New Diterpenes from the South African Soft Coral *Eleutherobia aurea*

Gregory J. Hooper,[†] Michael T. Davies-Coleman,*[,†] and Michael Schleyer[‡]

Department of Chemistry, Rhodes University, Grahamstown 6140, South Africa, and Oceanographic Research Institute, Durban, South Africa

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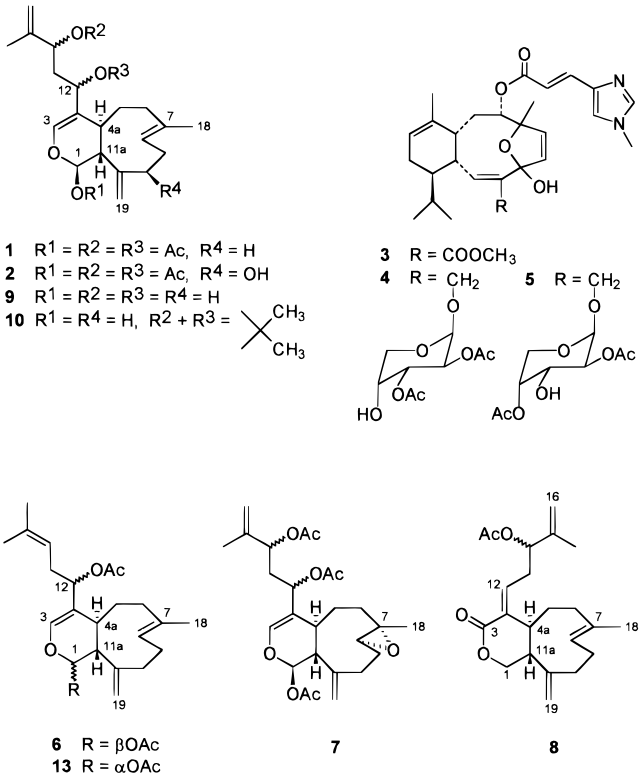
The known diterpenoids zahavin A and 9-deacetoxy-14,15-deepoxyxeniculin and the two new diterpenoids 7,8-epoxyzahavin A and xeniolide C were isolated from specimens of *Eleutherobia aurea* collected from Aliwal Shoal off the southern Kwazulu−Natal coast, South Africa. Standard spectroscopic methods were used for the structure determinations. The former three diterpenoids inhibit superoxide production in rabbit-cell neutrophils.

Both temperate and subtropical South African soft corals have proved to be a good source of new bioactive metabolites.[1−4] In continuation of our search for new antiinflammatory diterpenes from these organisms[2] we have examined specimens of *Eleutherobia aurea* Benayahu and Schleyer (1995),[5] collected from the Aliwal Shoal, an extensive reef system off the southern Kwazulu−Natal coast, South Africa. Interestingly, the taxonomy of this organism has recently been the subject of some debate.[3,4] The Indo-Pacific genera *Alcyonium* and *Eleutherobia* are taxonomically very similar, and this soft coral, originally identified as the new species *A. aureum*,[3] has now been reclassified as the new species *E. aurea*.[4,5] Although the chemistry of soft corals from the genus *Alcyonium* is well documented,[6] very little is known about the natural product constituents of the genus *Eleutherobia*.[4] Kashman *et al.* have recently examined the natural product chemistry of *E. aurea* collected from Sodwana Bay, on the northern Kwazulu−Natal coast and have found a surprising intraspecific variation in diterpene metabolites even from a single colony of these organisms.[3,4] The compounds isolated thus far from the Sodwana Bay specimens of *E. aurea* include the new xenicane diterpenes zahavins A (**1**) and B (**2**),[3] sarcodictyin A (**3**), and two new glycosides related to **3**, eleuthosides A (**4**) and B (**5**).[4] Our chemical and bioactivity investigations of specimens of *E. aurea* from the Aliwal Shoal (approximately 500 km south of Sodwana Bay) are presented in this paper.



