Metabolites of the Nudibranch *Chromodoris funerea* and the Singlet Oxygen Oxidation Products of Furodysin and Furodysinin

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Methanolic extracts of the nudibranch *Chromodoris funerea*, found in association with sponges of the genus *Dysidea*, contained 2-(2,4-dibromophenoxy)-3,4,5-tribromophenol (**1**), a major metabolite of *Dysidea herbacea*, *O*-methylfurodysinin lactone (**2**), and the furodysinin hydroperoxide **3**. The structure of the furodysinin hydroperoxide **3** was determined by X-ray analysis. Both *O*-methylfurodysinin lactone (**2**) and the furodysinin hydroperoxide **3** can be prepared by singlet oxygen oxidation of furodysinin (**4**), a metabolite of the sponge *Dysidea* sp., in methanol solution. Singlet oxygen oxidation of furodysinin (**4**) in nonpolar solvents gave a mixture of furodysinin lactone (**6**), the bis-epoxides **7** and **8**, and the epoxy lactones **9** and **10**, three of which were subsequently found in an acetone extract of *C. funerea*, together with a γ-hydroxybutenolide **11** that was prepared by oxidation of furodysin (**5**) with singlet oxygen.

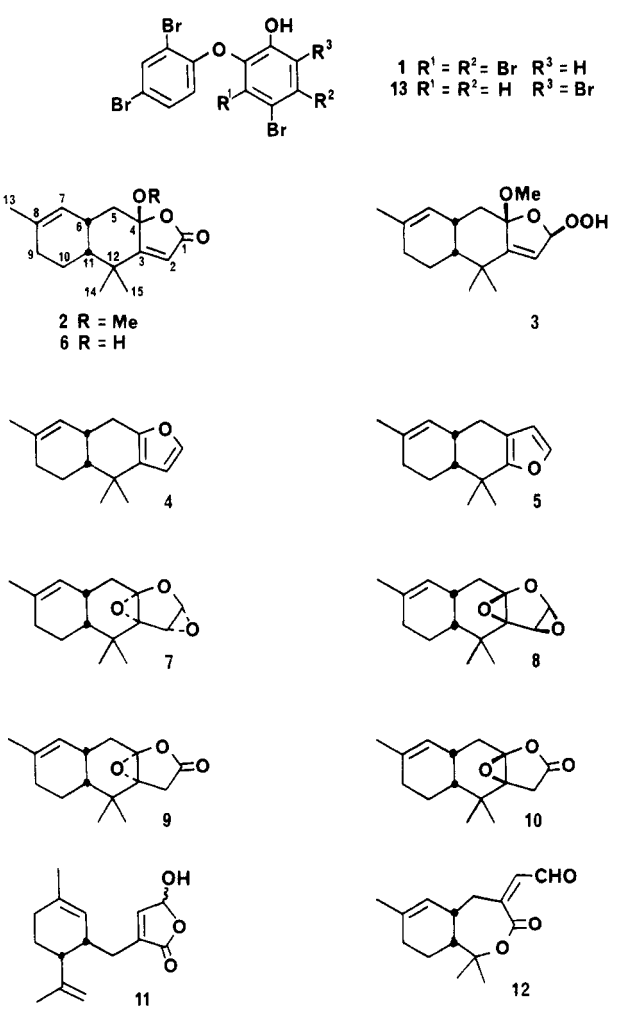
It is now well established that most dorid nudibranchs have evolved chemical defense mechanisms that employ chemicals obtained from food sources.[1] Sponge-eating dorid nudibranchs have specialized to such a degree that they cannot only tolerate the chemicals that generally protect sponges from predation but are able to select the most repugnant of these chemicals and sequester them for their own defense. A few nudibranchs are capable of de novo synthesis of defensive chemicals[2] but they are the exception rather than the rule. In this paper we will provide circumstantial evidence to suggest that it is possible for nudibranchs to modify dietary chemicals to produce more effective allemones.

Chromodoris funerea[3] was first collected in 1981 from a shallow area of Iwayama Bay, Palau. It was generally found on or near sponges of the genus *Dysidea*, at least three species of which are common in Iwayama Bay. Recent collections of *C. funerea* were obtained from the same shallow water location and from a marine lake located on an island inside Iwayama Bay and approximately 1 mile from the first site.

The first collection of eight animals was stored in methanol at −10 °C for 6 months. Chromatography of the dichloromethane soluble material from the methanolic extract of *C. funerea* on silica gel followed by LC on Partisil using 10% ether in hexane as eluant gave 2-(2,4-dibromophenoxy)-3,4,5-tribromophenol (**1**, 14 mg, 1.8 mg/animal), *O*-methylfurodysinin lactone (**2**, 8 mg, 1.0 mg/animal), and the furodysinin hydroperoxide **3** (5 mg, 0.65 mg/animal). The sample of 2-(2,4-dibromophenoxy)-3,4,5-tribromophenol (**1**) was identical in all respects with an authentic sample from *Dysidea herbacea*,[4] a sponge that was found at the collection site.

