181. Rarisetenolide, Epoxyrarisetenolide, and Epirarisetenolide, New-Skeleton Sesquiterpene Lactones as Taxonomic Markers and Defensive Agents of the Marine Ciliated Morphospecies *Euplotes rariseta*

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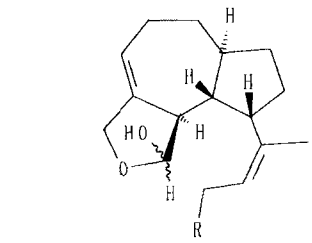
Rarisetenolide (**3**) and epoxyrarisetenolide (**4**), new-skeleton α,β-conjugated sesquiterpene lactones, were isolated from various strains of the unicellular ciliated protist *Euplotes rariseta* collected from marine coasts widely far apart from each other: northern and southern Australia, southern Brazil, and Canary Islands. A strain of *E. rariseta* from New Zealand gave epirarisetenolide (**5**) instead, revealing a subtle variability in secondary metabolism for this ciliated morphospecies. Nonetheless, these metabolites – which are the first non-aldehydic terpenoids so far isolated from ciliates – represent a unique whole that constitutes a reliable taxonomic tool at the morphospecies level. Epirarisetenolide (**5**) and rarisetenolide (**3**), in this order, showed higher toxicity towards nonproductive ciliates than the chemically more reactive natural epoxide **4** and the semisynthetic aldehyde/protected-aldehyde forms **8**/**7a**/**7b**. This inverse trend of biological *vs.* chemical effectiveness suggests that these cytotoxic agents interact noncovalently with membrane receptors.

1. Introduction. – We have recently shown that marine hypothrich ciliates in the genus *Euplotes* produce terpene aldehydes in protected (hemiacetal) form, to which the role of broadening agents of the niche size has been attributed. This is the case of euplotins A–C, isolated from the morphospecies[1]) *Euplotes crassus* (DUJARDIN, 1841), which depress the division rate, or kill at higher concentrations, most other interstitial ciliates except raptorial ciliates [1]. In contrast, raikovenal and epiraikovenal, produced by the morphospecies *Euplotes raikovi* AMAGALIEV, 1966 [2], as well as focardin (**1**) and epoxyfocardin (**2**) from the Antarctic ciliate *Euplotes focardii* VALBONESI et LUPORINI, 1990 [3], act as defensive agents against predatory ciliates while little affecting the other interstitial ciliates.

Evolutionarily significant is the conservation of the same euplotins by the different populations of *E. crassus* [4], implying a very strong selection pressure, whereas a variability was noticed among different populations of *E. raikovi* in the production of either raikovenal [2a] or epiraikovenal [2b].

We present here the first non-aldehydic terpenoids isolated from ciliates. They can reliably challenge traditional morphologic characters for inferring taxonomic relationships within the morphospecies *Euplotes rariseta* CURDS, WEST et DORAHY, 1974, and reveal an inverse trend of biological activity *vs.* chemical reactivity.

[1]) Morphospecies stands for species defined on morphological characteristics.



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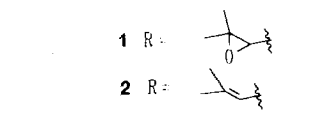

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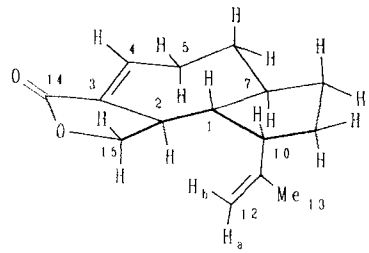

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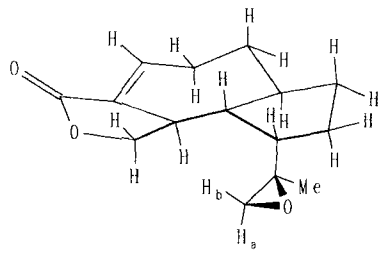

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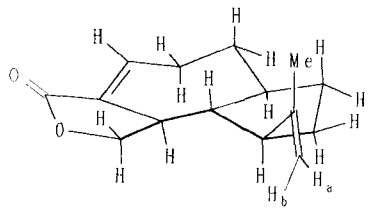