Results and Discussion

Specimens of the small, bright yellow soft coral with prominent white polyps, *E. aurea*, were collected by scuba from the Aliwal Shoal during winter 1994. The frozen soft coral was freeze dried and steeped in EtOAc. Initial Si gel flash chromatography of a portion of the EtOAc extract, followed by exhaustive HPLC on both normal and reversed-phase columns, yielded **1** (0.04% dry wt), 9-deacetoxy-14,15-deepoxyxeniculin (**6**, 0.12% dry wt), and two new compounds, 7,8-epoxyzahavin A (**7**, 0.03% dry wt) and xeniolide C (**8**, 0.01% dry wt).

The molecular formula of **1**, isolated as a colorless oil ($[\alpha]_D$ +21°, lit.[3] +7.3°), was established as $C_{26}H_{36}O_7$ from HREIMS data, while the ^{1}H- and ^{13}C-NMR spectral data for this compound were consistent with those reported for zahavin A.[3] Interestingly, no evidence of the C-10 hydroxylated analog of **1**, zahavin B (**2**), was found in the EtOAc extract of the Aliwal Shoal specimens of *E. aurea*. The relative configuration of the two exocyclic chiral centers in **1** has not been established, and a logical route to ascertain this stereochemistry appeared to be through initial saponification of **1** to give the triol **9**, followed by preparation of the isopropylidene or acetonide derivative (**10**) of **9**. The chemical shifts of the two acetonide methyl groups in the ^{13}C-NMR spectrum of **10** would then establish either a chair (δ 20 and 30) or a twist-boat (both δ 25) conformation for the acetonide ring and thus indicate either a *syn* or *anti* relative stereochemistry, respectively, for the exocyclic diol.[7] Several attempts at mild saponification of **1** were unsuccessful, and instead of isolating the expected saponification product, **9**, from the complex reaction mixture, only the rearranged compound (**11**) was obtained in low yield. The structure of this compound was established as follows.

HREIMS data established the molecular formula of **11** as $C_{20}H_{26}O_2$. The presence of a single aldehyde carbonyl and ten olefinic resonances in the ^{13}C-NMR

* To whom correspondence should be addressed. Phone: 27-461-318264. Fax: 27-461-25109. E-mail: chdc@warthog.ru.ac.za.
† Department of Chemistry.
‡ Oceanographic Research Institute.
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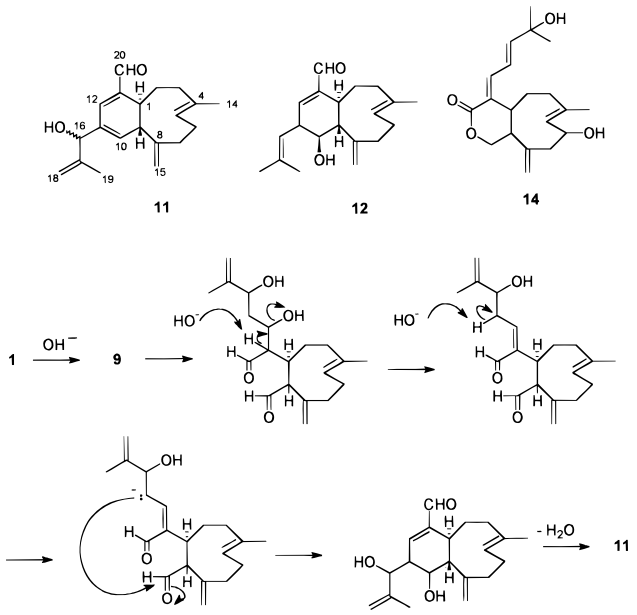
Scheme 1. Proposed rearrangement of **1** under basic conditions, to give **11**.