O-Methylfurodysinin lactone (**2**), $[\alpha]_D$ −85° (*c* 0.5, $CHCl_3$), was isolated as colorless crystals, mp 111–112 °C. The molecular formula, $C_{16}H_{22}O_3$, was obtained by high resolution mass spectroscopy. The infrared band at 1770 cm^{-1}, ^1H NMR signals at δ 3.12 (s, 3 H), and 5.69 (s, 1 H), and ^{13}C NMR signals at δ 171.9 (s), 129.2 (s), 117.9 (d), 106.9 (s), and 49.9 (q) all indicated the presence of a 3,4-disubstituted-4-methoxybutenolide moiety. Comparison of the remaining signals in the ^1H and ^{13}C NMR spectra

Chart I



with those of furodysinin (**4**)[5] strongly suggested that *O*-methylfurodysinin lactone (**2**) was derived by oxidation of the furan ring of furodysinin (**4**).

Furodysinin hydroperoxide (**3**), $[\alpha]_D$ −63.4° (*c* 0.5, $CHCl_3$), was obtained as colorless crystals, mp 142–143 °C, from ether–hexane solution. The highest peak in the mass

(1) Faulkner, D. J.; Ghiselin, M. T. *Mar. Ecol. Prog. Ser.* **1983**, *13*, 295.
(2) Cimino, G.; De Rosa, S.; De Stefano, S.; Morrone, R.; Sodano, G. *Tetrahedron* **1984**, *41*, 1093. Gustafson, K.; Andersen, R. J. *Tetrahedron* **1984**, *41*, 1101.
(3) Collingwood, C. *Trans. Limn. Soc. London, Zool.* (2) **1981**, *2*, 123.
(4) Cartê, B.; Faulkner, D. J. *Tetrahedron* **1981**, *37*, 2335.
(5) Kazlauskas, R.; Murphy, P. T.; Wells, R. J.; Daly, J. J.; Schonholzer, P. *Tetrahedron Lett.* **1978**, 4951. Dunlop, R. W.; Kazlauskas, R.; March, G.; Murphy, P. T.; Wells, R. J. *Aust. J. Chem.* **1982**, *35*, 95.

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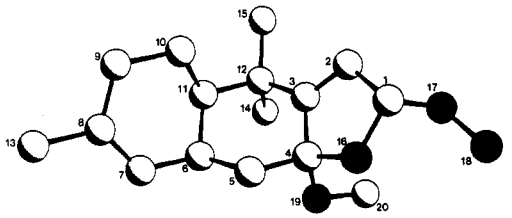


Figure 1. Computer-generated perspective drawing of the final X-ray model of the furodysinin hydroperoxide **3**. Carbon atoms are shown in white, oxygen atoms are in black, hydrogens are omitted for clarity, and the absolute configuration illustrated is an arbitrary choice.

spectrum, m/z 262 (M – H_2O) did not reveal the true molecular formula, $C_{16}H_{24}O_4$, that was obtained by elemental analysis. The ^1H NMR spectrum of hydroperoxide **3** was similar to that of butenolide **2** except that in the downfield region it contained two mutually coupled signals at δ 5.73 (d, 1 H, J = 1.4 Hz) and 5.41 (d, 1 H, J = 1.4 Hz) in place of the downfield singlet at δ 5.69 in the spectrum of **2**. The infrared spectrum contained a hydroxyl absorption at 3500 cm^{-1} but no carbonyl bands. The ^{13}C NMR spectrum contained signals at δ 152.5 (s), 118.9 (d), 110.7 (s), 108.2 (d) and 50.3 (q). These data suggested that the lactone carbonyl in **2** was replaced by a hemiacetal in **3** yet the molecular formula required one more oxygen. The reaction of **3** with potassium iodide in aqueous acetic acid to liberate iodine indicated that the additional oxygen was incorporated into a hydroperoxide group. Although hydroperoxy acetals had been reported as singlet oxygen oxidation products of furans,[6] we were nonetheless skeptical that such products could be stable crystalline compounds and therefore submitted the hydroperoxide for structural determination by X-ray analysis.

A computer-generated perspective drawing of the final X-ray model of furodysinin hydroperoxide (**3**) is given in Figure 1. Hydrogens are omitted for clarity, and the absolute configuration illustrated is an arbitrary choice. Bond angles and distances agree well with generally accepted values. The A-ring is in a half-chair conformation and the B-ring is in a chair conformation. The dihydrofuran ring is essentially planar with all internal dihedral angles less than 8°. The O16–C1–O17–O18 dihedral angle is –66°, and the O16–C4–O19–C20 dihedral angle is –60°.

