in steps up to a 1:1 ratio (total of 16 500-mL fractions). A portion of fraction 15 was subjected to HPLC using a C_{18} column and methanol–water (3:1) to give three pure compounds: **10**, 40 mg; **11**, 7 mg; and **12**, 70 mg. HPLC of portions of fractions 13 and 14 over C_{18} using methanol–water (7:3) afforded compounds **1**, **2**, and **10** (10, 8, and 4 mg, respectively).

Patellamide D (10): mp 144–145 °C (slow evaporation of methanol solution); IR 3380, 3325, 1660 (brd) cm^{-1}; $[\alpha]_D$ 32° (*c* 0.37, $CHCl_3$); HR EI MS 776.3099 $C_{38}H_{48}N_8O_6S_2$ (−5 ppm).

Lissoclinamide 5 (11): IR ($CHCl_3$) 3380, 3320, 3120, 1655, 1635 cm^{-1}; HR EI MS 739.2638 $C_{38}H_{41}N_7O_5S_2$ (3.7 ppm).

Lissoclinamide 4 (12): mp 152–154 °C (powder precipitated from ether); IR ($CHCl_3$) 3380, 3325, 1655, 1630 cm^{-1}; $[\alpha]_D$ 45° (*c* 0.7, $CHCl_3$); HR EI MS 741.2767 $C_{38}H_{43}N_7O_7S_2$ (15.2).

Lissoclinamide 6 (13): FT IR (film) 3380, 3325, 1677, 1640 cm^{-1}; HR EI MS 741.2818 $C_{38}H_{43}N_7O_5S_2$ (6.9 ppm).

X-ray Analysis of 10. Well-formed prismatic crystals of patellamide D were obtained by slow evaporation from a methanol solution. A crystal of dimensions 0.10 × 0.10 × 0.40 mm was selected for X-ray measurements. The cell parameters were determined from the least-square fit of $\pm 2\theta$ values of 48 reflections measured at 163 K using Cu $K\alpha_1$ radiation. Patellamide D crystallizes in the orthorhombic space group $P2_12_12_1$ with a = 13.976 (2), b = 24.360 (4), and c = 12.289 (3) Å; V = 4183.9 Å^3; Z = 4; D = 1.288 gm/cm^3 at 163 K. Intensities of all unique reflections within $0 \leq 2\theta \leq 150°$ were measured on an Enraf-Nonius CAD-4 diffractometer at 163 ± 2 K using Cu $K\alpha$ radiation. A $\theta - 2\theta$ scan technique was employed using a variable scan width of $(0.90 + 0.15 \tan \theta)°$. Intensities of three standard reflections were monitored every 2 h of X-ray exposure and they showed no significant variation. Out of the total of 4798 reflections, 4274 were considered observed on the basis of $I > 2\sigma(I)$. Data were corrected for Lorentz and polarization effects, but no absorption correction was made (μ = 15.2 cm^{-1}). The structure was solved by direct methods using the program MULTAN[34] and refined by a full-matrix least-squares routine[35] in which the quantity, $\sum\omega(|F_o| - |F_c|)^2$, was minimized. All the hydrogen atoms in the molecule were located from the difference Fourier maps and were refined with isotropic thermal parameters. One of the two water molecules in the structure was found to be disordered. The structure was refined to a final R factor of 0.034, R_w = 0.035 and an S = 1.67. The final difference map was featureless and contained peaks of height +0.2 e/Å^3. The molecular dynamics and energy minimization calculations were performed using the AMBER all atom force field.[33] Charges for the nonstandard peptide fragments of the molecule were calculated with the ab initio GAUSSIAN80 USCF program.

