

DITERPENOID ISOCYANIDES FROM THE MARINE SPONGE *HYMENIACIDON AMPHILECTA*

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A number of sesquiterpene isocyanides have been isolated from sponges of the Order Halichondrida.¹ The isolation of a diterpenoid diisocyanide 1 from a sponge of the unrelated genus *Adocia*² was unusual from both the chemical and chemotaxonomic viewpoint. In this paper we wish to report the structure of a second diterpenoid diisocyanide, 8,15-diisocyano-11(20)-amphilectene (2) from *Hymeniacidon amphilecta*, a sponge of the Order Halichondrida.

Ethanollic extracts of the Caribbean sponge *Hymeniacidon amphilecta*³ exhibited antibiotic activity against *S. aureus*, *B. subtilis* and *C. albicans*. Silica gel chromatography of the ether soluble material, guided by antibacterial testing, gave several active components. The major compound 2 (0.9% dry weight), crystallized from hexane, mp 105-106°C, and had the molecular formula C₂₂H₃₂N₂. The spectral data⁴ described a tricyclic diterpene having two tertiary isocyanide groups and an exocyclic methylene functionality. The ¹H NMR spectrum contained two unusual methyl signals at δ 1.43 and 1.45 which appeared as 1:1:1 triplets (J = 2Hz) which are typical of methyl groups on a carbon bearing isocyanide.⁵ Reduction of the diisocyanide 2 with lithium in liquid ammonia gave the basic hydrocarbon which had no tertiary methyl groups suggesting that diisocyanide 2 was a tricyclic compound related to diisocyanide 1. The structure of diisocyanide 2 was determined by X-ray diffraction analysis.

8,15-diisocyano-11(20)-amphilectene (2) crystallized in the monoclinic crystal class with

$a = 18.396(8)$, $b = 9.998(4)$, $c = 13.095(4)$ Å and $\beta = 123.90(2)^\circ$. Systematic extinctions and the known chirality indicated space group C2 and a calculated ($z = 4$) and observed density of $\sim 1.08\text{g/cc}$ implied one molecule of $\text{C}_{22}\text{H}_{32}\text{N}_2$ per asymmetric unit. All unique diffraction maxima were recorded on a four-circle diffractometer using graphite monochromated $\text{MoK}\alpha$ (0.71069 Å) radiation and a variable speed ω -scan technique. Of the 1877 reflections surveyed, 1532 (82%) were judged observed ($F_o \geq 3\sigma(F_o)$) after correction for Lorentz, polarization and background effects. An initial phasing model was found by a multiresolution weighted tangent formula approach.⁶ Hydrogens were located on a difference electron density synthesis and included in subsequent calculations.⁷ Full matrix least-squares refinements with anisotropic temperature factors for nonhydrogen atoms and isotropic temperature factors for hydrogens have converged to a conventional crystallographic residual of 0.047 for the observed data. No attempt to determine the absolute configuration was made.

Figure 1 is a computer-generated perspective drawing less hydrogens. The three cyclohexane rings are all in the chair conformation with the bridgehead substituents axially oriented. All of the other substituents are equatorially oriented except the exocyclic methylene for which no designation is needed. The two isocyanide groups have an antiparallel orientation. There are no intermolecular nonhydrogen atoms contacts less than 3.45 Å and a final difference map showed no unusually high electron density. See reference 8 for further structural information.

A second antimicrobial compound 3 (0.5% dry weight) was obtained as an oil⁹ having the molecular formula $\text{C}_{22}\text{H}_{34}\text{N}_2\text{O}$. The infrared spectrum of compound 3 contained signals at 3250 and 1685 cm^{-1} assigned to an amide functionality. The ^1H NMR spectrum contained two signals at δ 7.91 and 8.05 assigned to rotational isomers of a formamide group. Treatment of both the diisocyanide 2 and the formamide 3 with 98% acetic acid gave the same diformamide 4. In the ^1H NMR spectrum of formamide 3, the methyl signals at δ 1.35 and 1.39 did not show fine coupling, suggesting that the formamide group was at C-15. In the ^{13}C NMR spectrum of formamide 3, a 1:1:1 triplet ($J = 4\text{Hz}$) due to the carbon atom-bearing isocyanide was at δ 66.5. Similar signals in the ^{13}C NMR spectrum of diisocyanide 2 were at δ 66.9 and 56.5, suggesting that the isocyanide group was at C-8.