Since *Chromodoris funerea* had undoubtedly obtained the brominated phenol **1** from the sponge *Dysidea herbacea* on which it was found, we expected to find a dietary source of *O*-methylfurodysinin lactone (**2**) and furodysinin hydroperoxide (**3**) among the sponges of Iwayama Bay. A careful examination of available sponges revealed instead that one of the most common sponges of Iwayama Bay, a species of *Dysidea*, contained a mixture of furodysinin (**4**, 0.9% dry weight) and furodysin (**5**, 0.3% dry weight) but there was no trace (<0.01% dry weight) of the oxidation products. We therefore proposed that the nudibranch was eating the *Dysidea* species and was responsible for the oxidation of furodysinin (**4**) to an unidentified oxidation product that reacted with the methanol extraction solvent to produce the methoxy lactone **2** and hydroperoxide **3**.

The oxidation procedure could be duplicated in the laboratory by reacting a methanolic solution of furodysinin (**4**) at –78 °C with singlet oxygen, generated photolytically by using rose bengal as catalyst, to obtain the hydroperoxide **3** in 77% yield. The synthetic material was identical in all respects, including optical rotation, with the natural material. On heating or prolonged standing, the hydroperoxide **3** was slowly converted into the methoxy lactone **2**, confirming its structural assignment. Treatment of the hydroperoxide **3** with activated manganese dioxide in ether at 25 °C also caused its rapid conversion to the methoxy lactone **2** that was identical in all respects with the natural material.

Our results were somewhat different from those reported by Grode and Cardellina,[7] who isolated relatively small amounts of furodysinin (**4**) and furodysinin lactone (**6**) from an acetone extract of the sponge *Dysidea etheria* and a mixture of furans that included furodysinin (**4**), but no furodysinin lactone (**6**), from an acetone extract of the nudibranch *Hypselodoris zebra*. Since the methoxy group incorporated into the methoxy lactone **2** and the hydroperoxide **3** was undoubtedly from the extraction solvent, we decided to synthesize the potentially unstable oxidation products of furodysinin (**4**) and furodysin (**5**) and then search for these products in new samples of *C. funerea* that were to be stored in acetone.

The oxidation of furodysinin (**4**) with singlet oxygen gave complex mixtures of products that were highly dependent on reaction and workup conditions. In a typical experiment, a dichloromethane solution of furodysinin (**4**) was allowed to react at –78 °C with singlet oxygen generated by photolysis of oxygen in the presence of a polymer-bound rose bengal catalyst.[8] On completion, the reaction mixture was warmed to room temperature and analyzed by ^1H NMR, and the products were separated by LC on Partisil. The isolated products consisted of two bis-epoxides **7** and **8**, a mixture of two epoxy lactones **9** and **10** and furodysinin lactone (**6**). Treatment of a dichloromethane solution of furodysinin (**4**) with singlet oxygen at –78 °C followed by quenching the product with methanol and allowing the reaction mixture to warm to 25 °C gave the same array of products (**6**–**10**) together with *O*-methylfurodysinin lactone (**2**).[9]

Furodysinin lactone (**6**) was identified by comparison of its spectral data with literature values.[7] The bis-epoxides **7** and **8** and epoxy lactones **9** and **10** were all isomeric, having the molecular formula $C_{15}H_{20}O_3$. The bis-epoxides were characterized by ^1H NMR signals at δ 5.48 (d, 1 H, J = 1 Hz) and 3.55 (d, 1 H, J = 1 Hz) for **7** and 5.58 (d, 1 H, J = 1 Hz) and 3.80 (d, 1 H, J = 1 Hz) for **8** that could be assigned to the C-1 and C-2 protons. The remaining signals in the ^1H NMR spectra could all be assigned by assuming the expected furodysinin skeleton. Assignment of the relative stereochemistry of the two isomers was based on the argument that the C-6 proton signal in **8** (δ 2.83) should be at higher field than the C-6 proton signal in **7** (δ 2.99) since it lies within the ring current of the 3,4-epoxide ring.[10] Unfortunately we were unable to confirm this assignment by NOE observations. Although **7** was slightly more stable than **8**, both bis-epoxides slowly decomposed at room temperature and were particularly unstable to silica gel chromatography.

(6) Gollnick, K., Griesbeck, A. *Tetrahedron* **1985**, *41*, 2057.

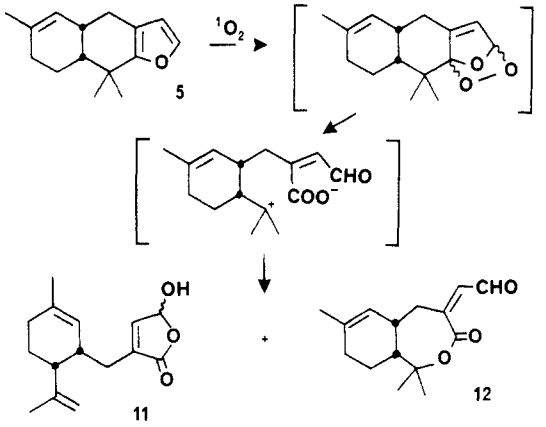
(7) Grode, S. H.; Cardellina, J. H., II. *J. Nat. Prod.* **1984**, *47*, 76.