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2. Results and Discussion. – 2.1. *The Structures.* The composition $C_{15}H_{20}O_2$ for rarisetenolide (**3**)[2]) is secured by matching of the number of C- and H-atoms derived from HR-MS with those from NMR spectra; the latter also reveal the presence of two C=C and one C=O bond, suggesting a tricyclic compound. The γ-lactone ring of **3** rests on characteristically deshielded diastereotopic γ-protons (2 H–C(15)), heterocorrelated with C(15) (*t*) and C(14)=O (*s*) (see *Table 1*, last entry)[3]). The presence of an α,β-conjugated C=C bond at the lactone group is supported by strong UV absorption at 225 nm, and its exocyclic position rests on heterocorrelation between C=O and H–C(4). Fusion of the γ-lactone moiety to a bicyclo[5.3.0]decene system bearing a vinyl side chain is supported by DDS, COSY, HMQC, and HMBC data in either $CDCl_3$ or C_6D_6; in the latter solvent, the proton resonances are better resolved (*Table 1*). NOE Data show that *a*) H–C(1), H–C(10), and the H–C(15) at δ 3.41 (*dd*) are situated on the same, arbitrarily called β[4]), face, *b*) H–C(2) and the H–C(15) at δ 4.15 (*t*) lie on the opposite face, and *c*) H_a–C(12) and 3 H–C(13) are *cis*-interrelated. Consistently, a NOE enhancement is observed between H–C(10) and H_b–C(12), suggesting that the C(11)=C(12) and H–C(10) bonds are eclipsed, probably to minimize allylic 1,3-strain [5]. Assignment of *trans*-fusion of the cycloheptene and cyclopentane rings, hindered by submerged ^{1}H-NMR signals for H–C(7), is based on *a*) large $J(7,6\beta)$, $J(7,1)$, and $J(7,8\beta)$ coupling constants, implying a *trans*-diaxial relationship between the relevant H-atoms, *b*) a homoallylic '*cisoid*' cou-

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[2]) Viewed in the preferred conformation.

[3]) Arbitrary C-atom numbering; for systematic numbering and names, see *Exper. Part.*

[4]) No absolute-configuration meaning is attributed to any of the new compounds described in this work.

pling between H–C(2) and H_a–C(5), *c*) identical couplings between H–C(4) and the two protons at C(5), and *d*) typical δ(C) values for C(1) and C(7). No NOESY maps can be noticed between H–C(1) and H–C(7) at such a high field (600 MHz) that the corresponding resonances for these protons are well resolved. These coupling patterns are nicely simulated by molecular-mechanics (MM) calculations. In further support of structure **3**, none of these couplings is predicted by MM calculations for the hypothetical 1,7-*cis*-fused stereoisomer. These stereochemical conclusions are corroborated by the agreement between the observed and calculated mean coupling constant $J(10,9\alpha) = 3.4$ Hz for mobile-envelope and half-chair conformations of the cyclopentane ring, like for focardin (**1**) [3]. Also the calculated most favorable conformation of the side chain of **3**, involving eclipsing of the H–C(10) and C(11)=C(12) bonds, is in accordance with the NMR observations.

Table 1. *NMR Data* (C_6D_6) *for Rarisetenolide* (**3**)[3][a])

	^{13}C-NMR	^{1}H-NMR
H–C(1)	52.34 (*d*)	0.73 (*dt*, $J(1,10) = 8.6$, $J(1,2) \approx J(1,7) = 11.5$)
H–C(2)	40.43 (*d*)	2.36 (*tddd*, $J(2,4) \approx J(2,5\alpha) = 3.9$, $J(2,15\alpha) = 9.0$, $J(2,15\beta) = 7.9$, $J(2,1) = 11.5$)
C(3)	132.32 (*s*)	–
H–C(4)	140.25 (*d*)	7.10 (*td*, $J(4,5) \approx J(4,2) = 3.3$, $J(4,5\beta) = 8.9$)
CH_2(5)	27.61 (*t*)	α: 1.54 (*qdd*, $J(5\alpha,2) \approx J(5\alpha,6\alpha) \approx J(5\alpha,4) = 3.7$, $J(5\alpha,6\beta) = 12.7$, $J_{gem} = 15.8$)
		β: 1.90 (*dddd*, $J(5\beta,6\beta) = 2.5$, $J(5\beta,6\alpha) = 4.8$, $J(5\beta,4) = 8.9$, $J_{gem} = 15.8$)
CH_2(6)	32.71 (*t*)	α: 1.45 (*m*)
		β: 0.64 (*dddd*, $J(6\beta,5\beta) = 2.0$, $J(6\beta,7) = 10.7$, $J(6\beta,5\alpha) = 12.7$, $J_{gem} = 14.9$)
H–C(7)	47.58 (*d*)	1.38 (*tddd*, $J(7,8\alpha) \approx J(7,6\alpha) = 1.6$, $J(7,8\beta) = 10.0$, $J(7,6\beta) = 10.7$, $J(7,1) = 11.5$)
CH_2(8)	33.97 (*t*)	α: 1.60 (*dddd*, $J(8\alpha,9\beta) = 3.7$, $J(8\alpha,7) = 6.8$, $J(8\alpha,9\alpha) = 9.6$, $J_{gem} = 13.5$)
		β: 0.92 (*dddd*, $J(8\beta,9\alpha) = 8.7$, $J(8\beta,7) = 10.0$, $J(8\beta,9\beta) = 11.4$, $J_{gem} = 13.5$)
CH_2(9)	28.10 (*t*)	α: 1.28 (*dddd*, $J(9\alpha,10) = 3.4$, $J(9\alpha,8\alpha) = 9.6$, $J(9\alpha,8\beta) = 11.4$, $J_{gem} = 13.7$)
		β: 1.55 (*dtd*, $J(9\beta,8\alpha) = 3.7$, $J(9\beta,8\beta) \approx J(9\beta,10) = 8.7$, $J_{gem} = 13.7$)
H–C(10)	49.33 (*d*)	2.33 (*dt*, $J(10,9\alpha) = 3.4$, $J(10,9\beta) \approx J(10,1) = 8.6$)
C(11)	147.16 (*s*)	–
CH_2(12)	114.06 (*t*)	a: 4.54 (*qd*, J(12a,Me) = 1.5, $J_{gem} = 2.4$)
		b: 4.41 (br. *qd*, J(12b,Me) = 0.7, $J_{gem} = 2.4$)
Me(13)	21.47 (*q*)	1.39 (*dd*, J(Me,12a) = 1.5, J(Me,12b) = 0.7)
C(14)	170.48 (*s*)	–
CH_2(15)	69.51 (*t*)	α: 4.15 (*t*, $J(15\alpha,2) = J_{gem} = 9.0$)
		β: 3.41 (*dd*, $J(15\beta,2) = 7.9$, $J_{gem} = 9.0$)

