1.08, $CHCl_3$).

To a suspension of CuBr (13 mg, 0.1 mmol) in ether (1 mL) at 0 °C was added allylmagnesium bromide (2.2 mL, 0.48 mmol), followed by the above-obtained tosyl-triflate **15** in ether (3 mL), and the reaction mixture was stirred at the same temperature for 3.5 h. Then n-Bu_2CuLi (1.4 mmol) in ether (3 mL) was introduced and the mixture was stirred at room temperature for 12 h. Usual workup followed by column chromatography gave 62 mg (58%) of **14**.

Preparation of L-Factor [(4*S*,5*S*)-(+)-5-Hydroxy-4-decanolide] (16). Through a solution of **14** (130 mg, 0.58 mmol) in 5 mL of MeOH cooled in a dry ice-acetone bath was passed a slightly excess of ozone. To the resulting pale blue solution was added dimethyl sulfide (1 mL), and the reaction mixture was slowly warmed to room temperature.

After evaporation of most of the solvent, the obtained crude aldehyde was dissolved in MeOH (5 mL) and treated with $AgNO_3$ (200 mg, 1.2 mmol) in H_2O (3 mL) and KOH (140 mg, 2.4 mmol) in H_2O (2 mL) at 0 °C for 0.5 h. At the end of the reaction the mixture was acidified with concentrated HCl and gently refluxed for 0.5 h. After removal of the insoluble substance by filtration through Celite, the filtrate was concentrated. The residue was poured into AcOEt and rinsed with saturated NaCl. Following solvent removal, the crude product was purified by column chromatography to give 83 mg (77%) of **16** as a colorless oil, which was crystallized in a refrigerator: mp 39–41 °C (lit.[30j] mp 42–44 °C); R_f 0.32 (ether); $[\alpha]^{20}_D$ +31.2° (c 0.96, $CHCl_3$) [lit.[30j] $[\alpha]^{21}_D$ +33.2° (c 1.11, $CHCl_3$)]; IR (neat) 3430, 2920, 2850, 1765, 1190, 780, 755; ^1H NMR 0.90 (3 H, t, J = 6.0 Hz), 1.2–1.8 (8 H, m), 1.9–2.7 (5 H, m), 3.3–3.7 (1 H, m), 4.42 (1 H, dt, J = 7.3, 4.4 Hz); ^{13}C NMR 13.89, 22.42, 23.95, 25.08, 28.58, 31.60, 32.85, 73.28, 82.92, 177.40; HRMS calcd for $C_{10}H_{18}O_3$ 186.1256, found 186.1263.

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Supplementary Material Available: Experimental data for **5**, **6**, **8–10**, and **14** (3 pages). Ordering information is given on any current masthead page.

Structure and Absolute Stereochemistry of the Epoxyquinol LL-C10037α and Related Metabolites from *Streptomyces* LL-C10037

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The structure of antibiotic LL-C10037α, produced by *Streptomyces* LL-C10037, was revised to the epoxyquinol **2** on the basis of a single-crystal X-ray diffraction analysis. Two other metabolites were isolated and characterized as 2-acetamido-4,5-dihydroxycyclohexenone and 2-acetamido-4-hydroxycyclohexenone. From application of the empirical "inverse quadrant rule" of Snatzke for epoxyquinols to the circular dichroism spectrum of **2**, the stereochemistry was inferred to be 4*S*,5*S*,6*S*. Preferring to rely on a non-empirical approach, the circular dichroism (CD) exciton chirality and X-ray crystallographic analyses of suitable derivatives were investigated to provide nonempirical approaches to establishing the absolute stereochemistry. Thus the 4-*p*-bromobenzoate of **2** was prepared, and the exciton chirality rules for interaction of the transition moments of the enone and benzoate chromophores were applied to the circular dichroism spectrum of this derivative, and a single-crystal X-ray diffraction analysis was carried out on the carbamate of **2** obtained with (*S*)-α-methylbenzylisocyanate. The exciton chirality analysis yielded 4*S* stereochemistry; thus, C-5 and C-6 also have the *S* configuration from the cis relationship established from the ^1H NMR spectrum. This absolute stereochemistry was also obtained from the X-ray diffraction analysis. The 4-*p*-bromobenzoates of **8** and **9** were also prepared, and exciton chirality analysis again indicated the 4*S* configuration for each.

Introduction

The antitumor metabolite LL-C10037α was isolated from *Streptomyces* LL-C10037 by researchers at Lederle Laboratories, and its gross structure was reported as **1**.[1] This structure represented the first reported occurrence of a γ-aminoepoxysemiquinone, and we wanted to explore its bioorganic implications.[2] As the initial part of our effort we repeated the spectral analysis with an authentic sample of LL-C10037α in order to confirm the structural assignment made earlier and to determine the relative configuration. In this paper data are presented for revising the structure of LL-C10037α, for establishing the structures of two additional metabolites produced by the same organism, and for defining the absolute stereochemistry of each metabolite.

(1) Lee, M. D.; Fantini, A. A.; Morton, G. O.; James, J. C.; Borders, D. B.; Testa, R. T. *J. Antibiot.* **1984**, *37*, 1149.

