135.10 (s), 150.82 (s), 157.42 (d), 159.66 (s), 170.07 (s), 197.38 (s). Anal. Calcd for $C_{15}H_{13}ClO_4$: C, 61.55; H, 4.48. Found: C, 61.27; H, 4.33.

X-ray Structure of 6. All X-ray data were collected on a Nicolet R3m/μ update of a $P2_1$ diffractometer with use of Mo Kα monochromated radiation. Crystal data: a = 4.577 (1) Å, b = 12.298 (2) Å, c = 24.253 (4) Å, space group $P2_1cn$, Z = 4, D(calcd) = 1.647 g cm^{-3}, and μ = 30.04 cm^{-1}. Emprical absorption correction was applied. The structure of **6** was refined by the block-cascade least-squares technique with hydrogen atoms allowed to ride at fixed distances from attached atoms. Refinement: R = 0.0410 for 184 parameters and 831 reflections, S = 1.05, $(\Delta/\sigma)_{max}$ = 0.012 with the largest residual peaks from a final difference map of −0.43 and +0.33 e Å^{-3}.

Acknowledgment. Financial support of this study by the Air Force Office of Scientific Research (Grant AFOSR-88-0132, A.P.M.), the Robert A. Welch Foundation (Grant B-963, A.P.M.; Grant P-074, W.H.W.), and the University of North Texas Faculty Research Committee (A.P.M.) is gratefully acknowledged.

Registry No. **4**, 116970-42-4; **5**, 116970-43-5; **6**, 116970-44-6; **7**, 116970-45-7; EDA, 623-73-4.

Supplementary Material Available: A structure drawing of **6** with thermal ellipsoids drawn at the 35% probability level and a list of atomic coordinates and isotropic thermal parameters, bond lengths, bond angles, anisotropic thermal parameters, and H-atom coordinates and isotropic thermal parameters for **6** (7 pages); a list of structure factors for **6** (7 pages). Ordering information is given on any current masthead page.

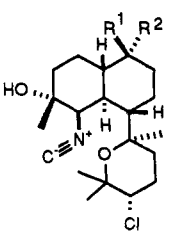
Polyfunctional Diterpene Isonitriles from Marine Sponge *Acanthella carvenosa*

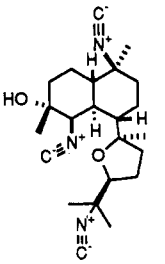
Siraj Omar, Carolyn Albert, Tashin Fanni, and Phillip Crews*

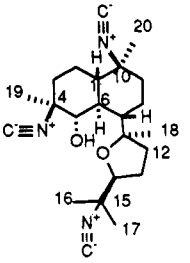
Department of Chemistry and Institute for Marine Sciences, University of California, Santa Cruz, California 95064

Received July 12, 1988

Orange sponges are conspicuous inhabitants of Fiji coral reefs. In 1984, our attention was drawn to an abundant, round orange Fijian sponge because its crude extracts were extremely active in an in vitro anthelmintic primary screen against parasitic stages of *Nippostrongylus brasiliensis*.[1] Bioassay guided isolation of the active constituents of this sponge, identified as *Acanthella carvenosa*,[2] commenced and involved both the original collection as well as large recollections made during two subsequent expeditions to Fiji in 1986 and 1987. While our work was in progress, Scheuer[3] reported structures of 11 richly functionalized diterpenoid antibiotics, the kalihinols, from Pacific collections of *Acanthella* sp. Structurally, the kalihinols fall into two main groups, the tetrahydropyran kalihinols, represented by kalihinol A (**1**),[3a] and the tetrahydrofuran kalihinols, represented by kalihinol F (**2**).[3b] Common to both groups is a *trans*-decalin skeleton bearing a hydroxyl function at carbon C-4; while multiple isocyano functions at C-5 and C-10 are found in seven cases. All five compounds of the first group have a chlorine atom at C-14 of the tetrahydropyranyl moiety, whereas in the second group the tetrahydrofuran moiety is functionalized at C-15 with NC, NCS, or Cl, or the *gem*-dimethyl is replaced by an isopropenyl. We now extend the structural breadth of this diterpenoid family by reporting the structure of a new tetrahydrofuran, isokalihinol F (**3**).