spectrum of **11** accounted for six of the eight degrees of unsaturation implied by the molecular formula and required this compound to be bicyclic. Comparison of the ^{1}H and ^{13}C chemical shifts of **11** with those of zahavin A indicated that the nine-membered ring B, together with its substituents, and the unsaturated terminal portion of the six-membered side chain had remained intact during the saponification. However, the disappearance of the resonances associated with the acetylated hemicacetal (ring A) indicated changes in this region of the molecule. The chemical shifts of the ring junction protons (δ 2.90, H-1 and δ 2.36, H-9) followed from the COSY spectrum in which contiguous coupling to these protons was observed from H-2 (δ 1.51). Further COSY coupling from H-9 to the olefinic proton (δ 5.91, d, $J_{9,10} = 5.3$ Hz, H-10) and long-range coupling from this latter proton to a second olefinic proton (δ 6.65, s, H-12) suggested a six-membered ring A incorporating a diene functionality. Definitive evidence for the structure of this ring, and its associated substitution pattern, followed from the HMBC spectrum of **11** in which three-bond HMBC correlations were observed, first, from the H-12 olefinic proton to the aldehyde carbonyl (δ 193.6), the C-1 ring junction carbon (δ 37.7), and the C-10 vinylic carbon (δ 134.3), and, second, from the H-10 olefinic proton to the C-12 vinylic carbon (δ 139.8), the C-16 carbinol carbon (δ 77.2), and C-1. A possible mechanism for the formation of **11** from **9**, and hence **1**, is presented in Scheme 1 and involves an initial opening of the cyclic acetal of **9** followed by base-induced elimination of (a) the C-12 alcohol moiety and (b) a proton at C-13 to yield a carbanion at this position. Intramolecular aldol-type condensation of this carbanion with the aldehyde functionality at C-1 and subsequent facile base-catalyzed dehydration would yield compound **11**. Interestingly, a similar compound, xeniafaraunol A (**12**), with the same basic bicyclo[7.4.0]tridecane carbon skeleton as **11**, was recently isolated from the Red Sea soft coral *Xenia faraunensis*.[8] The co-occurrence of xenicane diterpenes and **12** in *X. faraunensis* would suggest a biosynthetic link between these two diterpene groups, possibly analogous to that outlined in Scheme 1.

The second known compound, isolated in this investigation of *E. aurea* from Aliwal Shoal, was 9-deacetoxy-14,15-deepoxyxeniculin (**6**, $[\alpha]_D$ +127°). Unfortunately, no optical rotation was reported for the 9-deacetoxy-14,-15-deepoxyxeniculin originally obtained from *Xenia obscuronata*.[9] The molecular formula of $C_{24}H_{34}O_5$, established for **6** from HREIMS, was identical to that of tsitsixenicin A (**13**, $[\alpha]_D$ −64°), which we had previously isolated from the endemic South African soft coral *Capnella thyrsoidea*.[2] The close similarities in the ^{1}H- and ^{13}C-NMR data of these two compounds (see Table 1), together with the significant differences in optical rotations, suggested that **6** and **13** were diastereomeric. Previous attempts to establish the stereochemistry of an acetoxy group at C-12 in xenicane diterpenes by chemical transformations and spectroscopy have failed.[10,11] Therefore, although the stereochemistry at C-12 in both these compounds remains unassigned, an examination of coupling constants and a series of 1D NOE difference experiments revealed a difference in configuration at C-1. The slightly broadened H-11a singlet (δ 1.96) in the ^{1}H-NMR spectrum of **6** indicated a very small coupling to H-4a (δ 2.00) and is consistent with the *trans* fused bicyclic ring system found in other naturally occurring xenicane diterpenes, including **13**.[2,8] The H-1α configuration in **6** followed from the significant NOE enhancement (3%) of H-4a on irradiation of H-1(δ 5.84) and the $J_{1,11a}$ coupling constant (2.2 Hz). No enhancement of H-4a was observed in an analogous NOE difference experiment performed on **13**, while the $J_{1,4a}$ coupling constant was significantly larger (3.6 Hz), thus suggesting that the stereochemistry at C-1 in this compound is as shown.[2] Although **6** is the major compound in the extract of *E. aurea* from the Aliwal Shoal, it was not reported in the extracts of Sodwana Bay specimens of this species.