(2) Initial biosynthesis results have already been reported: Gould, S. J.; Shen, B.; Whittle, Y. G. *J. Am. Chem. Soc.* **1989**, *111*, 7932.

Table I

H	^1H NMR (400 MHz, DMSO-d_6) δ	^1H NMR (400 MHz, DMSO-d_6) multiplicity, J, Hz	^{13}C NMR (100.6 MHz, DMSO-d_6) δ
1			189.6
2			128.3
3	7.04	dd, J = 2.5, 2.7	128.3
4	4.79	ddd, J = 2.7, 3.1, 6.4	63.3
5	3.77	ddd, J = 2.5, 3.1, 4.2	53.7
6	3.55	d, J = 4.2	52.2
1′			169.5
2′ (CH_3)	2.04	s	23.7
OH	5.79	d, J = 6.4	
NH	9.04	br s	

Results and Discussion

Relative Stereochemistry of 2. The ^1H NMR data acquired by us for LL-C10037α were identical to that reported in the literature[1] (Table I). The only difference was in the interpretation of the chemical shifts of the two

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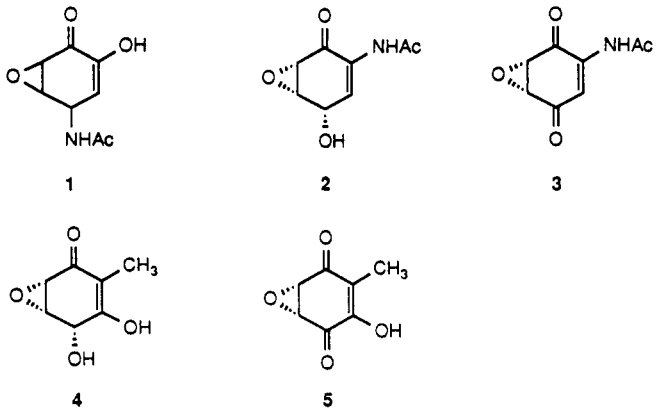
Table II

compound	$[\alpha]^{20}_D$	ΔE, R band (wavelength, nm)	ΔE, K band (wavelength, nm)
2	−202	−5.59 (326)	+3.10 (246)
epoxydon[5]	+93	+4.70 (341)	−5.86 (245)
4[8]	−269	−1.64 (318)	−0.42 (265)
desoxyepiepoxydon[9]	+221	+1.91 (340)	+3.5 (254)
chaloxone[10]	+271	+2.88 (330)	+7.5 (270)

acidic protons in the spectrum acquired in DMSO-d_6. Whereas we assigned the doublet at 5.8 ppm (J = 6.4 Hz) to the OH proton and the broad singlet at 9.0 ppm to the NH proton, the previous workers made the reverse assignments. Based on our interpretation of the NMR data, LL-C10037α should be 2-acetamido-4-hydroxy-5,6-epoxyquinol, **2**. This correction was confirmed by X-ray crystallography, which also indicated that the atoms C-2, C-3, C-5, and C-6 are roughly in a plane and C-1 and C-4 are both displaced to the same side of this plane. The hydroxyl group extended in an equatorial direction and bore a cis relationship with the oxygen of the oxirane ring. Thus, **2** would be the cis equatorial stereoisomer shown, or its mirror image. Remarkably, therefore, **2**, $[\alpha]^{20}_D$ −202° (*c* 0.334, MeOH), has the same gross structure and relative stereochemistry as (+)-MT35214, $[\alpha]^{20}_D$ +104° (*c* 1, MeOH),[3] obtained by acetylation of antibiotic MM14201 produced by *Streptomyces* sp. NCIB 11813.[3] The difference in the sign of their specific rotations indicates that they apparently form an enantiomeric pair.[4]

2 was oxidized to the epoxyquinone **3**, $[\alpha]^{20}_D$ +115.6° (*c* 0.5, MeOH), and its specific rotation compared to that which was reported for the corresponding epoxyquinone MT36531, $[\alpha]^{20}_D$ −99° (*c* 0.5, MeOH).[3] The same trend was observed,[4] providing further evidence in support of the enantiomeric relationship.



Absolute Stereochemistry of 2. A number of other epoxyquinols have been isolated from natural sources: (+)-epoxydon,[5] isoepoxydon,[6,7] panepoxydon,[6] (−)-terremutin,[8] desoxyepiepoxydon,[9] and chaloxone.[10] Generally, the absolute stereochemistry of the epoxide ring has been determined from circular dichroism (CD) data. An empirical correlation of the sign of the R band at approximately 340 nm of an epoxyquinol with compounds of known absolute stereochemistry resulted in formulation of an "inverse quadrant" rule, with the sign of the R band dictated by the octant in which the oxirane oxygen atom lies.[11] In Table II the CD data for **2** and some of these other metabolites are given. Since the R band of **2** is negative (ΔE_{326} −5.59), similar to terremutin, **4** (ΔE_{341} −1.64), **2** should have the oxirane oxygen lying below the plane of the cyclohexenone ring, as shown. Having previously established that **2** is a cis stereoisomer, its absolute stereochemistry would be 4*S*,5*S*,6*S*. However, although numerous epoxyquinols have been assigned in this manner, we were not content to rely solely on such an empirical correlation.