Acknowledgment. This work was supported by Grants CA-17256 (F.J.S.) and CA-17562 (D.v.d.H) awarded by the National Cancer Institute, DHEW, NOAA Sea Grant Project NA 83AA-D-00011, and NSF Grant EAR-8352055 (M.H.E.). We thank Dr. John Coll, James Cook University, Townsville, Australia, for inviting F.J.S. on a cruise of the R. V. Kirby, James Cook University, during which some specimens were collected, and Rick Willis for assistance in collection and freeze-drying of specimens. We thank Dr. W. C. Dunlap, Australian Institute of Marine Science, Townsville, Qld, Australia, for a generous gift of *L. patella*. We are grateful to Dr. Patricia Kott, Queensland Museum, Brisbane, Australia, for specimen identification. We thank the NSF, Grant CHE 8113507, and the University of Oklahoma Research Associates Fund for purchase of a high-field NMR spectrometer.

Supplementary Material Available: Bond lengths, bond angles, anisotropic thermal parameters for non-hydrogen atoms, and hydrogen atom parameters (10 pages). Ordering information is given on any current masthead page.

(34) Main, P.; Fiske, S. J.; Hull, S. E.; Lessinger, L.; Germain, G.; Declercq, J.-P.; Woolfson, M. M. MULTAN80, A system of computer programs for the automatic solution of crystal structures; University of York: England, 1980.

(35) Sheldrick, G. M. SHELX76, Program for Crystal structure determination. University of Cambridge: England, 1976.

Notes

Sulfircin: A New Sesterterpene Sulfate from a Deep-Water Sponge of the Genus *Ircinia*

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Received October 11, 1988

Relatively few sulfate-containing compounds have been reported from marine sources. Sponges have been the source of sterol sulfates[1] while saponin sulfates have been reported from echinoderms.[2] Recently, suvanine, a sesterterpene sulfate, has been reported from a sponge of the genus *Coscinoderma*.[3] From our efforts to identify antifungal agents from marine organisms, we report the isolation and structure elucidation of a new sesterterpene sulfate, **1**, for which we have assigned the common name sulfircin. Sulfircin was isolated as its *N,N*-dimethylguanidinium salt from a deep-water collection of the marine sponge *Ircinia* sp.[4] The presence of sulfate functionality in **1** makes it unusual, but even more unusual was its isolation as its *N,N*-dimethylguanidinum salt. To the best of our knowledge, suvanine is the only other example of a marine natural product isolated as its *N,N*-dimethylguanidinium salt.

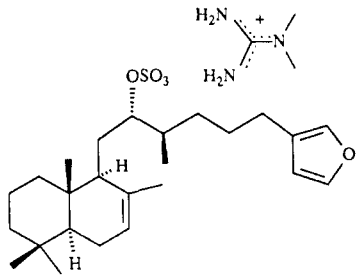
(1) See, for example: Fusetani, N.; Matsunaga, S.; Konosu, S. *Tetrahedron Lett.* **1981**, *22*, 1985–1988. Nakatsu, T.; Walker, R. P.; Thompson, J. E.; Faulkner, D. J. *Experimentia* **1983**, *39*, 759–761.

(2) See, for example: Kitagawa, I.; Kobayashi, M. *Tetrahedron Lett.* **1977**, 859–862; *Chem. Pharm. Bull.* **1978**, *26*, 1864–1873.

(3) Manes, L. V.; Crews, P.; Kernan, M. K.; Faulkner, D. J.; Fronczek, F. R.; Gandour, R. D. *J. Org. Chem.* **1988**, *53*, 570–575.

(4) Taxonomic classification carried out by Cristina Diaz as per Wiedenmayer and van Soest: Wiedenmayer, F. *A Monograph of the Shallow Water Sponges of the Western Bahamas*; Birkhauser Verlag: Basel, 1977; pp 287 (*Experientia* supplement 28). van Soest, R. W. M., personal communication. A voucher of the sponge is kept for reference at Harbor Branch Oceanographic Institution in Ft. Pierce.

0022-3263/89/1954-3472$01.50/0



1

The sponge was collected at a depth of 119 m off Fresh Creek, Andros, Bahamas in August of 1985, by using the Harbor Branch Oceanographic Institution's submersible the Johnson Sea-Link I. The methanol extract of the frozen sponge was chromatographed under reverse phase vacuum liquid chromatographic conditions[5] to yield, after recrystallization of the active fraction, pure 1 as colorless needles (mp 199–200 °C).