We propose that the new carbon skeleton be named amphilectane (5). Thus the diisocyanide

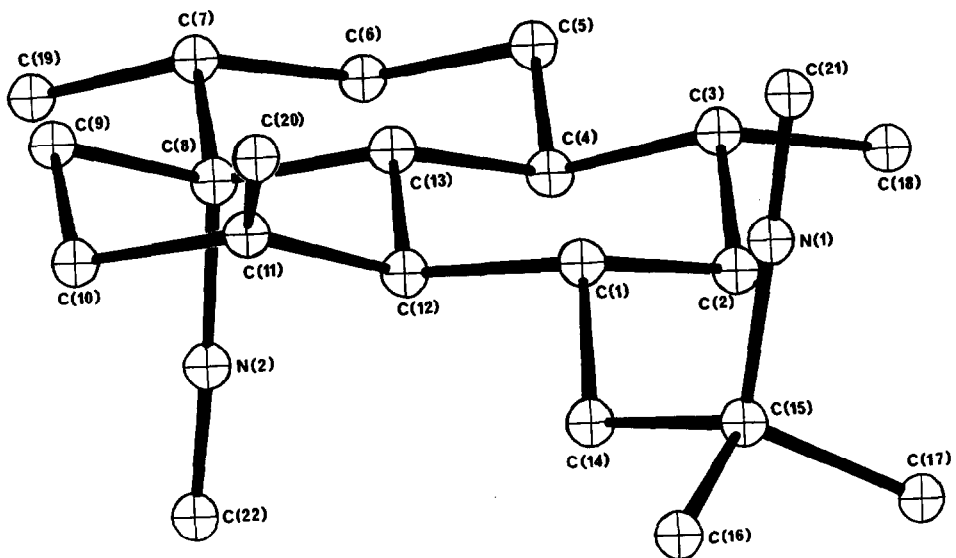
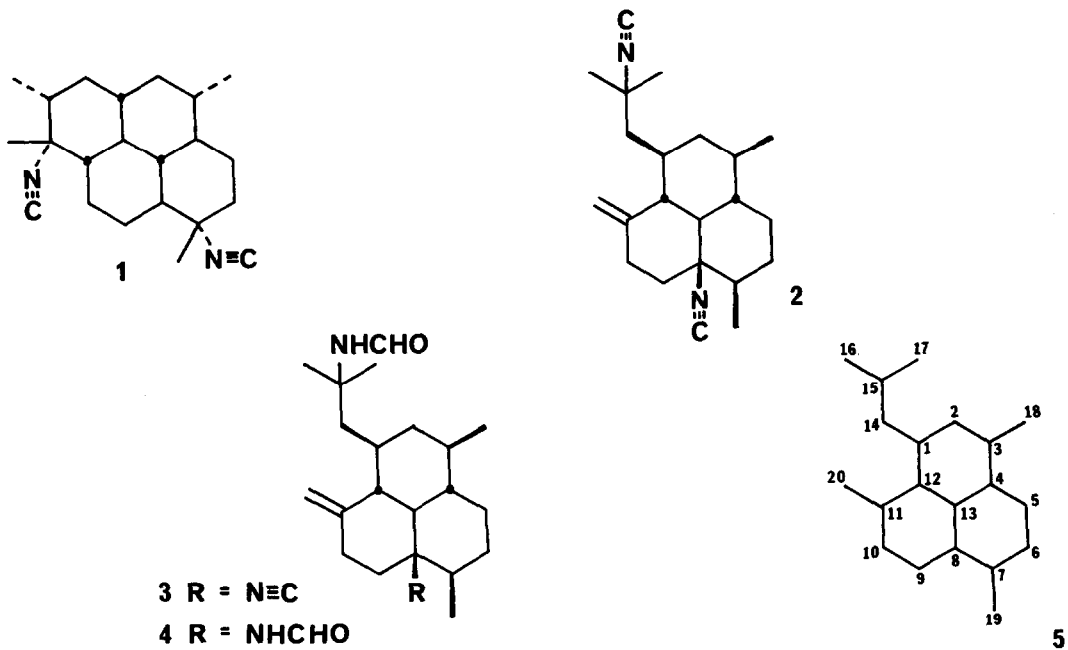


Figure. A computer generated drawing of 8,15-diisocyanato-11(20)-amphilectene(2). Hydrogens are omitted for clarity and no absolute configuration is implied.