(8) Eschaap, A. P.; Thayer, A. L.; Blossey, E. C.; Neckers, D. C. *J. Am. Chem. Soc.* **1975**, *97*, 3741.

(9) The formation of only one of two possible steroisomers of the methoxy lactone **2**, hydroperoxide **3** and the hydroxy lactone **6** deserves comment. We propose that two stereoisomeric endoperoxides are formed. In dichloromethane solution, the two endoperoxides give rise to two bis-epoxides and two epoxy lactones. The hydroxy lactone **6** can equilibrate to the more stable isomer via the corresponding keto acid. The formation of a single hydroperoxide can be explained by equilibration of the endoperoxide via a Criegee-type ozonide equilibration mechanism that can occur more readily in polar solvents.

(10) Jackman, L. M.; Sternhall, S. *Applications of Nuclear Magnetic Resonance Spectroscopy in Organic Chemistry*, 2nd ed.; Pergamon: Oxford, **1969**; p 99.

Scheme I. Mechanism for the Oxidation of Furodysin (5) with Singlet Oxygen To Produce the γ-Hydroxybutenolide 11 and the Lactone 12



The 1:1 mixture of epoxy lactones **9** and **10** had IR bands at 1800 and 1785 cm^{-1}, typical of β,γ-epoxy γ-lactones.[11] The ^1H NMR spectra of both molecules were compatible with structures based on the furodysinin skeleton with the C-2 methylene proton signals appearing at δ 2.77 (d, 1 H, J = 18 Hz) and 2.71 (d, 1 H, J = 18 Hz) in **9** and at 2.82 (d, 1 H, J = 19 Hz) and 2.74 (d, 1 H, J = 19 Hz) in **10**. The relatively rapid decomposition of epoxy lactone **9** left a sample significantly enriched with epoxy lactone **10**. The key ^1H NMR signals for both compounds could therefore be assigned without difficulty. We have assigned the stereochemistry of the epoxy lactones by comparing selected ^1H NMR data. The chemical shifts of the C-6 proton signals at δ 2.42 for **9** and 2.30 for **10** are in agreement with the observation that protons in the ring current of an epoxide ring are shifted upfield. In addition, examination of a molecular model suggests that the dihedral angle between H-5 (equatorial) and H-6 is smaller in **9** than in **10**, resulting in the observed difference in coupling constants (7.2 Hz in **9**, 5 Hz in **10**).

The reaction of furodysin (**5**) with singlet oxygen gave a 2:1 mixture of the γ-hydroxybutenolide **11** and the lactone **12**. The ^1H NMR spectrum of the γ-hydroxybutenolide **11** contained signals at δ 4.89 (br s, 1 H), 4.71 (br s, 1 H), and 1.80 (br s, 3 H) that are appropriate for an isopropenyl group, at 6.83 (d, 1 H, J = 1 Hz) and 6.09 (d, 1 H, J = 1 Hz) for the olefinic and hemiacetal protons, and at 1.65 (br s, 3 H) and 5.36 (br d, 1 H, J = 11 Hz) due to the olefinic methyl and olefinic proton on the six-membered ring. The lactone **12** is responsible for ^1H NMR signals at δ 9.82 (d, 1 H, J = 8 Hz), 6.14 (d, 1 H, J = 8 Hz), 1.56 (s, 3 H), and 1.53 (s, 3 H), assigned to the aldehyde, olefinic, and *gem*-dimethyl protons, respectively. The formation of **11** and **12** from furodysin (**5**) can be explained if an intermediate endoperoxide undergoes ring cleavage (Scheme I).

A collection of 18 animals from the same location in Iweyama Bay was stored in acetone for 2 weeks. Chromatography of the dichloromethane-soluble material from the acetone extract gave furodysinin (**4**), furodysin (**5**), and 2-(2,4-dibromophenoxy)-4,6-dibromophenol (**13**) as the major metabolites. Only the more stable of the minor metabolites could be isolated. Furodysinin lactone (**6**), the γ-hydroxybutenolide **11**, and one epoxy lactone **10** were purified by LC. The second epoxy lactone **9** and both bis-epoxides **7** and **8** were detected by ^1H NMR spectroscopy in impure fractions from the initial chromatographic separation but decomposed during subsequent chromatographic procedures.

The results presented in this paper provide support for the hypothesis that the furanosesquiterpenes are being oxidized by *C. funerea* to a product that can be stored but cannot be isolated because it either decomposes or reacts with solvent. From purely mechanistic considerations, a mixture of endoperoxides would be the most logical candidate for the oxidation product if it were not for the fact that the synthetic endoperoxides are unstable at ambient temperatures. A possible rationale for the oxidation of furans by the nudibranch is that the biological activities of the oxidation products are an order of magnitude greater than those of the furans. For example, in a fish feeding inhibition assay employing the "spotted kelpfish" *Gibbonsia elegans*,[12] furodysinin (**4**) caused food rejection at a concentration of 50 μg/mg pellet while *O*-methylfurodysinin lactone (**2**) and the furodysinin hydroperoxide **3** were effective at 10 μg/mg and 1–5 μg/mg, respectively.