Table III

compound	ΔE (wavelength, nm)
3	+6.78 (364), −10.00 (308), +6.29 (242), −6.61 (224)
5[13]	−1.34 (351), +1.87 (313)
G-7063-2[14]	−6.58 (376), +10.53 (327)

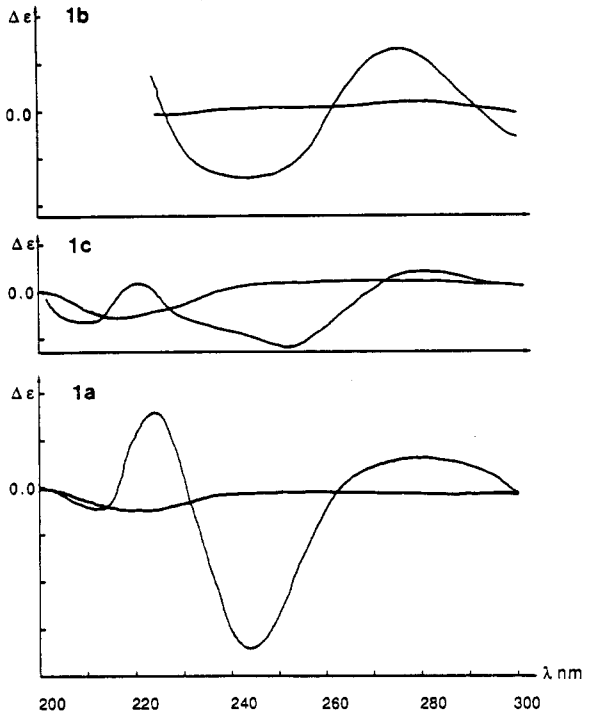


Figure 1. (a) CD spectrum of **6** in methanol. (b) CD spectrum of **10** in methanol. (c) CD spectrum of **11** in methanol.

The epoxyquinone **3** was also analyzed by CD, and the data were compared with that of other epoxyquinones in the literature. A standard has been terreic acid, **5**, whose absolute stereochemistry was established by chemical correlation with **4**.[8,12] Such compounds exhibit two Cotton effects for n → π* transitions between 300 and 400 nm. These transitions have been associated with the two individual C=O chromophores, and the difference in the band positions has been ascribed to the transitions from the energetically higher n orbital of the two carbonyl

(3) Box, S. J.; Gilpin, M. L.; Gwynn, M.; Hanscomb, G.; Spear, S. R.; Brown, A. G. *J. Antibiot.* **1983**, *36*, 1631.
(4) The differences in the absolute magnitudes are apparently due to purity of the samples; the Beecham group had relatively little to work with (M. L. Gilpin, private communication).
(5) (a) Closse, A.; Mauli, R.; Sigg, H. P. *Helv. Chim. Acta* **1966**, *49*, 204–213. (b) Sakamura, S.; Niki, H.; Obata, Y.; Sakai, R.; Matsumoto, T. *Agric. Biol. Chem.* **1969**, *33*, 698–703.
(6) Kis, A.; Closse, A.; Sigg, H. P.; Hruban, L.; Snatzke, G. *Helv. Chim. Acta* **1970**, *53*, 1570–1597.
(7) Sekiguchi, J.; Gaucher, G. M. *Biochem. J.* **1979**, *182*, 445.
(8) Read, G.; Ruiz, V. M. *J. Chem. Soc. C* **1970**, 1945–1948.
(9) Nagasawa, H.; Suzuki, A.; Tamura, S. *Agric. Biol. Chem.* **1978**, *42*, 1303–1304.
(10) Fex, T.; Wickberg, B. *Acta Chem. Scand. B* **1981**, *35*, 97–98.
(11) Snatzke, G.; Snatzke, F. In *Fundamental Aspects and Recent Developments in ORD and CD*; Ciardelli, F., Salvadori, P., Eds.; Heyden and Sons: London, 1973; pp 109–121.
(12) Miller, M. W. *Tetrahedron* **1968**, *24*, 4839–4851.

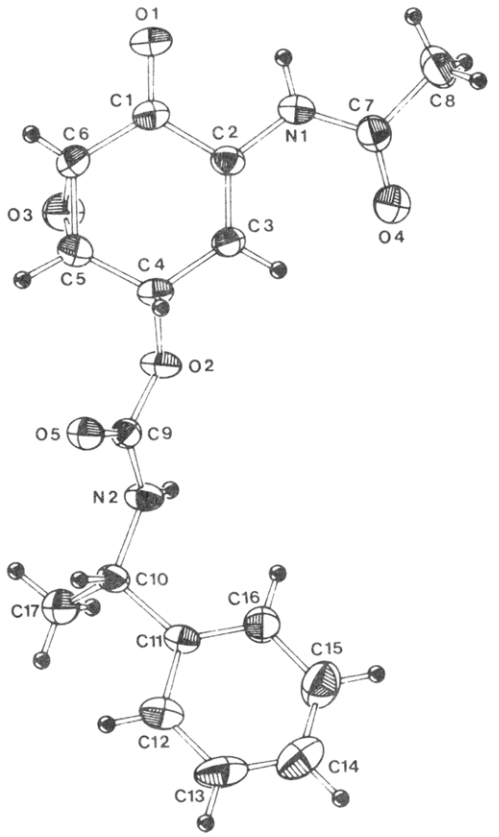
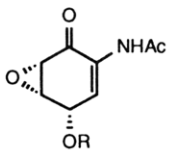


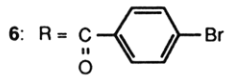
Figure 2. X-ray crystal structure of **7**.