1 R_1=NC, R_2=Me
4 R_1=Me, R_2=NCS
5 R_1&R_2 = $=CH_2$
6 R_1=Me, R_2= NC

2 **3**

Results and Discussion

Aqueous methanol extracts of freshly collected sponge *A. carvenosa* yielded a dark viscous oil. The crude oil was then purified between aqueous methanol and the series: hexanes, CCl_4, and CH_2Cl_2. Purification proceeded on the dichloromethane partition fraction, which showed high in vitro anthelmintic activity. The workup of collection no. 86-8 illustrates our purification strategy. Its CH_2Cl_2 partition fraction was subjected to flash chromatography (silica gel) followed by repeated reversed-phase HPLC (10 μm ODS, 25 × 1.0 cm; 85% MeOH–15% H_2O), which afforded a major crystalline component, kalihinol A (**1**), whose spectral properties were identical with those reported by Scheuer.[3] Other minor components were obtained including known kalihinols F (**2**) and X (**4**) and an unknown compound, **3**. The latter crystallized as colorless long needles from diethyl ether (mp 180–182 °C; $[\alpha]^{20}_D$ +13.6°). Its molecular formula, $C_{23}H_{33}N_3O_2$ (calcd MW 383.257), was deduced from LRCIMS (isobutane) data, m/z 384 (M^+ + H) and an integrated ^{13}C NMR spectrum. The ^{13}C (C_6D_6) NMR spectrum of compound **3** showed a resemblance to that of kalihinol F (**2**). Most importantly, the resonances for **3** at 87.5 (s) (C-11) and 82.4 (d), (C-14) ppm were consistent with a tetrahydrofuran moiety, and broad triplets ($J \approx 5$ Hz) at 61.5, 59.8, and 59.6 ppm intimated that an isocyano group was attached to each of these carbons. A *trans*-decalin ring was assigned to **3** on biogenetic grounds and because of its similar ^{13}C NMR δ's with **2** at C-1, C-2, C-9, and C-20. However, in contrast to **2**, the three isocyano functions in **3** were assigned to carbons C-4, C-10, and C-15, while a secondary hydroxyl function, as a sharp ^{13}C NMR peak at 76.7 (d) ppm (whose methine ^1H was at δ 3.44), was assigned to carbon C-5. The specific evidence supporting the placement of the OH at C-5 and its assigned stereochemistry is as follows. The 2D ^1H–^1H homo and ^{13}C–^1H hetero COSY NMR spectra in C_6D_6 enabled all NMR resonances of **3** to be assigned. The ^1H–^1H COSY NMR spectrum showed strong correlations from ^1H NMR peak at 3.44 ppm (dd, J = 8.4, 3.3 Hz, H-5) to the peak at 0.95 ppm (m, H-6), and to the hydroxyl peak at 6.2 ppm (d, J = 3.3 Hz). A large coupling constant (J = 8.4 Hz) was observed between proton H-5 and axial H-6, which indicated that H-5 must also be axial. The remaining stereochemical features were next confirmed. The stereochemistry of the tetrahydrofuranyl ring substituents

(1) We thank Dr. Tom Matthews and his staff for this data according to the assay described by: Jenkins, D. C.; Armitage, R.; Carrington, T. S. *Z. Parasitenkd.* **1980**, *63*, 261.

(2) Our collections of *Acanthella carvenosa* (Fam. Axinellidae, Order Axinellida) were identified by C. Diaz (Harbor Branch Oceanographic Institution/SeaPharm Project, Fort Pierce, FL).

(3) (a) Chang, C. W. J.; Patra, A.; Roll, D. M.; Scheuer, P. J.; Matsumoto, G. M.; Clardy, J. *J. Am. Chem. Soc.* **1984**, *106*, 4644. (b) Patra, A.; Chang, C. W. J.; Scheuer, P. J.; van Duyne, G. D.; Matsumoto, G. M.; Clardy, J. *J. Am. Chem. Soc.* **1984**, *106*, 7981. (c) Chang, C. W. J.; Patra, A.; Baker, J. A.; Scheuer, P. J. *J. Am. Chem. Soc.* **1987**, *109*, 6119.