Examination of the broad band decoupled and DEPT carbon NMR spectra of 1 indicated the presence of 28 carbons with 45 attached hydrogens. The presence of a dimethylguanidinium moiety in 1 was suggested by a six-proton singlet observed at 2.96 ppm in the proton NMR spectrum, the carbon NMR resonances observed at 158.3 (s) and 37.8 (q), and the infrared absorbances at ν_{max} 3410–3180, 1620 cm^{-1}. The presence of a monosubstituted furan was suggested by the proton NMR resonances observed at 7.25, 7.13, and 6.17 ppm and the carbon NMR resonances observed at 142.8 (d), 138.9 (d), 125.2 (s), and 111.2 (d) ppm. An additional oxygen substitution was indicated by the resonances observed at 4.45 (dd, J = 11.1, 3.1 Hz) and 83.8 (d) in the proton and carbon NMR spectra, respectively. The presence of a methyl-substituted olefin was suggested by the olefinic proton resonance observed at 5.27 (br s) ppm, which showed one-bond coupling to the carbon observed at 122.2 (d) and long-range coupling to the carbons observed at 135.8 (s) and 22.4 (q) ppm and to the methyl protons observed at 1.61 ppm (br s) in the appropriate proton homonuclear or long- and short-range C–H two-dimensional NMR correlation experiments.[6] Three additional methyl singlets and one methyl doublet were observed at 0.79 (3 H, s), 0.76 (3 H, s), 0.66 (3 H, s), and 0.87 (3 H, d, J = 7.0 Hz) ppm in the proton NMR spectrum of 1. Only one major ion at m/z 627 and a small ion at 781 resulting from addition of dithiothreitol from the FAB matrix to the 627 ion was obsd. in the FAB mass spectrum of 1. High-resolution measurement of the 627 ion indicates a formula of $C_{31}H_{59}N_6O_5S$ (627.2490 obsd, 627.2468 calcd), which does not agree with the observed NMR data. The molecular formula determined through X-ray crystallography is $C_{28}H_{49}N_3O_5S$, which is in agreement with the NMR data. The difference between the observed ion and the expected molecular ion is $C_3H_{10}N_3$, which would correspond to addition of a second *N,N*-dimethylguanidinium ion to the protonated molecular ion.

As the spectral data suggested that 1 was a novel compound and crystals suitable for X-ray diffraction could be obtained from a solution of 1 in dichloromethane/methanol/water by slow evaporation at room temperature, the structure of 1 was determined by X-ray crystallography. Diffraction measurements were collected on a crystal of dimensions 0.37 × 0.25 × 0.13 mm by using a Rigaku AFC5S fully automated diffractometer and graphite-monochromated Cu Kα radiation (λ = 1.541 78 Å). Preliminary indications of the unit cell based on 25 randomly selected reflections revealed monoclinic symmetry with the following lattice parameters: a = 7.098 (2) Å, b = 14.776 (2) Å, and c = 14.579 (4) Å with β = 97.76°. The space group, on the basis of the observed systematic extinctions, was assigned as $P2_1$ (No. 4), Z = 2 with one molecule of composition $C_{28}H_{49}O_5N_3S$ forming the asymmetric unit. The volume was 1515 $Å^3$, and the calculated density was 1.17 g/cm^3. There were 2357 reflections collected with $2\theta \leq 120°$; of those reflections, 1531 (65%) with $I \geq 3\sigma(I)$ were adjudged observed.