HREIMS data established the molecular formula of **7** as $C_{26}H_{36}O_8$. The molecular formula of **7** and **1** differed by a single oxygen atom and by the replacement of the vinylic proton resonance (δ 5.32, H-8), evident in the ^{1}H-NMR spectrum of **1**, with an oxymethine doublet of doublets (δ 2.95, J = 3.7, 10.8 Hz, H-8) in the ^{1}H-NMR spectrum of **7**. These were the only major differences in the ^{1}H-NMR spectra of these two compounds. This observation, supported by the expected differences in the ^{13}C-NMR spectra of **1** and **7**, indicated that the latter compound was the 7,8-epoxy derivative of zahavin A. It is unclear, however, if **7** is indeed a natural product or an oxidation artifact arising from the isolation procedure. There are conflicting reports of the ease of oxidation of the Δ^7-olefin in xenicane diterpenes. Both Scheuer[10] and Kashman[13] suggested that 7,8-epoxidation in xenicanes occurs on standing in air, while Higuchi and co-workers were able to obtain only a 13% yield of the epoxidation product of a similar xenicane diterpene on stirring a $CHCl_3$ solution of the compound in air for two weeks.[11] The appearance of the characteristic H-8 oxymethine resonance in the initial ^{1}H-NMR spectrum of the EtOAc crude extract of *E. aurea* and the perceived stability of **1**, on prolonged standing in $CDCl_3$, mitigates against the probability that **7** is an isolation artifact. The 7,8-*E*-epoxide configuration in **7** was assigned from NOE difference experiments. The observed enhancement of the α-proton, H-4a (19%), on irradiation of H-8 and enhancement of the β-proton,

Table 1. ^{13}C NMR Data for Compounds **6**, **8**, and **13**[a] and ^{1}H, COSY, and HMBC Data for Compound **8**[b]

	6	**13**	**8**			
atom	δ_C	δ_C	δ_C	δ_H[c]	COSY to	HMBC to
1	92.0 (d)	92.3 (d)	70.9 (t)	4.06 dd (5.8, 11.4), 3.52 t (12.0)	H-11a	C-3, C-4a, C-11a, C-11
3	136.5 (d)	142.6 (d)	170.6 (s)			
4	116.9 (s)	116.1 (s)	136.8 (s)			
4a	37.8 (d)	39.7 (d)	43.8 (d)	2.88 dd (3.6, 10.8)	H-5, H-11a	C-3, C-4, C-5, C-6
5	30.0 (t)	32.3 (t)	37.6 (t)	1.62 m, 1.45 m	H-4a, H-6	C-7
6	40.2 (t)	39.7 (t)	40.3 (t)	2.21 m, 2.17 m	H-5	
7	135.6 (s)	134.4 (s)	135.3 (s)			
8	124.4 (d)	124.3 (d)	124.6 (d)	5.41 brt (7.8)	H-9, H-9′, H-18	C-6, C-18
9	25.2 (t)	25.5 (t)	24.8 (t)	2.47 m, 2.12 m	H-8, H-10	
10	35.5 (t)	35.9 (t)	34.6 (t)	2.32 m, 2.12 m	H-9	C-11, C-11a
11	151.1 (s)	149.3 (s)	152.3 (s)			
11a	49.5 (d)	50.1 (d)	49.4 (d)	2.12m	H-1, H-1′, H-4a	
12	72.2 (d)	74.7 (d)	133.2 (d)	6.33 t (7.3)	H-13, H-13′	C-3, C-4a
13	33.4 (t)	31.4 (t)	31.0 (t)	2.60 m, 2.50 m	H-12, H-14	C-4, C-12, C-14, C-15
14	118.8 (d)	118.9 (d)	75.5 (d)	5.25 t	H-13, H-13′	C-12, C-13, C-15, C-16
15	134.5 (s)	135.6 (s)	142.0 (s)			
16	18.0 (q)	18.1 (q)	113.5 (t)	5.00 s, 4.95 s	H-17	C-14, C-15, C-17
17	25.8 (q)	25.7 (q)	18.5 (q)	1.76 s	H-16	C-14, C-15, C-16
18	16.7 (q)	17.0 (q)	18.5 (q)	1.70s	H-8	C-6, C-7, C-8
19	113.2 (t)	113.4 (t)	113.1 (t)	4.95 s, 4.84 s		C-10, C-11a
CO*CH*$_3$	21.2 (q)	21.5 (q)	21.1 (q)	2.06s		*CO*CH$_3$
	21.0 (q)	21.0 (q)				
*CO*CH$_3$	169.7 (s)	169.5 (s)	169.9 (s)			
	170.0 (s)	170.2 (s)				

[a] 100 MHz, $CDCl_3$. [b] 400 MHz, $CDCl_3$. [c] Coupling constants (Hz) in parentheses.

