cm^{-1}; HRMS: M^+, m/z obsd 318.2562, $C_{21}H_{34}O_2$ required 318.2558; low-resolution MS: m/z (relative intensity) 318 (1), 287 (6), 249 (50), 217 (14), 161 (100), 81 (89), 69 (28).

Asperketal D (4). The ketal **4** was obtained as a white solid after final purification by HPLC (Partisil 10 silica with 5% EtOAc in isooctane). The extract yielded 10 mg (0.03% of the crude extract) of **4**, mp 75–76 °C. Asperketal D exhibited $[\alpha]^{20}_D$ +126° (*c* 0.60, C_6H_6) and displayed the following spectral features: IR (film) 2940, 1660, 1450, 1360, 1140, 1010, 970, 880 cm^{-1}; HRMS: M^+, m/z obsd 304.2388, $C_{20}H_{32}O_2$ required 304.2402; low-resolution MS: m/z (relative intensity) 304 (44), 275 (45), 165 (46), 161 (69), 153 (59), 126 (76), 81 (100), 69 (87).

Asperketal E (5). The ketal **5** was obtained as an oil after final purification by HPLC (Partisil 10 silica with 5% EtOAc in isooctane). The extract yielded 9 mg (0.03% of the crude extract) of **5**, which showed $[\alpha]^{20}_D$ +54° (*c* 0.57, C_6H_6) and displayed the following features: IR (film) 2940, 1665, 1450, 1360, 1140, 1030, 960, 880 cm^{-1}; HRMS: M^+, m/z obsd 304.2406, $C_{20}H_{32}O_2$ required 304.2402; low-resolution MS: m/z (relative intensity) 304 (100), 275 (63), 165 (27), 161 (67), 152 (23), 126 (43), 81 (47), 69 (37).

Asperketal F (6). The ketal **6** was isolated as a white solid after final purification by HPLC (Partisil 10 silica with 5% EtOAc in isooctane). The extract yielded 14 mg (0.04% of the crude extract) of **6**, mp 54–55 °C. Asperketal F showed $[\alpha]^{20}_D$ +71° (*c* 0.77, C_6H_6) and displayed the following spectral features: IR (film) 2940, 1640, 1455, 1360, 1010, 980, 905, 885 cm^{-1}; HRMS: M^+, m/z obsd 304.2394, $C_{20}H_{32}O_2$ required 304.2402; low-resolution MS: m/z (relative intensity) 304 (34), 275 (100), 165 (24), 161 (63), 153 (1), 134 (40), 126 (51), 81 (84), 69 (99).

Obscuronatin (7). Obscuronatin (**7**) was obtained as an oil after purification by HPLC (Partisil 10 silica with 5% EtOAc in isooctane). The extract yielded 9 mg (0.03% of the crude extract) of **7**. The ^1H NMR spectrum of **7** (in C_6D_6) showed peaks at 5.31 (1 H, dd, 15.7, 10.1 Hz), 5.25 (1 H, br t, 7.0), 5.04 (1 H, br d, 15.7), 4.92 (1 H, br d, 11.0), 2.62 (1 H, m), 2.27–1.85 (7 H, m), 1.69 (3 H, br s), 1.60 (3 H, br s), 1.55–1.10 (6 H, m), 1.54 (3 H, br s), 1.06 (3 H, s), 0.92 (3 H, d, 6.7) ppm, in full accord with published data.[7] ^{13}C NMR resonances were also within ±0.04 ppm of the reported data for this compound.[7]

Acknowledgment. This research is a result of generous financial support from the National Science Foundation under Grants CHE-83-15546 and CHE-86-20217. We are grateful for continued support from the Oceanography Division at NSF, which supported our utilization of the research vessel *Columbus Iselin*. We thank the administration of the Grenadine Islands for permission to do research in their territorial waters.

Fascaplysin, an Unusual Antimicrobial Pigment from the Marine Sponge *Fascaplysinopsis* sp.

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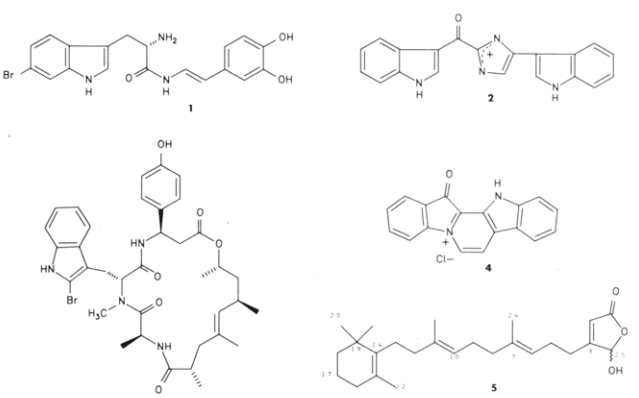
The structure of fascaplysin (**4**), a novel nitrogenous pigment exhibiting antimicrobial and cytotoxic properties, has been determined by spectral and X-ray analyses. The known compound luffariellolide (**5**) was also isolated.