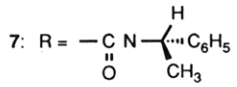
groups into the π^* orbital.[13] Additionally, there may be some intramolecular hydrogen bonding at one end of the quinone system.[13,15]

The sign of the CD spectrum of **3** (Table III) is opposite that of **5**, indicating that the former has 5*R*,6*S* stereochemistry, consistent with that assigned to **2**. However, both assignments are based on the same underlying empirical rule since **5** was assigned by comparison with **4**. We therefore chose to establish the absolute stereochemistry unequivocally with a nonempirical method:[16] either the exciton chirality method, pioneered by Nakanishi,[17] or single-crystal heavy-atom X-ray crystallographic analysis.

The *p*-bromobenzoate **6** was prepared in 75% yield by treating **2** with *p*-bromobenzoyl chloride, triethylamine, and a catalytic amount of (dimethylamino)pyridine (DMAP) in tetrahydrofuran (THF). Unfortunately, an acceptable crystal for the X-ray analysis could not be obtained. However, the CD spectrum of **6** (Figure 1a) showed a split Cotton effect (ΔE_{244} −9.2 and ΔE_{280} +1.9), which is due to the interaction between the bromobenzoate and the enone chromophores. Applying the exciton chirality rule,[17] this negative first Cotton effect corresponds to a negative chirality, and the projection of the two chromophores should be counterclockwise; the resulting absolute stereochemistry is that shown.

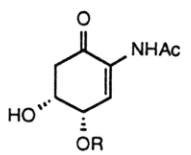


6: R = 

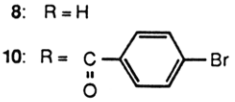
7: R = 

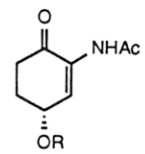
Simultaneously, we prepared a number of urethanes of **2** with optically active isocyanates. One of these, **7**, obtained by reaction with (*S*)-(−)-α-methylbenzyl isocyanate in THF at reflux was carefully recrystallized from toluene. This yielded a crystal suitable for X-ray analysis. The ORTEP drawing in Figure 2 clearly shows the same 4*S*,5*S*,6*S*,10*S* stereochemistry.

Structure and Absolute Stereochemistry of LL-C10037β and LL-C10037γ. Two additional metabolites of *Streptomyces* LL-C10037 have been isolated and purified by column chromatography on silica gel. One, more polar than **2**, has been named LL-C10037β, and the other, slightly less polar than **2**, has been named LL-C10037γ. The UV and IR spectra indicated that each was also a 2-acetamidocyclohexenone. From the presence of one methylene adjacent to the ketone carbonyl (δ 2.00 and 2.78), three exchangeable hydrogens, and two carbinol protons (δ 4.17 and 4.58), the diol structure **8** was assigned to the more polar metabolite. The cis stereochemistry was derived from the H-4/H-5 coupling constant (3.4 Hz). The proton NMR spectrum of the third compound contained resonances from two adjacent methylenes, one next to the ketone, and structure **9** was therefore assigned. The $^1H/^1H$ COSY spectra of each contained all cross peaks consistent with these structures.

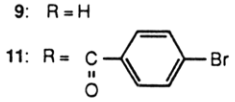


8: R = H

10: R = 



9: R = H

11: R = 

In order to determine the absolute stereochemistry of each of these new metabolites, the *p*-bromobenzoates **10** and **11** were prepared. The CD spectra of each (Figure 1, parts b, c) displayed the same negative split Cotton effect that had been observed for **6**. Therefore, applying the exciton chirality rules confirmed the biogenetic expectation that these compounds have the same absolute stereochemistry as **2**.

Conclusions

Antibiotic LL-C10037α has now been shown to have the expoxyquinol structure **2**. Two additional related metabolites of *Streptomyces* LL-C10037 were isolated and characterized as **8** and **9**.

While numerous other naturally occurring epoxyquinols have been reported over a 22-year period, until recently[16]—while our work was in progress—in no case had the absolute stereochemistry of any of these been established by an unambiguous, nonempirical analysis. Absolute stereochemistry for the epoxide carbons had only been inferred from empirical correlations of the signs of Cotton effects in the circular dichroism spectra ("inverse quadrant rule").[11]

We have prepared the carbamate **7** of **2** and (*S*)-(−)-α-methylbenzyl isocyanate and analyzed a single-crystal by X-ray diffraction, unambiguously yielding the absolute