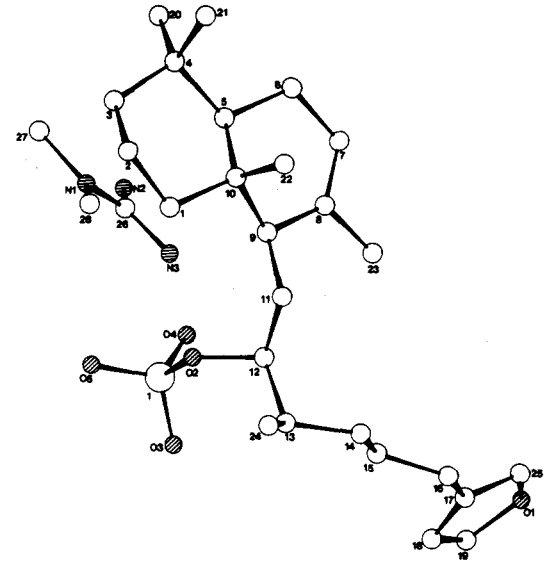


Figure 1. PLUTO drawing of sulfircin.[9]

The structure, shown in Figure 1, was solved by using MITHRIL.[7] Once it was recognized that the molecule contained a sulfate group, the structure solution proceeded normally. The positions of the hydrogen atoms were calculated. The full-matrix refinement[8] of the non-hydrogen atoms and inclusion of the hydrogen scattering factor have resulted in convergence of the crystallographic reliability factors to the following values: unweighted residual of 0.0555 and a weighted residual of 0.067.

A multitude of furan-containing sesterterpenes have been isolated from the sponge genus *Ircinia*. Sulfircin represents the first bicarbocyclic member of this class of compounds and is structurally similar to suvanine. In vitro bioassay of sulfircin against the fungal pathogen *Candida albicans* gave a minimum inhibitory concentration of 25 $\mu g/mL$.

Experimental Section

General Experimental. NMR spectra were obtained on a Bruker AM 360 NMR spectrometer fitted with an Aspect 3000 computer. Infrared spectra were recorded on a Perkin-Elmer 1310 IR spectrometer. Ultraviolet spectra were recorded on a Perkin-Elmer Lambda 3B UV/vis spectrophotometer.

(5) Blunt, J. W.; Calder, V.; Fenwick, G. D.; Lake, R. J.; McCombs, J. D.; Munro, M. H. G.; Perry, N. *J. Nat. Prod.* **1987**, *50*, 290–292.

(6) Aue, W. P.; Bartholdi, E.; Ernst, R. R. *J. Chem. Phys.* **1976**, *64*, 2229–2246. Bax, A.; Morris, G. *J. Magn. Reson.* **1981**, *42*, 501–505.

(7) Structure solution methods: C. J. Gilmore, MITHRIL, an integrated direct methods computer program. *J. Appl. Crystallogr.* **1984**, *17*, 42–46, University of Glasgow, Scotland. Texsan-Texray Structure Analysis Package, Molecular Structure Corporation (1985).

(8) Least-squares function minimized: $\sum w(|F_o| - |F_c|)^2$ where $w = 4F_o^2/\sigma^2(F_o^2)$, $\sigma^2(F_o^2) = [S^2(C + R^2B) + (pF_o^2)^2]/Lp^2$, S = scan rate, C = total integrated peak count, R = ratio of scan time to background counting time, B = total background count, Lp = Lorentz-polarization factor, p = p factor. Standard deviation of an observation of unit weight: $\sum w(|F_o| - |F_c|)^2/(No - Nv)]^{1/2}$ where No = number of observations.

(9) SKK-PUB and URANUS, programs to generate tables and plots respectively, written by Simon K. Kearsley, Yale University, 1985.