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in 3 was concluded to be analogous as in 2 because of their identical ^{13}C shifts at C-12 to C-18. Four out of five of the methyl ^1H NMR peaks in 3 appeared as broadened singlets due to coupling with the ^{14}N of the isocyano group (NC). The similar high-field ^{13}C (C_6D_6) shifts assigned to methyl carbons C-19 (19.9 (q)) and C-20 (20.3 (q)) indicated that both methyl groups were axial; whereas the equatorial methyl C-19 in kalihinol F (2) had a much higher value of 28.6 ppm. Given the methyl stereochemistry assigned in 3, both isocyano functions at carbons C-4 and C-10 must be equatorial. Finally, the multiplicities of axial protons, H_{ax}-2 and H_{ax}-8, respectively at 0.70 ppm (dddd, J = 13.6, 13.6, 12.4, 3.2 Hz) and 0.52 ppm (dddd, J = 13.8, 13.8, 12.8, 3.9 Hz), suggests that the *trans*-decalin skeleton of isokalihinol F (3) adopts a chair–chair conformation in solution. This is consistent with the chair–chair decalin conformation reported for crystalline kalihinol A (1),[3a] but not with that of the kalihinol F, which occupies a boat–boat conformation in the solid state.[3b]

Significant differences were observed in the kalihinol constituents of the two large collections of *A. carvenosa* obtained from Fiji (no. 86-8 and no. 87-34). As noted above, the 1986 collection (no. 86-8) yielded kalihinols A (1) (major component), F (2), X (4), and isokalihinol F (3). By contrast the 1987 collection (no. 87-34) yielded kalihinols Y (5) (major component), X (4), and Z (6). Anthelmintic (in vitro) screening conducted with pure compounds (at 50 μg/mL) against *N. brasiliensis* revealed that kalihinol Y (5) was extremely active, while kalihinols A (1), X (4), and Z (6) were very active, but isokalihinol F (3) was completely inactive. Attempts to interconvert the C-4 and C-5 substituents in 1 under acidic conditions showed that this compound was completely stable. Thus, the unusual arrangement of substituents in the A ring of 3 was not an artifact formed during workup. Biologically active diterpenoid isonitriles have also been found in *Amphimedon* sp.,[4] *Hymeniacidon amphilecta*,[5] and in *Halichondria* sp.[6] The variations in the kalihinol compositions that we and Scheuer have observed suggest that *Acanthella* is capable of rich isonitrile diterpenoid biosynthesis, and this is a subject under current investigation in our laboratory.

Experimental Section

NMR spectra were recorded on a JEOL FX-100 PFT spectrometer (99.5 MHz for ^1H and 25.0 MHz for ^{13}C) or on a GN-300 spectrometer (300 MHz for ^1H and 75 MHz for ^{13}C). High-field ^1H NMR spectra were also recorded on a Bruker 500 spectrometer (at Syntex Research Inc., Palo Alto) operating at 500 MHz. Multiplicities of ^{13}C NMR peaks were determined from APT data, and 2D COSY NMR experiments were done on the GN-300 instrument. Mass spectrometry data were obtained on a Finnigan 4000 (6000 LS7 computer system). High-performance liquid chromatography (HPLC) was done on a Waters liquid chromatograph with a Regis 10 μm ODS or 10 μm silica gel column (25 × 1.0 cm). All solvents were distilled and dried for HPLC and were spectral grade for spectroscopy. Rotations were measured on a Perkin-Elmer 141 polarimeter.

Two-Dimensional NMR Procedures. Standard pulse sequences were used for the homo COSY and the hetero COSY experiments.

Isolation Procedures. The sponge *A. carvenosa* was either extracted fresh or preserved for a short period before extraction. It was cut into small pieces and repeatedly extracted with aqueous MeOH. The extract was concentrated by distillation of the methanol under reduced pressure, and the remaining aqueous extract was solvent extracted with methylene chloride (CH_2Cl_2), which was then evaporated, yielding a dark viscous oil. The crude oil was then partitioned between aqueous MeOH and the solvent series of hexanes, CCl_4, CH_2Cl_2, and aqueous MeOH. Each partition fraction was assayed for bioactivities. The hexanes, the CCl_4, and the CH_2Cl_2 partition fractions were then separately chromatographed on flash column (Aldrich silica gel, grade 60,60A) and further purified on a normal-phase HPLC column (10 μm silical gel, 25 × 1.0 cm; hexanes/ethyl acetate solvent system) or on a reversed phase column (10 μm ODS, 25 × 1.0 cm; aqueous MeOH solvent system).