(13) Thiericke, R.; Stellwaag, M.; Zeeck, A.; Snatzke, G. *J. Antibiot.* **1987**, *42*, 1549–1554.
(14) Noble, M.; Noble, D.; Sykes, R. B. *J. Antibiot.* **1977**, *30*, 455.
(15) Read, G. *J. Chem. Soc.* **1965**, 6587–6589.
(16) The structure and absolute stereochemistry of a marine epoxyquinol have recently been reported with—to our knowledge—the first instance where both CD analysis (empirical "inverse quadrant rule") and X-ray crystallographic analysis have been done on the same compound: Higa, T.; Okuda, R. K.; Severns, R. M.; Scheuer, P. J.; He, C. H.; Changfu, X.; Clardy, J. *Tetrahedron* **1987**, *43*, 1063–1070.
(17) Harada, N.; Nakanishi, K. *Circular Dichroic Spectroscopy, Exciton, Coupling in Organic Stereochemistry*; Oxford University Press: Oxford, 1983; pp 238–247.

stereochemistry of **2** as 4*S*,5*S*,6*S*. We also prepared the *p*-bromobenzoate **6** of **2**, as well as those—**10** and **11**—of **8** and **9**, respectively. The CD spectra of these derivatives were analyzed for the interactions of the enone and benzoate transition moments; this nonempirical use of circular dichroism also yielded 4*S* absolute stereochemistry for all three.

In this study as well as Scheuer's,[16] application of the "inverse quadrant rule" to the CD spectra of the parent epoxyquinols yielded the correct absolute configuration for C-5/C-6. Thus, this rule may now be more reliably invoked.

Work is now proceeding toward isolation of the epoxidases from *Streptomyces* LL-C10037[2] and NCIB 11813 that generate the enantiomeric epoxides of **2** and (+)-MT35214, respectively.

Experimental Section

General Procedures. ^1H NMR spectra (400 MHz) and ^{13}C NMR spectra (100.6 MHz) were taken on a Bruker AM 400 spectrometer. All ^{13}C NMR spectra were broadband decoupled. Five-millimeter NMR tubes were used for all NMR measurements. ^1H and ^{13}C NMR samples were referenced with TMS or *t*-BuOH. IR spectra were recorded on a Nicolet 5DXB FTIR spectrometer. Low-resolution mass spectra were taken on a Varian MAT CH-7 spectrometer. High-resolution mass spectra were taken on a Kratos MS 50 TC spectrometer.

UV spectra were recorded on a IBM 9420 UV-visible spectrophotometer, and CD spectra were recorded on a Durrum JASCO Model J-10 circular dichroism spectrometer. X-ray crystal diffraction analysis was carried out on a Rigaku AFC6R diffractometer.

Melting points were taken on a Büchi melting point apparatus and are uncorrected. Flash chromatography was carried out on silica gel (EM Reagents, Keiselgel 60, 230–400 mesh). Silicar CC-4 was purchased from Mallinckrodt. Analytical thin-layer chromatography (TLC) was carried out on precoated Keiselgel 60 F_{254} (either 0.2-mm aluminum sheets or 0.25-mm glass plates) and visualized by long- and/or short-wave UV. Anhydrous solvents were prepared by distillation over sodium or lithium aluminum hydride. (*S*)-(−)-α-methylbenzyl isocyanate was purchased from Aldrich.

Standard Culture Conditions. *S.* LL-C10037 was maintained at 5 °C as spores on sterile soil. A loopful of this material was used to inoculate 50 mL of seed medium containing 1.0% glucose, 2.0% soluble potato starch, 0.5% yeast, 0.5% N-Z Amine A 59027, and 0.1% $CaCO_3$ in glass distilled water, all adjusted to pH 7.2 with 2% KOH. The seed inoculum, contained in a 250-mL Erlenmeyer flask, was incubated for 3 days at 28 °C, 200 rpm. Production broths (200 mL in 1-L Erlenmeyer flasks), consisting of 1.0% glucose, 0.5% bactopeptone, 2.0% molasses (Grandma's Famous light unsulfured), and 0.1% $CaCO_3$ in glass distilled water and adjusted to pH 7.2 with 10% HCl prior to sterilization, were subsequently inoculated 5% v/v with vegetative inoculum from seed broths. The production broths were incubated for 120 h.

Isolation. The fermentation was filtered through cheesecloth and Celite. The filtrate was adjusted to pH 4.7 with solid KH_2PO_4 saturated with $(NH_4)_2SO_4$ and extracted repeatedly with EtOAc (typically eight times). After concentration in vacuo, the residue was dissolved in a minimum volume of methanol and adsorbed onto a small quantity of silica gel. This was applied to the top of a column of flash grade silica gel (25 g/200 mL fermentation) prepared in 40% hexane/EtOAc. After low polarity colored impurities had been eluted, the solvent was changed to 20% hexane/EtOAc and elution yielded **2**, which was recrystallized from methanol. Once **2** was eluted, the solvent was changed to EtOAc. The fractions containing **8** were pooled and concentrated to dryness. The residue was further purified by preparative silica gel TLC plates (2 mm, 20 × 20 cm) developed with $CHCl_3$/CH_3OH/AcOH (92:7:1). The band containing **8** was collected and eluted with EtOAc. The eluted EtOAc solution was concentrated in vacuo to give **8**, which was recrystallized from EtOAc. The fractions containing **9** were pooled and concentrated to yield **9**, which was recrystallized from EtOAc–methanol.