Experimental Section

***Chromodoris funerea*: Collection, Extraction, and Chromatography.** Eight specimens of *Chromodoris funerea* (81-085) were collected by hand in the shallow waters (−2 m) of Iwayama Bay, Palau, Western Caroline Islands. The nudibranchs were stored in methanol (~300 mL) at −10 °C for 6 months. The solvent was decanted and the nudibranchs were washed with fresh methanol (2 × 200 mL). The combined methanol extracts were evaporated and the resulting aqueous suspension (~75 mL) was extracted with dichloromethane (3 × 50 mL). The combined extracts were dried over anhydrous sodium sulfate and filtered, and the solvent was evaporated to obtain a green oil (272 mg, 34 mg/animal). The oil was chromatographed on TLC grade silica gel using eluants of increasing polarity from hexane to ethyl acetate. The fraction eluted with 5% ethyl acetate in hexane was subjected to LC on Partisil using 10% ether in hexane as eluant to obtain 2-(2,4-dibromophenoxy)-3,4,5-tribromophenol (**1**, 14 mg, 1.8 mg/animal), *O*-methylfurodysinin lactone (**2**, 8 mg, 1 mg/animal), and the furodysinin hydroperoxide **3** (5 mg, 3.6 mg/animal). The ^1H NMR spectra of the crude extract and chromatographic fractions did not contain signals due to furodysinin (**4**) or furodysin (**5**).

***O*-Methylfurodysinin lactone (2)**: mp 111–112 °C (pentane): $[\alpha]_D$ −84.8° (*c* 0.5, $CHCl_3$): IR ($CHCl_3$) 2950, 1770, 1640, 1440, 1300, 1275, 1110, 1055 cm^{-1}; ^1H NMR (CCl_4) δ 1.20 (m, 2 H), 1.22 (s, 3 H), 1.35 (s, 3 H), 1.47 (t, 1 H, J = 13.5 Hz), 1.60 (br s, 3 H), 1.65 (m, 1 H), 1.94 (br d, 2 H, J = 6.6 Hz), 2.29 (dd, 1 H, J = 3.8, 13.5 Hz), 2.73 (m, 1 H), 3.12 (s, 3 H), 5.34 (br d, 1 H, J = 5.3 Hz), 5.69 (s, 1 H); ^{13}C NMR (C_6D_6) δ 171.9 (s), 133.5 (s), 129.2 (s), 124.2 (d), 117.9 (d), 106.9 (s), 49.9 (q), 47.6 (d), 40.6 (t), 38.2 (s), 31.0 (t), 30.5 (d), 25.6 (q), 25.0 (q), 23.3 (q), 18.5 (t); HRMS, obsd *m/z* 262.1559, $C_{16}H_{22}O_3$ requires 262.1569

The furodysinin hydroperoxide 3: mp 142–143 °C (ether/hexane); $[\alpha]_D$ −63.4° (*c* 0.5; $CHCl_3$); IR ($CHCl_3$) 3500, 3000, 1430, 1320, 1290, 1170, 1120, 1055 cm^{-1}; ^1H NMR (CCl_4) δ 1.14 (s, 3 H), 1.26 (s, 3 H), 1.29 (t, 1 H, J = 13.5 Hz), 1.58 (br s, 3 H), 1.90 (br d, 1 H, J = 13.5 Hz), 1.90 (br d, 1 H, J = 6 Hz), 2.10 (dd, 1 H, J = 3.7, 13.5 Hz), 2.68 (m, 1 H), 3.08 (s, 3 H). 5.33 (br d, 1 H), 5.41 (d, 1 H, J = 1.4 Hz), 5.73 (d, 1 H, J = 1.4 Hz), 8.48 (br s, 1 H, D_2O exchangeable); ^{13}C NMR (C_6D_6) δ 152.5 (s), 133.0 (s), 125.5 (d), 118.9 (d), 110.7 (s), 108.2 (d), 50.3 (q), 47.0 (d), 42.5 (t), 37.0 (s), 31.3 (d), 31.3 (t), 27.0 (q), 25.6 (q), 23.4 (q), 18.8 (t); HRMS, obsd *m/z* 262.1565, $C_{16}H_{22}O_3$ ($M-H_2O$) requires 262.1569. Anal. (Galbraith Labs, Inc.) Found for $C_{16}H_{24}O_4$: C, 68.74; H, 8.83. Calcd: C, 68.53; H, 8.63.

Single-Crystal X-ray Structure Determination of the Furodysinin Hydroperoxide 3. A roughly cubic crystal with approximate sides of 0.5 mm was cut from a much longer crystal. Preliminary X-ray photographs displayed orthorhombic symmetry, and accurate lattice constants of *a* = 6.584 (1), *b* = 15.307

(11) Cimino, G.; De Stefano, S.; Minale, L. *Experientia* **1974**, *30*, 18.