H-11a (5%), on irradiation of the C-18 methyl protons, together with the absence of any enhancement of H-8 resulting from this latter irradiation, supported the assignment as shown.[11,12] Molecular modeling studies (using the modeling program Hyperchem) of both α- and β-epoxide configurations clearly showed that the former configuration is consistent with the NOE enhancements observed with calculated H-4a,H-8 and H-11a,Me-18 interatomic distances of approximately 2 Å.

A molecular formula of $C_{22}H_{30}O_4$ was established from HREIMS data for the second new compound from *E. aurea*, xeniolide C (**8**, $[\alpha]_D$ +96°). The presence of two carbonyl (δ 170 and 169) and eight olefinic resonances (δ 152, 142, 136, 135, 133, 124, 113, 113) in the ^{13}C-NMR spectrum of **8** accounted for six of the eight degrees of unsaturation suggested from the molecular formula. The remaining two degrees of unsaturation required **8** to be bicyclic. A number of similarities in the ^{1}H- and ^{13}C-NMR data between **8** and **1** implied, once again, the same nine-membered ring B system in both these compounds. Additional common resonances in the ^{1}H-NMR spectra of these two compounds included the exocyclic oxymethine (δ 5.25, t, J = 6.4 Hz, H-14), the olefinic methylene (δ 5.0, s, 2H-16), and the methyl (δ 1.76, s, 3H-17) signals and tentatively confirmed the structure of the terminal portion of the six-carbon side chain in **8**. These observations were supported by standard COSY, HMQC, and HMBC NMR data (see Table 1). The major differences between the ^{1}H-NMR spectra of **8** and **1** were therefore confined to ring A in which the absence of H-1 oxymethine and H-3 enol ether proton resonances in the ^{1}H-NMR spectrum of the former compound indicated that **8** did not possess the 1-acetoxydihydropyran ring A structure of **1**. The δ-lactone structure of ring A in **8** was therefore determined as follows. A COSY spectrum coupled the ring junction proton H-11a (δ 2.12) to the deshielded oxymethylene protons with multiplets centered at δ 4.06 (dd, J = 5.8, 11.4 Hz, H-1) and δ 3.52 (t, J = 12 Hz, H-1′), while an HMBC spectrum revealed three-bond correlations from both the oxymethylene protons to the lactone carbonyl (δ 170, C-3) and C-4a (δ 44). Further HMBC correlations from the exocyclic vinylic proton H-12 (δ 6.33, t, J = 7.3 Hz) to the lactone carbonyl and C-4a, together with a contiguous coupling sequence from H-12 to H-14, in the COSY spectrum of **8** unequivocally established the structure of the side chain and placed this side chain at C-4. Finally, the ^{1}H- and ^{13}C-NMR data of **8** compared favorably with the reported data for xeniolide A (**14**) previously isolated from the soft coral *Xenia macrospiculata*.[14]

The antiinflammatory activity previously observed for tsitsixenicin A and its analogs[2] served as a precedent for a similar investigation of the antiinflammatory activity of the xenicane diterpenes from *E. aurea*. Superoxides (O_2^-) are associated with the production of prostaglandins released during the inflammation process. Therefore, compounds that inhibit the release of superoxides may find use as potential antiinflammatory drugs. Rabbit-cell neutrophils can be stimulated to release superoxide, a process that can be monitored via a spectrophotometric method involving the reduction of excess superoxide by cytochrome c. Compounds **1**, **6**, and **7** showed good inhibition of superoxide release from fMLP stimulated rabbit neutrophils (>90%) at a concentration of 20 μg mL^{-1}, which is consistent with the results previously reported for the tsitsixenicins.[2] These results should be interpreted with caution however, because the results obtained with rabbit neutrophils cannot always be directly extrapolated to human-cell neutrophils, which are generally less active in this bioassay.[15] Compounds **1**, **6**, and **7** were not assayed against human-cell neutrophil superoxide release because of the only moderate results obtained for the closely related tsitsixenicins in this bioassay.[2,15]

Experimental Section

General Experimental Procedures. IR spectra were recorded on a Perkin-Elmer series 7 FT-IR spectrophotometer. The ^{1}H- and ^{13}C-NMR spectra were recorded on a Bruker AMX400 spectrometer. Optical rotations were measured on a Perkin-Elmer 141 pola-

rimeter, and LRMS were recorded on a Hewlett-Packard 5988A mass spectrometer. HRMS were obtained by Dr. P. Boshoff of the Mass Spectrometry Unit, Cape Technikon, Cape Town. HPLC separations were performed on Whatman Magnum 9 Partisil and Phenomenex Selectosil C-18 columns.