2-Acetamidoepoxyquinone 3. To 50 mL of a CH_2Cl_2 solution containing **2** (200 mg, 1.1 mmol) was added NaOAc (90 mg, 1.1 mmol) and PCC (355 mg, 1.65 mmol). The resulting solution was stirred at room temperature for 1.5 h. The brown reaction mixture was then filtered through a Celite pad, and the residue was washed with CH_2Cl_2. The combined CH_2Cl_2 filtrate was concentrated in vacuo to give a brown residue, which was further purified on a silica gel column (1.5 × 15 cm) eluted with CH_2Cl_2. The fractions containing the product were combined and concentrated in vacuo to provide a yellow solid. After recrystallization from EtOAc, 100 mg of bright yellow crystals were obtained in 50% yield: mp 135–136 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 7.89 (1 H, bs), 7.51 (1 H, d, *J* = 2.2 Hz), 3.91 (1 H, d, *J* = 3.7 Hz), 3.83 (1 H, dd, *J* = 3.7, 2.2 Hz), 2.22 (3 H, s); $[\alpha]^{22}_D$ = +115.6° (*c*, 0.5 in MeOH).

***p*-Bromobenzoate of 2 (6).** To a cold (0 °C) stirred solution of **2** (40.0 mg, 0.219 mmol) and triethylamine (24.3 mg, 0.24 mmol) in THF (2.0 mL) was slowly added *p*-bromobenzoyl chloride (60.0 mg, 0.273 mmol) in THF (1.0 mL). DMAP (1.4 mg, 0.012 mmol) in THF (0.7 mL) was then added, and the reaction solution was allowed to warm to room temperature. After 17 h, the reaction was quenched and extracted with $CHCl_3$ (5 × 3.0 mL). The combined organic solution was washed with H_2O (2 × 1.0 mL), dried over Na_2SO_4, and evaporated to give a white solid (85.0 mg). Recrystallization from hexane/EtOAc afforded 70 mg of **6** in 75% yield as colorless needles: mp 155–157 °C; IR (KBr) 3290, 1738, 1682, 1528, 1312, 1265, 1085, 1013, 877, 752 cm^{-1}; UV max (ε) 201 (19600), 246 nm (14700); ^1H NMR (400 MHz, acetone-d_6) δ 2.21 (s, 3 H), 3.66 (d, 1 H, *J* = 4.3 Hz), 4.13 (ddd, 1 H, *J* = 2.6, 2.9, 4.3 Hz), 6.25 (dd, 1 H, *J* = 2.9, 3.0 Hz), 7.40 (dd, 1 H, *J* = 2.9, 3.0 Hz), 8.02, 8.13 (AA'BB', 4 H, *J* = 8.7 Hz), 8.4 (bs, 1 H); ^{13}C NMR (100.6 MHz, acetone-d_6) δ 189.3, 170.3, 165.6, 132.9, 132.4, 131.2, 129.7, 128.9, 120.5, 66.8, 52.8, 52.1, 24.2; CD (CH_3OH) Δε(330 nm) = −2.49, Δε(280 nm) = +1.9, Δε(244 nm) = −9.2, Δε(223 nm) = +6.4; low-resolution mass spectrum (EI) 186.0, 185.0, 184.0, 183.0 (100), 157.0, 155.0, 140.0, 124.0, 110.0, 43.0. Anal. Calcd: C, 49.20; H, 3.30; N, 3.83. Found: C, 49.23; H, 3.06; N, 3.75.

8: mp 160.5–161.0 °C; IR (KBr) 3431, 3427, 3355, 3311, 1670, 1664, 1656, 1650, 1533, 1529, 1375 cm^{-1}; UV max (ε) 265 (52700), 209 mn (81800); ^1H NMR (400 MHz, methanol-d_4) δ 2.10 (s, 3 H), 2.67 (dd, 1 H, *J* = 3.5, 16.6 Hz), 2.78 (dd, 1 H, *J* = 6.5, 16.6 Hz), 4.18 (dddd, 1 H, *J* = 1.3, 3.4, 3.5, 6.5 Hz), 4.59 (dd, 1 H, *J* = 3.4, 3.8 Hz), 7.51 (dd, 1 H, *J* = 1.3, 3.8 Hz); ^{13}C NMR (100.6 MHz, methanol-d_4) δ 193.7, 172.2, 133.7, 129.8, 70.1, 68.6, 42.9, 23.9; low-resolution mass spectrum (EI) 185.0, 156.0, 144.0, 143.0, 125.0, 114.0, 96.0, 71.0, 70.0 (100); $[\alpha]^{22}_D$ = +26.3° (*c* 0.268, in MeOH). Anal. Calcd: C, 51.93; H, 5.95; N, 7.57. Found: C, 51.93; H, 5.95; N, 7.51.