(12) Pawlik, J. R.; Albizati, K. F.; Faulkner, D. J. *Mar. Ecol. Prog. Ser.* **1986**, *30*, 25.

(1), c = 15.387 (2) Å were determined from a least-squares fit of 15 moderate angle 2θ values. Systematic extinctions, crystal density, and the presence of chirality were uniquely accommodated by space group $P2_12_12_1$ with one molecule of composition $C_{16}H_{24}O_4$ forming the asymmetric unit. All unique diffraction maxima with $2\theta \leq 114°$ were collected by using a variabe speed, 1° ω-scan and graphite monochromated Cu K$\bar{\alpha}$ radiation (1.54178 Å). Of the 1249 reflections surveyed in this fashion, 1130 (90%) were judged observed ($|F_o| \geq 3\sigma(F_o)$) after correction for Lorentz, polarization, and background effects.[13] These reflections were used in all subsequent calculations. A phasing model was achieved by using a multisolution tangent formula approach followed by tangent formula recycling of a plausible 12-atom fragment.[14] All of the non-hydrogen atoms were found in this way. Hydrogens were located on difference electron density syntheses following partial refinement. Block-diagonal least-squares refinements with anisotropic non-hydrogen atoms and isotropic hydrogens have converged to a standard crystallographic residual of 0.372 for the observed data. Crystallographic parameters are available and are described in the paragraph entitled supplementary material available.

Extraction of *Dysidea* sp. *Dysidea* sp. (81-066) was collected by hand at Iwayama Bay. The sponge (50 g dry weight) was stored frozen for 2 years and then lyophilized and extracted with ethyl acetate. Evaporation of the ethyl acetate gave a green oil that was chromatographed on TLC grade silica gel eluting with solvents of increasing polarity from hexane through ether to ethyl acetate. The fraction eluted with hexane was purified by LC on μ-Partisil using hexane as eluant to obtain furodysinin (**4**, 450 mg, 0.9% dry weight) and furodysin (**5**, 150 mg, 0.3% dry weight). The physical and spectral data were identical with those reported by Kazlauskas et al.[5]

A sample of *Dysidea* sp. that had been stored in methanol for 2 years was extracted by using the same procedure that was used on *C. funerea*. The oxidized products **2** and **3** could not be detected (limits of detection <0.01% dry weight).

Oxidation of Furodysinin (4) with Singlet Oxygen. (a) Methanol Solution. A solution of furodysinin (**4**, 125 mg) in methanol (100 mL) containing rose bengal (2 mg) was stirred at −78 °C under an atmosphere of oxygen and was irradiated with a 200-W incandescent lamp. After 1 h the reaction was judged complete and the solvent was evaporated. The residue was taken up in 1:1 ether–hexane and filtered through a small pad of silica gel. Evaporation of the ether gave the crystalline furodysinin hydroperoxide **3** (125 mg, 77% theoretical) that was identical in all respects with the natural material.

Activated manganese dioxide[15] was added to a solution of the furodysinin hydroperoxide **3** (100 mg) in ether (50 mL) and the solution was stirred for 1.5 h at 25 °C. The solution was filtered and the product chromatographed on Partisil using 25% ether in hexane as eluant to obtain *O*-methylfurodysinin lactone (**2**, 39.4 mg, 42% theoretical).

(b) Dichloromethane Solution. A solution of furodysinin (4, 8 mg) in dry dichloromethane (5 mL) containing polystyrene-bound rose bengal catalyst[8] was stirred at −78 °C under an atmosphere of oxygen and was irradiated with a 200-W tungsten lamp. After 2.5 h the reaction was judged complete by TLC. the solution was filtered and the solvent evaporated to obtain a crude product (8.4 mg, 91.5% based on $C_{15}H_{20}O_3$). The product was separated by LC on Partisil to obtain a 1:1 mixture of epoxy lactones **9** and **10** (1.7 mg, 18.6% theoretical), bis-epoxide **7** (1.33 mg, 14.6% theoretical), bis-epoxide **8** (0.54 mg, 5.9% theoretical), and furodysinin lactone (**6**, 0.96 mg, 10.5% theoretical).

(c) Dichloromethane Solution. Methanol Quench. A solution of furodysinin (**4**, 5.0 mg) in dichloromethane was photo-oxidized at −78 °C using the procedure above. After 2.5 h, methanol (100 μL) was added and the solution was worked up as before. A ^{1}H NMR analysis of the product revealed 35% *O*-methylfurodysinin lactone, 15% furodysinin lactone, 15% bis-epoxide **7**, 10% bis-epoxide **8**, 10% epoxy lactone **9**, and 15% epoxy lactone **10** (all values to nearest 5%).

Bis-epoxide 7: oil; IR 1075, 1060, 1045 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 5.48 (d, 1 H, J = 1 Hz), 5.34 (br s, 1 H), 3.55 (d, 1 H, J = 1 Hz), 2.99 (m, 1 H), 2.34 (m, 2 H), 2.12 (dd, 1 H, J = 13, 8 Hz), 1.91 (br q, 1 H, J = 8 Hz), 1.77 (m, 1 H), 1.67 (br s, 3 H), 1.62 (dd, 1 H, J = 13, 10 Hz), 1.51 (m, 1 H), 1.21 (s, 3 H), 1.15 (s, 3 H).