Biological Material. *E. aurea* (class Octocorallia, order Alcyonacea, family Alcyoniidae) was collected at a depth of 20 m from the Aliwal Shoal off southern Kwazulu−Natal, South Africa in June 1994. Comparison of a specimen of *E. aurea* collected from Aliwal Shoal with the voucher specimens of this species, *E. aurea* Benayahu and Schleyer (1995) described from Sodwana Bay, Durban Harbor and Park Rhynie, revealed the same color, gross morphology, and shape and size of surface and internal sclerites. A voucher specimen of the Aliwal Shoal specimen of *E. aurea* is located in the marine invertebrate collection housed at Rhodes University (SAF 94-013).

Isolation Procedures. The soft coral was immediately frozen after collection and later freeze dried (210 g). All of the freeze-dried soft coral was extracted with EtOAc to give a brown gum (7.0 g), a portion of which (5.3 g) was initially flash chromatographed on Si gel (70%, 50% EtOAc−hexane and EtOAc). Excessive amounts of cholesterol were removed from several of the flash chromatography fractions by crystallization from MeOH. Selected flash chromatography fractions were subsequently subjected to both normal (90% and 85% hexane−EtOAc) and reversed-phase (30% H_2O−MeCN and 20% H_2O−MeOH) HPLC to yield compounds **1** (65 mg), **6** (184 mg), **7** (54 mg), and **8** (21 mg).

Zahavin A (1): colorless oil; $[\alpha]^{21}_D$ +21.5° (*c* 0.77, $CHCl_3$, lit.[3] + 7.3°); IR (film) 2960, 2920, 2850, 1735, 1365, 1230, 1155, 1010 cm^{-1}; ^{1}H- and ^{13}C-NMR data consistent in all respects with published data;[3] EIMS (70 eV) *m/z* (int %) no M^+, 417 (1), 297 (4), 243 (9), 149 (30), 105 (23), 91 (20), 83 (23), 69 (16), 43 (100); HREIMS *m/z* 460.2448 (calcd for $C_{26}H_{36}O_7$, 460.2459).

9-Deacetoxy-14,15-deepoxyxeniculin (6): colorless oil; $[\alpha]^{21}_D$ +126.9° (*c* 0.86, $CHCl_3$); IR (film) 2970, 2930, 2860, 1735, 1670, 1445, 1370, 1230, 1155, 1015, 1015 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.50 (1H, m, H-5), 1.60 (3H, br s, 3H-16), 1.64 (3H, br s, 3H-18), 1.68 (3H, br s, 3H-17), 1.87 (1H, m, H-5′), 1.94 (1H, m, H-6), 1.96 (1H, m, H-11a), 2.00 (1H, m, H-4a), 2.03 (3H, s, Ac), 2.05 (3H, s, Ac), 2.11 (1H, m, H-9), 2.22 (1H, m, H-6′) 2.25 (2H, m, 2H-10), 2.33 (2H, m, 2H-13), 2.45 (1H, m, H-9′), 4.83 (1H, br s, H-19), 4.87 (1H, br s, H-19′), 5.07 (1H, t, *J* = 6.6 Hz, H-14), 5.26 (1H, d, *J* = 5.9 Hz, H-8), 5.35 (1H, t, *J* = 6.2 Hz, H-12), 5.84 (1H, d, *J* = 2.2 Hz, H-1), 6.37 (1H, s, H-3); ^{13}C NMR data, see Table 1; EIMS (70 eV) *m/z* (int %) no M^+, 359 (2), 349 (21), 342 (4), 333 (13), 291 (14), 231 (25), 149 (25), 91 (37), 69 (38), 43 (100); HREIMS *m/z* 402.2419 (calcd for $C_{24}H_{34}O_5$, 402.2406).