Collection and Isolation of Sulfircin (1). The sponge was collected at a depth of 119 m off Fresh Creek, Andros, Bahamas in August of 1985, by using the Harbor Branch Oceanographic Institution's submersible the Johnson Sea-Link I. One hundred grams of the frozen sponge (wet weight) was extracted exhaustively by homogenization with methanol in a blender. The extract was filtered and then concentrated by distillation under reduced pressure to yield 1.27 g of a pale yellow oil. The oil was chromatographed under reverse phase vacuum liquid chromatographic conditions by using a C-18 stationary phase and a step gradient of acetonitrile/water as eluent. The fraction eluting with acetonitrile/water (4:1) contained a microcrystalline solid, which after recrystallization from dichloromethane/methanol yielded 90 mg of sulfircin (1): colorless needles, mp 199–200 °C (dichloromethane/methanol); $[\alpha]^{20}_D$ +5° (*c* 0.006, ethanol); UV (EtOH) λ_{max} 208 nm (ϵ 1024); IR (KBr) ν (cm^{-1}) 3340, 3200, 2920, 1620, 1445, 1380, 1360, 1210, 1050, 1020, 960, 900, 870, 820, 746, 775; ^1H NMR (360 MHz, $CDCl_3/CD_3OD$ (4:1)) δ 7.25 (t, J = 1.2 Hz, H19), 7.13 (br s, H25), 6.17 (d, J = 1.2 Hz, H18), 5.27 (br s, H7), 4.45 (dd, J = 11.3, 3.1 Hz, H12), 2.84 (6 H, s, H27abc and H28abc), 2.34 (2 H, t, J = 7.5 Hz, H16ab), 2.21 (m, H13), 2.16 (m, H9), 1.84 (2 H, m, H6ab), 1.72 (m, H1a), 1.61 (3 H, br s, H23abc), 1.60 (m, H15a), 1.54 (m, H11a), 1.47 (m, H15b), 1.42 (m, H2a), 1.36 (m, H2b), 1.33 (m, H14a), 1.29 (m, H3a), 1.17 (m, H5), 1.14 (m, H14b), 1.09 (m, H3b), 1.07 (m, H1b), 0.87 (3 H, d, J = 7.0 Hz, H24abc), 0.79 (3 H, s, H20 or H21abc), 0.76 (3 H, s, H21 or H20abc), 0.66 (3 H, s, H22abc); ^{13}C NMR (90 MHz, $CDCl_3/CD_3OD$ (4:1)) 158.3 (s, C26), 142.8 (d, C19), 138.9 (d, C25), 135.8 (s, C8), 125.2 (s, C17), 122.2 (d, C7), 111.1 (d, C18), 83.9 (d, C12), 50.1 (d, C5), 48.8 (d, C9), 42.3 (t, C3), 38.7 (t, C1), 37.8 (q, 2 C, C27 and C28), 37.0 (d, C13), 36.6 (s, C4 or C10), 33.2 (q, C20), 33.1 (s, C4 or C10), 33.1 (t, C14), 28.3 (t, C15), 26.6 (t, C11), 24.9 (t, C16), 23.9 (t, C6), 22.4 (q, C23), 21.9 (q, C21), 18.9 (t, C2), 13.6 (q, C22), 13.1 (q, C24); FAB MS (magic bullet) m/z 781.4, 627.4, 613.4, 599.5, 562.3, 177.0.

Acknowledgment. Mass spectral determinations were performed at the University of Illinois. This is Harbor Branch Oceanographic Institution Contribution no. 712.

Registry No. 1, 120927-18-6.

Supplementary Material Available: Positional parameters, B(eq) values, bond angles and distances, torsion or conformation angles, and U values for compound 1 (7 pages). Ordering information is given on any current masthead page.

Selective Decarbalkoxylation of β-Keto Esters

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Received November 3, 1988

Decarbalkoxylation of a β-keto ester is a common synthetic manipulation. A wide variety of procedures have been developed for this transformation.[2] We now report a novel observation: the rate of decarbalkoxylation of a substituted β-keto ester is a predictable function of the substitution pattern.[3]