9: mp 122.5–123.5 °C; IR (KBr) 3350, 3329, 1687, 1676, 1649, 1629, 1560, 1540, 1535, 1373, 1343 cm^{-1}; UV max (ε) 265 (51500), 212 nm (71800); ^1H NMR (400 MHz, methanol-d_4) δ 1.89 (dddd, 1 H, *J* = 4.6, 8.8, 12.3, 17.0 Hz), 2.09 (s, 3 H), 2.27 (ddddd, 1 H, *J* = 1.2, 4.7, 4.8, 4.9, 17.0 Hz), 2.46 (ddd, 1 H, *J* = 4.7, 12.3, 17.0 Hz), 2.53 (ddd, 1 H, *J* = 4.6, 4.8, 17.0 Hz), 4.60 (ddd, 1 H, *J* = 2.9, 4.9, 8.8 Hz), 7.65 (dd, 1 H, *J* = 1.2, 2.9 Hz); ^{13}C NMR (100.6 MHz, methanol-d_4) δ 194.9, 172.2, 134.9, 133.0, 66.5, 35.1, 32.7, 29.3; low-resolution mass spectrum (EI) 169.0, 127.0, 126.0 (100), 110.0, 98.0, 82.0, 71.0, 70.0, 53.0; $[\alpha]^{22}_D$ = +20.3° (*c* 0.249, in MeOH). Anal. Calcd: C, 56.83; H, 6.56; N, 8.28. Found: C, 57.05; H, 6.33; N, 8.38.

***p*-Bromobenzoate of 8 (10).** To a cold (0 °C) stirred solution of **8** (43.0 mg, 0.232 mmol) and triethylamine (23.5 mg, 0.232 mmol) in THF (2.0 mL) was slowly added *p*-bromobenzoyl chloride (51.0 mg, 0.232 mmol) in THF (1.0 mL). DMAP (1.0 mg) in THF (0.05 mL) was then added, and the reaction solution was allowed to warm to room temperature. After 28 h, the reaction mixture was cooled, diluted with CH_2Cl_2 (2.0 mL), and quenched by adding crushed ice. The organic layer was separated; the remaining aqueous solution was extracted with EtOAc (3 × 2.0 mL). The combined organic solution was dried over Na_2SO_4 and concentrated in vacuo to afford 94.0 mg of a solid, which was further purified on a Silicar CC-4 column eluting with 50% hexane/EtOAc to afford 22.0 mg (26% yield) of a glass: IR (KBr) 3442, 3349, 1720, 1675, 1590, 1516, 1372, 1328, 1267, 1104, 1008, 756 cm^{-1}; UV max (ε) 245 nm (17000); ^1H NMR (400 MHz, acetone-d_6) δ 2.13 (s, 3 H), 2.90 (dd, 1 H, *J* = 7.8, 16.8 Hz), 2.96

(dd, 1 H, J = 4.0, 16.8 Hz), 4.58 (1 H, m), 4.71 (1 H, d, J = 4.7 Hz), 5.99 (1 H, dd, J = 3.5, 3.8 Hz), 7.60 (1 H, bd, J = 3.8 Hz), 7.71, 8.04 (AA'BB', 4 H, J = 8.4 Hz), 8.39 (bs, 1 H); ^{13}C NMR (100.6 MHz, acetone-d_6) δ 192.6, 169.9, 165.7, 135.1, 132.6, 132.4, 130.3, 128.5, 121.5, 72.4, 67.8, 42.9, 24.3; CD (CH_3OH) $\Delta\epsilon$(315 nm) = -0.97, $\Delta\epsilon$(274 nm) = +1.46, $\Delta\epsilon$(240 nm) = -1.70; low-resolution mass spectrum (EI) 202.0, 200.0, 185.0, 183.0 (100), 167.0, 157.0, 155.0, 125.0, 43.0.

p-Bromobenzoate of 9 (11). To a cold (0 °C) stirred solution of **10** (28.0 mg, 0.167 mmol) in CH_2Cl_2 (5.0 mL) were added triethylamine (18.9 mg, 0.20 mmol), p-bromobenzoyl chloride (43.98 mg, 0.20 mmol) in THF (1.0 mL), and DMAP (1.0 mg). The reaction solution was allowed to warm to room temperature. After 30 h, the reaction mixture was cooled and quenched by adding crushed ice. The organic layer was separated, and the remaining aqueous solution was extracted with CH_2Cl_2 (3 × 2.0 mL). The combined organic solution was washed with H_2O (3 × 1.0 mL) and dried over Na_2SO_4. The dried extract was concentrated in vacuo to afford 64.0 mg of a solid, which was further purified on a silica gel 60 column (10 × 1.5 cm), eluting with 50% hexane/EtOAc to give 24.0 mg of partially pure (**11**). This material was chromatographed again with 25% hexane/EtOAc as eluting solvent to afford 12.0 mg (20% yield) of a colorless solid: mp 96–99 °C; IR (KBr) 3349, 1718, 1684, 1667, 1591, 1508, 1323, 1282, 1289, 1245, 1118, 1105, 897, 847, 757 cm^{-1}; UV max (ϵ) 240 (30600), 246.8 nm (24800); ^{1}H NMR (400 MHz, acetone-d_6) δ 2.11 (s, 3 H), 2.2 (m, 1 H), 2.5 (m, 1 H), 2.64 (ddd, J = 4.7, 9.9, 17.3 Hz), 2.80 (ddd, 1 H, J = 4.8, 7.0, 17.2 Hz), 5.92 (ddd, J = 3.8, 4.2, 7.6 Hz), 7.78 (d, 1 H, J = 3.7 Hz), 7.72, 7.98 (AA'BB', 4 H, J = 8.3 Hz), 8.39 (bs, 1 H); ^{13}C NMR (100.6 MHz, acetone-d_6) δ 193.4, 169.9, 165.5, 134.8, 132.7, 132.2, 130.3, 128.5, 124.9, 69.7, 34.3, 34.0, 28.7; CD (CH_3OH) $\Delta\epsilon$(280 nm) = +0.55, $\Delta\epsilon$(252 nm) = -3.79, $\Delta\epsilon$(220 nm) = +2.01; high-resolution mass spectrum (EI) calcd 351.01062, 353.00862, found 351.01040, 353.00860; low-resolution mass spectrum (EI) 353.0, 351.0, 311.0, 309.0, 202.0, 200.0, 185.0 (100), 183.0, 167.8, 157.0, 155.0, 126.0, 110.0, 109.0, 43.0.