7,8-Epoxyzahavin A (7): colorless oil; $[\alpha]^{21}_D$ +121.7° (*c* 0.34, $CHCl_3$); IR (film) 2970, 2940, 2870, 1740, 1440, 1372, 1235, 1162, 1020, 915 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.08 (1H, m, H-6), 1.30 (3H, br s, 3H-18), 1.49 (1H, m, H-9), 1.67 (1H, m, H-5), 1.72 (3H, br s, 3H-17), 1.90 (2H, m, 2H-13), 2.03 (3H, s, Ac), 2.05 (3H, s, Ac), 2.06 (1H, m, H-4a), 2.07 (3H, s, Ac), 2.08 (1H, m, H-5′), 2.25 (1H, m, H-6′), 2.28 (1H, m, H-9′), 2.39 (2H, m, 2H-10), 2.42 (1H, m, H-11a), 2.95 (1H, dd, *J* = 3.7, 10 Hz, H-8), 4.89 (1H, br s, H-16), 4.95 (1H, br s, H-16′), 4.96 (1H, br s, H-19), 5.04 (1H, br s, H-19′), 5.19 (1H, dd, *J* = 5.3, 8.2, H-14), 5.29 (1H, dd, *J* = 4.7, 8.2, H-12), 5.93 (1H, d, *J* = 2.1 Hz, H-1), 6.40 (1H, s, H-3); ^{13}C NMR (100 MHz, $CDCl_3$) 16.9 (q, C-18), 18.3 (q, C-17), 3 × 21.0 (q, 3 × Ac), 24.7 (t, C-9), 29.2 (t, C-5), 32.1 (t, C-10), 37.1 (d, C-4a), 38.9 (t, C-13), 39.5 (t, C-6), 47.9 (d, C-11a), 59.8 (s, C-7), 62.8 (d, C-8), 68.6 (d, C-12), 73.1 (d, C-14), 91.6 (d, C-1), 112.8 (t, C-16), 115.2 (t, C-19), 116.9 (s, C-4), 137.5 (d, C-3), 142.8 (s, C-15), 148.0 (s, C-11), 169.5 (s, Ac), 169.8 (s, Ac), 170.0 (s, Ac); EIMS (70 eV) *m/z* (int %) no M^+, 417 (1), 356 (5), 297 (6), 243 (15), 149 (23), 145 (23), 105 (23), 91 (27), 81 (25), 69 (17), 43 (100); HREIMS *m/z* 476.2419 (calcd for $C_{26}H_{36}O_8$, 476.2408).

Xeniolide C (8): colorless oil; $[\alpha]^{21}_D$ +96.4° (*c* 1.17, $CHCl_3$); IR (film) 2970, 2925, 2870, 1735, 1645, 1438, 1365, 1225, 1025 cm^{-1}; ^{1}H- and ^{13}C-NMR data, see Table 1; EIMS (70 eV) *m/z* (int %) no M^+, 298 (9), 246 (60), 203 (20), 145 (37), 105 (48), 91 (82), 79 (57), 43 (100); HRIEMS *m/z* 358.2158 (calcd for $C_{22}H_{30}O_4$, 358.2143).

Attempted Saponification of Zahavin A. Dry K_2CO_3 (56 mg) was dissolved in H_2O (0.5 mL) and added to a stirred solution of **1** (18.0 mg) in MeOH (6 mL). After 16 h, H_2O (4.5 mL) was added and the MeOH removed under reduced pressure. The aqueous layer was extracted with EtOAc, which was then dried over anhyd. Na_2SO_4 and concentrated *in vacuo*. TLC and ^{1}H-NMR spectroscopy suggested a complex reaction mixture with no evidence of the starting material. The rearrangement compound **11** (1.3 mg) was separated from this mixture and purified by normal-phase HPLC (80% hexane−EtOAc).