Bis-epoxide 8: oil; IR 1065, 1040 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 5.58 (d, 1 H, J = 1 Hz), 5.41 (br s, 1 H), 3.80 (d, 1 H, J = 1 Hz), 2.83 (m, 1 H), 2.33 (m, 2 H), 2.09 (dd, 1 H, J = 13, 12 Hz), 1.93 (m, 1 H), 1.88 (m, 1 H), 1.70 (m, 1 H), 1.67 (br s, 3 H), 1.17 (s, 3 H), 1.04 (s, 3 H).

1:1 mixture of epoxy lactones 9 and 10: oil; IR ($CHCl_3$) 1800, 1735 (sh), 1170, 1160, 1070, 1065 cm^{-1}; HRMS, obsd m/z 248.1415, $C_{15}H_{20}O_3$ requires 248.1413.

Epoxy lactone 10: ^{1}H NMR ($CDCl_3$) δ 5.46 (br d, 1 H, J = 6 Hz), 2.82 (d, 1 H, J = 19 Hz), 2.74 (d, 1 H, J = 19 Hz), 2.44 (dd, 1 H, J = 15, 5 Hz), 2.30 (m, 1 H), 2.04 (m, 2 H), 1.90 (dd, 1 H, J = 15, 11 Hz), 1.63 (br s, 3H), 1.11 (s, 3 H), 1.10 (s, 3 H).

Epoxy lactone 9: (by subtraction) ^{1}H NMR ($CDCl_3$) δ 5.46 (br, 1 H), 2.77 (d, 1 H, J = 18 Hz), 2.71 (d, 1 H, J = 18 Hz), 2.42 (m, 1 H), 2.38 (dd, 1 H, J = 19, 7 Hz), 2.08 (dd, 1 H, J = 19, 12 Hz), 1.63 (br s, 3 H), 1.18 (s, 3 H), 1.16 (s, 3 H).

Oxidation of Furodysin (5) with Singlet Oxygen. A solution of furodysin (**5**, 25 mg) in dichloromethane was oxidized at −78 °C with singlet oxygen using the procedure above. After 3 h, the solvent was evaporated to obtain an oil (27.5 mg) part of which was purified by LC on Partisil ODS (70% MeOH/H_2O) to obtain a 2:1 mixture of γ-hydroxybutenolide **11** and lactone **12** (10.5 mg, 53% theoretical) that eluted as a single peak. A pure sample of the γ-hydroxybutenolide **11** was obtained from *C. funerea* (see below).

γ-Hydroxybutenolide 11: IR ($CHCl_3$) 3100–3500 (br), 1770, 1010 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 6.83 (d, 1 H, J = 1 Hz), 6.09 (d, 1 H, J = 1 Hz), 5.36 (br d, 1 H, J = 11 Hz), 4.89 (br s, 1 H), 4.71 (br s, 1 H), 3.40 (br s, 1 H, OH), 2.58 (m, 1 H), 1.80 (br s, 3 H), 1.65 (br s, 3 H); HRMS obsd m/z 248.1416, $C_{15}H_{20}O_3$ requires 248.1413.

Lactone 12: (by subtraction) ^{1}H NMR ($CDCl_3$) δ 9.82 (d, 1 H, J = 11 Hz), 6.14 (d, 1 H, J = 11 Hz), 1.56 (s, 3 H), 1.53 (s, 3 H).

Extraction of *C. funerea* with Acetone. Eighteen specimens of *C. funerea* (85-010) were collected from the same location and stored for 2 weeks in acetone. The acetone was decanted and evaporated and the residue partitioned between dichloromethane (3 × 50 mL) and water (30 mL). The combined extracts were dried over sodium sulfate and the solvent evaporated to obtain an oil (122 mg) that was subjected to flash chromatography on silica gel. The fraction eluted with hexane contained a 1:2 mixture of furodysinin (**4**) and furodysin (**5**) (46 mg total). A more polar fraction (20.7 mg) contained bis-epoxides **7** and **8**, epoxy lactones **9** and **10** and the tetrabromophenol **13**, as judged by the ^{1}H NMR spectrum. Chromatography of this fraction by LC on Partisil (eluant: 10% EtOAc/hexane) gave the tetrabromophenol **13** (6 mg), identical with authentic material, and epoxy lactone **10** (1.6 mg). A third more polar fraction from the flash chromatography was fractionated by LC on Partisil (eluant: 45% EtOAc/hexane) to obtain furodysinin lactone (**6**, 3.1 mg) and the γ-hydroxybutenolide **11** (2 mg).