A variety of biologically active metabolites containing an indole ring have been identified from marine sponges.[2,3] Among these are the tryptophan/tryptamine derivatives clionamide (**1**),[4,5] topsentin-A (**2**),[6] and jaspamide (**3**).[7] We now wish to report the isolation and structure determination of fascaplysin (**4**), a novel pentacyclic quaternary salt from the Fijian sponge *Fascaplysinopsis* Bergquist sp.[8] Fascaplysin inhibits the growth of several microbes, including *Staphylococcus aureus* (15-mm zone at 0.1 μg/disk), *Escherichia coli* (8-mm zone at 5 μg/disk), *Candida albicans* (11-mm zone at 1 μg/disk), and *Saccharomyces*

Chart I



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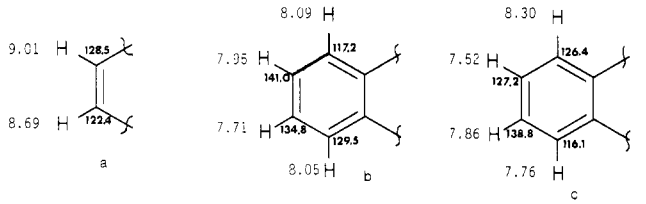
0022-3263/88/1953-3276$01.50/0

cerevisiae (20-mm zone at 0.1 μg/disk); it also exhibits an IC_{50} of 0.2 μg/mL against the murine leukemia L1210.

Fascaplysinopsis sp. was collected near Ndravuni, Fiji, and kept frozen until workup. Extraction of the lyophilized sponge with MeOH followed by Kupchan partitioning led to a mildly cytotoxic CCl_4 fraction and a $CHCl_3$ partition displaying more substantial cytotoxic and antimicrobial activity. Purification of the CCl_4 extract by column chromatography over Bio-Beads S-X12 (hexanes/CH_2Cl_2, 7:3) followed by HPLC on Partisil (isooctane/EtOAc, 7:3) led to the isolation of luffariellolide (**5**) as a colorless oil. This metabolite, which was recently described as an antiinflammatory constituent from the Palauan sponge *Luffariella variabilis*,[9] exhibited weak L1210 cytotoxicity (IC_{50} of 3.3 μg/mL) but did not display any antimicrobial activity.

The more polar $CHCl_3$ partition fraction was repeatedly chromatographed over Sephadex LH-20 using MeOH. Purification by reverse-phase HPLC (ODS-3, MeOH/H_2O/HOAc, 60:39.8:0.2) or crystallization from MeOH then yielded fascaplysin (**4**) as blood red prisms. Both the EI and FAB mass spectra of **4** afforded intense ions at m/z 271, indicating a formula of $C_{18}H_{11}N_2O$. This formula required 14.5 unsaturations and indicated that fascaplysin must be a salt.

The fully aromatic nature of **4** was revealed by the presence of ten hydrogens bound to sp^2 carbons (δ 7.5–9.0) and 17 carbons in the aromatic region (δ 116–150) of the NMR spectra. A signal at δ 186.4 in the ^{13}C NMR spectrum denoted a single carbonyl carbon, while a broad exchangeable signal at δ 11.5 (DMSO-d_6) suggested the presence of an N–H functionality. ^1H NMR decoupling experiments provided evidence for two ortho-disubstituted benzene rings and an isolated AB system. ^{13}C–^1H NMR correlation and long-range coupling experiments permitted definitive assignments of the protonated carbons as shown in partial structures a–c but did not allow further expansion of these pieces.



Since the structure of fascaplysin could not be unambiguously assigned, suitable crystals of **4** were submitted for X-ray diffraction analysis. The final X-ray model of compound **4** is illustrated in Figure 1. The entire molecule is planar within experimental error, and the interatomic distance and angles agree well with generally accepted values. The chloride anion makes a close contact of 2.1 Å with the NH hydrogen.

Compound **4** crystallized from $CHCl_3$ upon very slow evaporation as beautiful red crystals. Preliminary X-ray diffraction photographs showed monoclinic symmetry. Accurate lattice constants of a = 7.934 (1) Å, b = 10.961 (1) Å, c = 16.148 (2) Å, and β = 100.80 (2)° were determined from a least-squares fitting of 15 diffractometer-measured 2θ values. The space group was unambiguously $P2_1/n$ from systematic extinctions, and a density of 1.48 g/cm^3 indicated an asymmetric unit of $C_{18}H_{11}ClN_2O$.