Compound 11: colorless oil; $[\alpha]^{21}_D$ −76.4° (*c* 0.13, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.43 (1H, s, H-20), 6.65 (1H, s, H-12), 5.91 (1H, d, *J* = 5.3 Hz, H-10), 5.49 (1H, t, J = 8.3 Hz, H-5), 5.19 (1H, s, H-18), 5.00 (1H, s, H-18′), 4.90 (1H, s, H-15), 4.66 (1H, s, H-15′), 4.61 (1H, s, H-16), 2.90 (1H, m, H-1), 2.45 (1H, m, H-6), 2.36 (1H, m, H-9), 2.30 (1H, m, H-3), 2.30 (2H, m, H-7), 2.17 (1H, m, H-3′), 2.17 (1H, m, H-6′), 1.69 (3H, s, H-14), 1.63 (3H, s, H-19), 1.51 (2H, m, H-2); ^{13}C NMR (100 MHz, $CDCl_3$) δ 193.6 (d), 156.3 (s), 145.1 (s), 140.9 (s), 139.8 (d), 136.1 (s), 134.3 (d), 130.2 (s), 125.0 (d), 114.3 (t), 111.6 (t), 77.2 (d), 49.5 (d), 39.8 (t), 37.7 (d), 35.8 (t), 34.4 (t), 25.3 (t), 18.7 (q), 16.9 (q); HREIMS *m/z* 298.1939 (calcd for $C_{20}H_{36}O_2$, 298.1933).

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References and Notes

(1) Lin, Y.; Bewley, C. A.; Faulkner, D. J. *Tetrahedron* **1993**, *49*, 7977−7984; Hooper, G. J.; Davies-Coleman, M. T. *Tetrahedron Lett.* **1995**, *36*, 3265−3268; Korin-Goldshlager, G.; Klein, P.; Rudi, A.; Benayahu, Y.; Schleyer, M.; Kashman, Y. *J. Nat. Prod.*

1996, *59*, 262−266; Fridkovsky, E.; Rudi, A.; Benayahu, Y.; Kashman, Y.; Schleyer, M. *Tetrahedron Lett.* **1996**, *38*, 6909−6910.
(2) Hooper, G. J.; Davies-Coleman, M. T. *Tetrahedron* **1995**, *51*, 9973−9984.
(3) Rudi, A.; Ketzinel, S.; Goldberg, I.; Stein, Z.; Kashman, Y; Benayahu, Y.; Schleyer, M. *J. Nat. Prod.* **1995**, *58*, 1581−1586.
(4) Ketzinel, S.; Rudi, A.; Schleyer, M.; Benayahu, Y.; Kashman, Y. *J. Nat. Prod.* **1996**, *59*, 873−875.
(5) Benayahu, Y.; Schleyer, M. *Oceanogr. Res. Inst. Invest. Rep. (Durban)* **1995**, *68*, 1−12.
(6) Faulkner, D. J. *Nat. Prod. Rep.* **1995**, *12*, 223, and previous reviews cited in this series.
(7) Dann, A. E.; Davis, J. B.; Nagler, M. J. *J. Chem. Soc., Perkin Trans. 1*, **1979**, 158−160; Rychnovosky, S. D.; Skalitzky, D. J. *Tetrahedron Lett.* **1990**, *31*, 945−948.
(8) Kashman, Y.; Saltoun, M.; Rudi, A.; Benayahu, Y. *Tetrahedron Lett.* **1994**, 35, 8855−8858.
(9) Kashman, Y.; Groweiss, A. *J. Org. Chem.* **1980**, *45*, 3814−3827.
(10) Coval, S. J.; Scheuer, P. J.; Matsumoto, G. K.; Clardy, J. *Tetrahedron* **1984**, *40*, 3823−3828.
(11) Miyamoto, T.; Takenaka, Y.; Yamada, K.; Higuchi, R. *J. Nat. Prod.* **1995**, *58*, 924−928.
(12) Ochi, M.; Kataoka, K.; Tatsukawa, A.; Kotsuki, H.; Shibata, K.; *Heterocycles* **1994**, *38*, 151−158.
(13) Professor Yoel Kashman, personal communication.
(14) Kashman, Y.; Groweiss, A. *Tetrahedron Lett.* **1978**, 4833−4836.
(15) Dr. Dave Burgoyne, personal communication.

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