(S)-(-)-α-Methylbenzylurethane of 2 (7). To a dry 10.0-mL two-necked round-bottomed flask, equipped with a condenser and a septum cap, a solution of **2** (44 mg, 0.24 mmol) in THF (4.0 mL) and (S)-(-)-α-methylbenzyl isocyanate (35.4 mg, 0.24 mmol) were introduced via the septum cap. The resulting solution was heated at reflux under a nitrogen atmosphere for 24 h. The wine-colored reaction mixture was diluted with CH_2Cl_2 (20.0 mL) and decolorized with active charcoal. Filtration and concentration in vacuo yielded a residue that was recrystallized from CH_2Cl_2 to give white fine needles of **7** (76 mg, 96%): mp 173–175 °C; IR (KBr) 3339, 3320, 1692, 1534, 1373, 1243, 1063, 875, 702 cm^{-1}; UV max (ϵ) 274 (43000), 218 nm (72000); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.9 (1 H, s, exch), 7.2–7.4 (6 H, m), 6.0 (1 H, d, J = 7.4 Hz, exch), 5.8 (1 H, m), 4.9 (1 H, d, J = 7.2 Hz), 4.0 (1 H, m), 3.6 (1 H, d, J = 3.8 Hz), 2.1 (3 H, s), 1.5 (3 H, d, J = 6.8 Hz); ^{13}C NMR δ (100.6 MHz, $CDCl_3$) 188.3, 168.0, 154.3, 142.9, 129.2, 128.8, 127.6, 125.9, 121.3, 66.5, 51.6, 51.4, 51.1, 24.6, 22.4; $[\alpha]^{20}_D$ = -125.6° (c 0.05, in MeOH). Anal. Calcd: C, 61.81; H, 5.49; N, 8.48. Found: C, 61.56; H, 5.44; N, 8.18.

X-ray Work on $C_{17}NO_5H_{17}$ (7). A crystal of dimensions 0.20 × 0.10 × 0.05 mm was used for collection of data. Unit cell parameters were refined from a least-squares analysis of the angle settings of 13 reflections in the range 22° < 27° < 35°. Intensity data were collected with the ω-2θ scan technique and a scan speed of 32° min^{-1} in ω. The intensities of three standard reflections monitored throughout the data collection exhibited an average fluctuation of 2.1%. From 1802 reflections measured to (sin $\theta/\lambda)_{max}$ = 0.5947 Å^{-1} with the range of indices $0 \le h \le 8$, $0 \le k \le 43$, and $0 \le l \le 5$, 1235 unique data having $F_o^2 \ge 3\sigma(F_o^2)$ were obtained.

All calculations were performed on a μVAX II computer with programs from the TEXRAY crystallographic software package.[18] Atomic positions for all non-hydrogen atoms were derived from the direct methods program MITHRIL.[19] Hydrogen atoms attached to C atoms were placed in calculated positions (C-H = 0.95 Å) and of the attached C atom. Following two cycles of least-squares refinement, the remaining hydrogen atoms, H(8) and H(11), were located from a difference electron density map; the positional parameters and an isotropic thermal parameter were subsequently refined for each of these atoms. Final refinement of F_o with 224 variable, 1235 observations, and $F_o^2 > 3\sigma(F_o^2)$ affords the residuals R = 0.038 and R_w = 0.049, where the weights are derived from counting statistics and a value of p = 0.05. In the final cycle Δ/σ - 0.01 and the maximum excursion in the final difference electron density map = 0.32 e Å^{-3}. The data were not corrected for absorption.

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Kuanoniamines A, B, C, and D: Pentacyclic Alkaloids from a Tunicate and Its Prosobranch Mollusk Predator *Chelynotus semperi*

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From a Micronesian tunicate and its predator, a prosobranch mollusk *Chelynotus semperi*, we have isolated five alkaloids, the known shermilamine B (**1**) and four new pentacyclic compounds, kuanoniamines A–D (**2–5**). The structures were established by extensive NMR analysis and correlations. Cytotoxicity (IC_{50}) against KB cells ranged from >10 μg/mL for **3** to 5 μg/mL for **1** and **5** to 1 μg/mL for **2**.

The sequestering of selective metabolites by opisthobranch mollusks from dietary sources has been an extensively studied area of chemical marine ecology.[1] Similar studies of mollusks from other gastropod subclasses are,

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