All unique diffraction maxima with $2\theta < 114°$ were collected using graphite-monochromated Cu Kα radiation

(9) Albizati, K. F.; Holman, T.; Faulkner, D. J.; Glaser, K. B.; Jacobs, R. S. *Experientia* **1987**, *43*, 949–950. The spectral data observed for luffariellolide were essentially identical with published data.

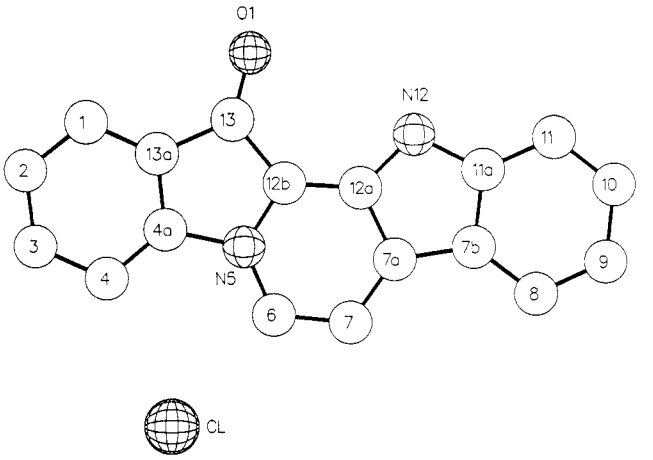
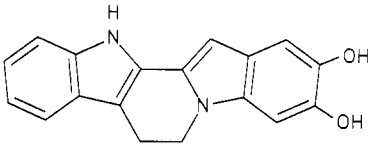


Figure 1. Computer-generated perspective drawing of the X-ray model of fascaplysin. Hydrogens are omitted for clarity.

(1.54178 Å) and variable-speed 1° ω scans. Of the 1852 reflections measured, 1490 (60%) were judged observed ($F_o > 3\sigma(F_o)$) after correction for Lorentz, polarization, and background effects.[10]

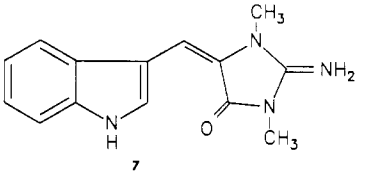
Finding a phasing model was uneventful. Block diagonal least-squares refinements with anisotropic heavy atoms and fixed hydrogens have converged to a standard crystallographic residual of 0.051. Additional crystallographic details are available in the Supplementary Material described at the end of this paper.

Fascaplysin represents the first naturally occurring member of the pentacyclic ring system 12*H*-pyrido[1,2-*a*:3,4-*b*']diindole whose closest analogue is the synthetic benzindole–pyrrocoline **6**.[11]



6

The sponge *Fascaplysinopsis reticulata* is well-known for its production of the tryptophan-derived metabolite aplysinopsin (**7**)[12,13] but this is the first report of a bisindole derivative from this genus.



7

Experimental Section

Two-dimensional ^{13}C–^1H NMR experiments were obtained on an IBM NR/200 FTNMR spectrometer in trifluoracetic acid-*d*. The pulse sequences used for COSY, HCCORR, RELAY, and

(10) All crystallographic calculations were done on a PRIME 9955 computer operated by the Cornell Chemistry Computing Facility. Principal programs employed were as follows: FOBS86, by G. Van Duyne, Cornell University, 1986; MULTAN 80 and RANTAN 80, written by P. Main, S. E. Hull, L. Lessinger, G. Germain, J. P. Declercq, and M. M. Woolfson, University of York, England, 1980; BLS78A, an anisotropic block diagonal least-squares refinement written by K. Hirotsu and E. Arnold, Cornell University, 1980; PLIPLOT, by G. Van Duyne, Cornell University, 1984; TABLES, by G. Van Duyne, Cornell University, 1986.

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COLOC experiments were those furnished in the IBM software manual. $^{13}C-^{1}H$ long-range couplings were set for maximum sensitivity at J = 4, 7, and 10 Hz.

Isolation. The dark brown sponge *Fascaplysinopsis* sp. was collected by SCUBA at Ndravuni, Fiji, and frozen. The sponge was then lyophilized (135.7 g, dry weight) and extracted with MeOH. The MeOH residue (42 g) was partitioned successively with 10% aqueous MeOH/hexanes, 20% aqueous MeOH/CCl_4, and 40% aqueous MeOH/$CHCl_3$. The majority of luffariellolide (0.45 g, 0.33% dry weight) was concentrated in the CCl_4 extract (2.64 g), while fascaplysin was located in the $CHCl_3$ extract (7.06 g). The $CHCl_3$ fraction was repeatedly chromatographed over Sephadex LH-20 in MeOH to give **4** as a brick red glass that crystallized from either MeOH or $CHCl_3$ (2.08 g, 2% dry weight).