Scheme I

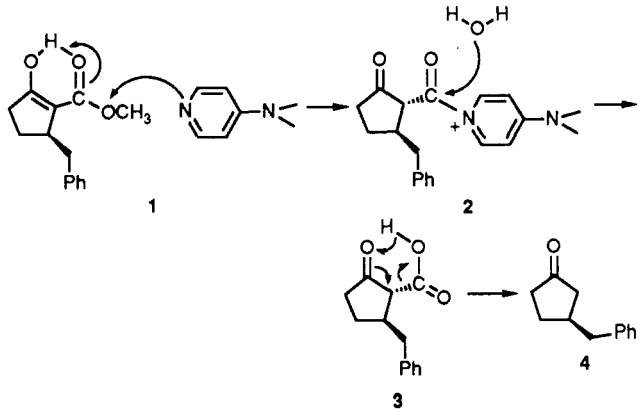
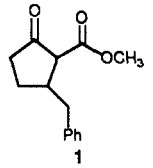
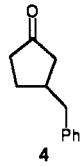
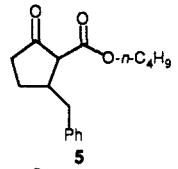
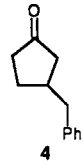
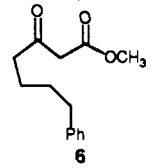
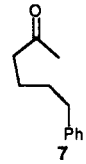
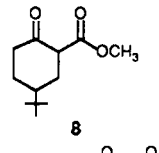
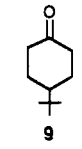
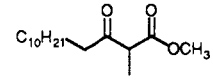


Table I

reactant	product	pH	half-life	yield, %
 1	 4	5	2.5 h	70
		7	6 h	70
		9	2.5 h	58
 5	 4	7	6 h	71
 6	 7	7	1 day	68
 8	 9	7	8 days	61
 10		7	no reaction	

The genesis of this project was the chance observation that 4-(dimethylamino)pyridine (4-DMAP), which we had shown to be an effective catalyst for transesterification of β-keto esters (toluene, 90 °C),[4] effects smooth decarbalkoxylation (1 → 4, Scheme I) of those same β-keto esters[5] if the toluene contains a little water.

The mechanism of this transformation is straightforward. Acylation of 4-DMAP by the ester 1, to give 2, is assumed to be the first step in the transesterification. Interception of the acylated pyridinium species by water would give 3, which at the elevated reaction temperature would spontaneously decarboxylate, to give the product.

To delineate the generality of this procedure, we submitted a representative series of β-keto esters to the reaction conditions (Table I). To our surprise, a substantial difference in rate appeared. As with the transesterification,

(1) Science and Engineering Scholar, University of Delaware Honors Program.

(2) A variety of reagent combinations have been used to effect decarbalkoxylation of β-keto esters. For leading references, see: (a) DMSO/NaCl: Krapcho, P. A.; Weimaster, J. F.; Eldridge, J. M.; Jahngen, E. G. E., Jr.; Lovey, A. J.; Stephens, W. P. *J. Org. Chem.* **1978**, *43*, 138. (b) DABCO: Huang, B. S.; Parish, E. J.; Miles, D. H. *J. Org. Chem.* **1974**, *39*, 2647. (c) Al_2O_3/dioxane: Greene, A. E.; Cruz, A.; Crabbe, P. *Tetrahedron Lett.* **1976**, 2707. (d) $Ba(OH)_2$: Miller, R. B.; Nash, R. D. *Tetrahedron* **1974**, *30*, 2961. (e) TMSI: Ho, T.-L. *Synth. Commun.* **1979**, *9*, 233. (f) Boric acid: Ho, T.-L. *Synth. Commun.* **1981**, *11*, 7. (g) $MgCl_2 \cdot nH_2O$/HMPA: Tsuda, Y.; Sakai, Y. *Synthesis* **1981**, 119. (h) Propane-1,2-diol: Aneja, R.; Hollis, W. M.; Davies, A. P.; Eaton, G. *Tetrahedron Lett.* **1983**, *24*, 4641.

(3) The enhanced lability of cyclopentanone carboxylates has been noted previously: Greene, A. E.; Luche, M.-J.; Serra, A. A. *J. Org. Chem.* **1985**, *50*, 3957, and ref. 2c, above.

(4) Taber, D. F.; Amedio, J. C., Jr.; Patel, Y. K. *J. Org. Chem.* **1985**, *50*, 3618.

(5) In the absence of 4-DMAP, no decarbalkoxylation is observed.

0022-3263/89/1954-3474$01.50/0