Isokalihinol F (3): colorless long needles; crystallized from diethyl ether, mp 180–182 °C, changed to brown liquid; $[\alpha]^{20}_D$ +13.6° (c 0.018, $CDCl_3$); molecular weight 383.257 calculated for $C_{23}H_{33}N_3O_2$; LREIMS, m/z (percent) 383 (trace, M^+), 357 (1, M^+ – NC), 330 (1, M^+ – NC – HCN), 303 (10, M^+ – NC – 2HCN), 285 (5, M^+ – NC – 2HCN – H_2O), 152 (38), 125 (10), 84 (100); LRCIMS (isobutane), m/z (percent) 384 (2, M^+ + H), 357 (30, M^+ – NC), 330 (45, M^+ – NC – HCN), 303 (100, M^+ – NC – 2HCN), 285 (19, M^+ – NC – 2HCN – H_2O), 152 (10); NMR shifts in ppm from Me_4Si, assignments based on assessing the number of attached protons and COSY data [[atom number] ^{13}C (C_6D_6) δ's at 75 MHz, ^1H (C_6D_6) δ's (multiplicites, J's (Hz), integration) at 500 MHz] [1] 47.0 (d), 0.90 (m, 1 H); [2] 21.4 (t), 1.63 (m, H_{eq}) and 0.70 (dddd, J = 13.6, 13.6, 12.4, 3.2, H_{ax}); [3] 37.0 (t), 1.50 (m, 2 H); [4] 61.5 (br t, distorted due to NC, $J \approx 5$ Hz); [5] 76.7 (d), 3.44 (dd, J = 8.4, 3.3, 1 H); [6] 43.0 (d), 0.95 (m, 1 H); [7] 53.8 (d), 1.20 (m, 1 H); [8] 26.3 (t), 1.02 (m, H_{eq}) and 0.52 (dddd, J = 13.8, 13.8, 12.8, 3.9, H_{ax}); [9] 40.6 (t), 1.52 (m, 2 H); [10] 59.8 (br t, distorted due to NC, $J \approx 5$ Hz); [11] 87.5 (s); [12] 39.0 (t), 1.18 and 1.48 (m, 2 H); [13] 25.0 (t), 1.50 (m, 2 H); [14] 82.4 (d), 3.07 (dd, J = 6.0, 2.2, 1 H); [15] 59.6 (br t, distorted due to NC, $J \approx 5$ Hz); [16] 25.4 (q), 1.12 (br s, 3 H); [17] 25.8 (q), 0.75 (br s, 3 H); [18] 18.2 (q), 0.87 (s, 3 H); [19] 19.9 (q), 1.27 (br s, 3 H); [20] 20.3 (q), 0.80 (br s, 3 H) (NC ^{13}C δ's at 158.2, 157.0, and 155.3 were weak signals).

Acknowledgment. Partial research support was from NOAA, National Sea Grant College Program, Department of Commerce, University of California project number R/MP-41. The U.S. Government is authorized to produce and distribute reprints for governmental purposes. Other grant support was from Syntex Inc. We thank Jim Loo (UCSC) and Dr. Mike Maddox (Syntex) for assistance with NMR measurements. We also are grateful to the Fiji government for their cooperation.

Registry No. 1, 91294-83-6; 2, 93426-91-6; 3, 117229-41-1; 4, 109979-17-1; 5, 109979-18-2; 6, 110043-50-0.

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Aromatization and Disproportionation of 1,3- and 1,4-Cyclohexadienes by Potassium 3-Aminopropylamide[1]

N. Venkatasubramanian and Samuel Siegel*

Department of Chemistry and Biochemistry, University of Arkansas, Fayetteville, Arkansas 72701

Received May 19, 1988

Recently, we reported the isomerization and aromatization of the 1,3-di-*tert*-butylcyclohexadienes in several strongly basic media including potassium 3-aminopropylamide (KAPA)[2] in 1,3-diaminopropane (DAP).[3]

(1) Presented in part at the 33rd Southwest/37th Southeast Regional meeting of the American Chemical Society, October 8–11, 1985, Memphis, TN. Support by Grant CHE-7826661 from the National Science Foundation is gratefully acknowledged.

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