Fascaplysin (4): mp 232-235 °C dec; FTIR (neat) 1712, 1618, 1594, 1502, 1462, 1450, 1080, 1061 cm^{-1}; UV (MeOH) λ_{max} 413 (ϵ 3100), 333 (3560), 301 (6800), 274 (5450), 262 (6400), 214 (7000) nm; (MeOH + OH^-, irreversible over time) 453 (ϵ 2700), 427 (2975), 398 (2295), 332 (5465), 274 (5801), 237 (6480), 210 (30,374) nm; ^{1}H NMR (TFA-*d*) δ 9.01 (1 H, d, J = 5.93 Hz), 8.69 (1 H, d, J = 5.93 Hz), 8.30 (1 H, d, J = 7.25 Hz), 8.09 (1 H, d, J = 7.91 Hz), 8.05 (1 H, d, J = 7.25 Hz), 7.95 (1 H, dd, J = 7.25, 7.91 Hz), 7.86 (1 H, dd, J = 7.25, 7.91 Hz), 7.76 (1 H, d, J = 7.91 Hz), 7.71 (1 H, dd, J = 7.25, 7.25 Hz), 7.52 (1 H, dd, J = 7.25, 7.25 Hz); ^{1}H NMR (DMSO-d_6) δ 13.5 (1 H, br s, N-H); ^{13}C (TFA-*d*) δ 186.4 (s), 149.9 (s), 149.8 (s), 145.4 (s), 141.0 (d), 138.8 (d), 135.0 (s), 134.8 (d), 129.5 (d), 128.5 (d), 127.2 (d), 126.4 (d), 126.1 (s), 123.5 (s) 122.4 (d), 122.2 (s), 117.2 (d), 116.1 (d); HRFABMS m/z 271.08606, $C_{18}H_{11}N_2O$ requires 271.08716; m/z 121 (100%); FABMS (matrix of glycerol + 18-crown-6 ether) MH^+ 535.

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Registry No. **4**, 114719-57-2; **5**, 111214-45-0.

Supplementary Material Available: Tables of fractional coordinates, thermal parameters, interatomic distances, interatomic angles, and torsional angles for **4** (6 pages). Ordering information is given on any current masthead page.

Photoisomerization of Some *N*-Aryl α,β-Unsaturated Iminium Salts[1]

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A series of *N*- and *C*-diaryl-substituted α,β-unsaturated iminium salts have been prepared in which a systematic change in the substituents on the aromatic rings was made. The photochemically induced isomerizations of these salts were examined in trifluoroacetic acid, a medium in which thermally induced isomerizations of these cations were generally slow. Photoisomerization about both the C=C and C=N partial double bonds of these iminium salts was observed, leading eventually to the formation of a photostationary state consisting of four isomers. Apart from cations containing a nitro substituent, the ratios of the quantum efficiencies for C=C vs C=N isomerization remained essentially constant as substituents on the two aryl rings were changed, suggesting that the perpendicular excited singlet states involved in these reactions have "biradical" character. The reduction potentials of the iminium salts were measured. Isomerization about the C=N bond of one of the salts was shown to occur by a photoinitiated electron transfer from tris(2,2'-bipyridine)ruthenium(II) dichloride.

Irradiation of α,β-unsaturated iminium ions in solution induces photoisomerization about both double bonds of the molecules, the C=N^+ and C=C bonds.[2,3] These reactions have application to the chemistry of vision, wherein the retinylidene iminium ion isomerizes about a specific C=C bond on light absorption.[4] The mechanism of this reaction of the visual pigments and the factors that determine the observed regioselectivity are of great interest.

In our previous work we have described the photoisomerizations of several *N*-alkyl-, C_3-aryl-, or C_3-alkyl-substituted α,β-unsaturated iminium salts.[2,3] The substituents present on these iminium salts seem to influence the regioselectivity of the photoisomerizations. It is possible that these effects are caused by a change in electron distribution in the excited states of these salts and that the substituent present can determine the preference for isomerization about either the C=N or C=C bond.[5]

In the present work we have carried out a detailed investigation of the isomerizations of a series of *N*- and *C*-diaryl-substituted α,β-unsaturated iminium salts in which a systematic change in the electron demand of the groups on the aryl rings is made. The purpose of these studies was to probe the effect of charge-stabilizing groups on the regioselectivity of the photoisomerizations about the C=N^+ and C=C bonds.[6]

Results

The iminium salts selected for this work were the *E,E* isomers **1a-i** in which the substituents on the two phenyl rings were varied. These cations were all isolated as crystalline, air-stable perchlorate salts by previously described procedures.[6] Their characterization by ^{1}H NMR

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