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(13) All crystallographic calculations were done on a PRIME 850 computer operated by the Cornell Chemistry Computing Facility. Principal programs employed were REDUCE and UNIQUE, data reduction programs by M. E. Leonowicz, Cornell University, 1978; MULTAN 78, MULTAN 80, and RANTAN 80, systems of computer programs for the automatic solution of crystal structures from X-ray diffraction data (locally modified to perform all Fourier calculations including Patterson syntheses) written by P. Main, S. E. Hull, L. Lessinger, G. Germain, J. P. Declercq, and M. M. Woolfson, University of York, England, 1978 and 1980; DIRDIF written by P. T. Buerskens et al., University of Nijmegen, Netherlands, 1981; MITHRIL, an automatic solution package written by C. J. Gilmore, University of Glasgow, Scotland, 1982; BLS78A, an anisotropic block diagonal least-squares refinement written by K. Hirotsu and E. Arnold, Cornell University, 1980; PLUTO78, a crystallographic illustration program by W. D. S. Motherwell, Cambridge Crystallographic Data Centre, 1978; and BOND, a gram to calculate molecular parameters and perpare tables written by K. Hirotsu, Cornell University, 1978.

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Supplementary Material Available: Tables of fractional coordinates, thermal parameters, interatomic distances, and interatomic angles for the furodysinin hydroperoxide 3 (4 pages). Ordering information is given on any current masthead page.

Solvolysis in Mixed Solvents with Complementary Electrophilic and Nucleophilic Properties. Hexafluoro-2-propanol and 1,3-Propanedithiol

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Solvolytic rates for alkyl tosylates and brosylates were determined for various binary compositions of hexafluoro-2-propanol (HFIP) and 1,3-propanedithiol (PDT). N_{OTs} and Y_{OTs} values were determined by using the Bentley–Schleyer approach. Solvent ionizing power dropped off sharply as PDT replaced HFIP in the mixtures, and a Y_{OTs} value of −4.7 was estimated for pure PDT. A plot of N_{OTs} vs. molar composition of HFIP was nonlinear with solvent nucleophilicity dropping off at high HFIP concentrations. Nevertheless, HFIP–PDT mixtures were, surprisingly, 3 to 4 orders of magnitude more nucleophilic than HFIP–H_2O mixtures although pure PDT is estimated to be only 2.5 orders of magnitude more nucleophilic than pure water. To explain these observations, it is suggested that, at high concentrations of electrophilic solvents, hydrogen bonding to the oxygen or sulfur atom in the more nucleophilic solvent molecules reduces the nucleophilicity of these solvents. The good correlation between rates of a wide variety of substrates in HFIP–PDT and 97% aqueous HFIP suggests that these substrates react by a k_c (or k_Δ) mechanism in these solvents.

The effects that the solvent has on the course of displacement reactions has attracted considerable attention from chemists since Ingold and his colleagues began sorting out the nature of displacement of covalently bound groups.[1,2] Grunwald and Winstein[2,3] made the first major attempt to quantify these effects when they proposed to use eq 1 to correlate these effects for the S_N1 solvolysis of

$$\log (k/k_0) = mY \quad (1)$$

tert-butyl chloride. This two-parameter equation has survived and proven useful despite its simplicity. In recent years there has been growing pressure to improve the correlations.

Grunwald and Winstein originally assumed that *tert*-butyl chloride, the model substrate, ionized without measureable nucleophilic solvent assistance. Hence, they assigned it a substrate sensitivity value, m, of 1.0. Bentley[4] has been the main advocate of the view that failure of some solvents, especially hindered fluorinated alcohols, to be correlated by eq 1 is evidence that *tert*-butyl chloride is experiencing nucleophilic solvent assistance from many hydroxylic solvents. Kevill's work lends support to this view.[5]

Raber and Harris[6] took advantage of the difference in the nucleophilicity of fluorinated and nonfluorinated alcohols in devising their probe for solvent nucleophilicity. However, Abraham, Kamlet, and Taft[7] have taken issue with the importance that Bentley, Harris, and Raber have given solvent nucleophilicity. They point out that their solvatochromatic method[8] suggests that differences in susceptibility to electrophilicity may be the cause of exceptional results.

Clearly, it seems likely that the Grunwald–Winstein treatment fails to satisfactorily deal with susceptibility of *tert*-butyl chloride to either electrophilicity or nucleophilicity. Extended Grunwald–Winstein treatments, which have parameters for ionizing power (or electrophilicity?) and nucleophilicity, have been proposed.[3,9,10] However, because of different susceptibilities to electrophilicity by the leaving group, comparisons with the same leaving group are required.[11] Despite this drawback and some failures in special cases,[12] the four-parameter approach of Bentley and Schleyer,[9] eq 2, is useful in evaluating solvent properties.

$$\log (k/k_0) = mY + lN \quad (2)$$

Kamlet and Taft's multiparameter approach, eq 3,[7,8] is

$$\log (k/k_0) = h\,\delta_H{}^2/100 + s\pi^* + a\alpha + b\beta \quad (3)$$

the most sophisticated approach currently undergoing scrutiny. Despite the enthusiasm for the solvatochromatic

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