2 is named 8,15-diisocyano-11(20)-amphilectene. The carbon skeleton can be derived by cyclization of the basic linear diterpene skeleton as was proposed for diisocyanoadociene (1).²

Both the diisocyanide 2 and the formamide 3 inhibited the growth of *S. aureus*, *B. subtilis* and *C. albicans* at 100 µg/disc. A recent examination of a second collection of this sponge suggests that there may be considerable variation in the diterpene isocyanide content of this sponge.

Acknowledgments

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References and Notes

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2. J. T. Baker, R. J. Wells, W. E. Oberh nsli, and G. B. Hawes, J. Amer. Chem. Soc. **98**, 4012 (1976).
3. Collected in July 1977 at Glover Reef, Belize, using SCUBA at 10-20 m depth.
4. $[\alpha]_D - 56^\circ$ (1.0, CHCl₃); ir (CCl₄) 2130, 1650 cm⁻¹; ¹H NMR (CCl₄) δ 4.84 (s, 1H), 4.66 (s, 1H), 1.45 (t, 3H, J = 2Hz), 1.43 (t, 3H, J = 2Hz), 0.99 (d, 3H, J = 7Hz), 0.97 (d, 3H, J = 7Hz); ¹³C NMR (d₆ benzene) 159.9 (t, J = 4Hz), 158.1 (t, J = 4Hz), 150.1(s), 106.6(t), 66.9 (t, J = 4Hz), 56.5 (t, J = 4Hz), 55.5(d), 46.3(d), 45.7(t), 42.9(d), 41.2(t), 40.9(d), 39.7(t), 35.6(d), 33.9(t), 33.5(t), 31.6(d), 30.4(t), 30.2(q), 30.0(q), 19.9(q), 15.9(q); HRMS, found 324.2553, C₂₂H₃₂N₂ requires 324.2565.
5. B. J. Burreson, C. Christophersen, and P. J. Scheuer, Tetrahedron **31**, 2015 (1975).
6. G. Germain, P. Main and M. M. Woolfson, Acta Crystallogr., **A27**, 368 (1971).
7. All crystallographic calculations were done on a Prime 400 computer operated by the Materials Science Center and the Department of Chemistry, Cornell University. The principal programs used were: REDUCE and UNIQUE, data reduction programs, M. E. Leonowicz, Cornell University, 1978; BLS, block diagonal least squares refinement, K. Hirotsu, Cornell University, 1978; ORFLS (modified), full matrix least squares, W. R. Busing, K. O. Martin and H. S. Levy, Oak Ridge, ORNL-TM-305; ORTEP, crystallographic illustration program, C. Johnson, Oak Ridge, ORNL-3794, BOND, structural parameters and errors, K. Hirotsu, Cornell University, 1978; MULTAN-76, direct methods and fast fourier transform, G. Germain, P. Main and M. Woolfson, University of York.
8. Tables of fractional coordinates and thermal parameters, bond distances, bond angles and observed and calculated structure factors are available from Jon Clardy, Department of Chemistry, Cornell University, Ithaca, NY 14853.
9. $[\alpha]_D - 24^\circ$ (1.0, CHCl₃); ir (neat) 3250, 2130, 1685 cm⁻¹; ¹H NMR (CCl₄) δ ~8.0 (1H,m) 4.80 (s, 1H), 4.63 (s, 0.3H), 4.56 (s, 0.7H), 1.39 (s, 3H), 1.35 (s, 3H), 1.00 (d, 3H, J = 7Hz), 0.92 (bd, 3H, J = 7Hz); ¹³C NMR 163.2, 160.1, 160.0(t, J = 4Hz), 150.4, 150.3, 106.5, 106.4, 66.5 (t, J = 4Hz), 55.7, 53.8, 52.6, 48.3, 46.7, 45.5, 42.8, 41.8, 41.4, 40.9, 39.9, 35.8, 33.9, 33.4, 32.9, 30.7, 30.1, 29.2, 28.9, 27.9, 24.9, 20.0, 15.8; HRMS, found 342.2684, C₂₂H₃₄N₂O requires 342.2671.

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