[a]) α and β refer to protons lying below or above the plane of the paper, respectively, on which structure **3** is drawn.

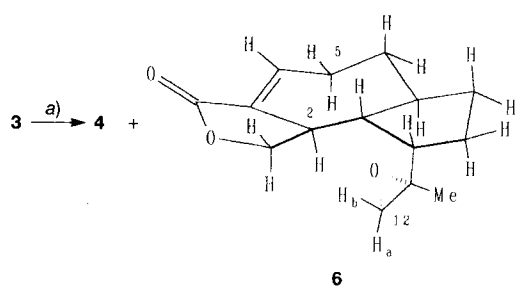
An oxirane group in **4**[2]) is suggested by both NMR and MS data. The configuration rests on strong NOEs between 3 H–C(13) and both H_a–C(12) and H–C(2), implying eclipsing of the H–C(10) and C(11)–C(12) bonds; this is also nicely simulated by MM calculations. Definitive evidence for structure **4** comes from semisynthesis, as discussed below.

Overall similar MS suggest that epirarisetenolide (**5**)[2]) must be isomeric with rarisetenolide (**3**); different fragmentations for the two compounds (*Exper. Part*) suggest some structural dissimilarity anyway. Scarce availability of **5** only allowed us to record 1D ^{1}H-NMR spectra. These, too, suggest that **5** is isomeric with **3** while revealing the site of structural dissimilarity. Thus, H–C(10) in **5** is shielded by 0.4 ppm with respect to **3**,

and also the neighboring protons – *i.e.*, the exocyclic methylene protons and 2 H–C(15) – are affected, while the resonances for the other protons are superimposable for the two compounds. This suggests that the two compounds are epimeric at C(10). This proposal is corroborated by the coupling pattern of H–C(10), which is different in the two compounds, for **5** (*dt*, $J(10,9\alpha) = 6.5$, $J(10,9\beta) \approx J(10,1) = 10.1$ Hz) matching expectations from MM calculations.

2.2. *The Reactivity.* Functional-group elaboration of rarisetenolide (**3**) was attempted both to confirm the above structural attributions and to furnish derivatives for structure-bioactivity correlations. The first task was easily accomplished *via* epoxidation of **3** with 3-chlorobenzenecarboperoxoic acid (3-$ClC_6H_4CO_3H$) yielding a 9:1 mixture of nature-identical epoxyrarisetenolide (**4**) and the unnatural diastereoisomer **6**[2]) (*Scheme 1*). Deshielding of H–C(2), and particularly of H_α–C(15), of **6** suggests that these two protons lie quite close to the epoxide O-atom, demanding a preferential conformation having C(12) above the plane of the paper as represented in *Scheme 1*; MM emulations are in agreement. Attack by the electrophile from the less hindered face of the vinyl group in the preferred conformation **3** can be imagined to give the natural epoxide **4**; similarly, attack by the electrophile from the opposite face of the vinyl group (or, more likely, from the less hindered face of the minor conformer) gives **6**. This rationalizes the good stereoselectivity (9:1) observed for this epoxidation.

Scheme 1



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a) 3-$ClC_6H_4CO_3H$, CH_2Cl_2, $NaHCO_3$, r.t., overnight.

The second task was accomplished by reduction of the lactone moiety in **3** to a hemiacetal group, providing aldehydic forms. Reaction of **3** with DIBAL (diisobutylaluminium hydride) furnished a 7:3 mixture of the epimeric hemiacetals **7a** and **7b**, in equilibrium with 7% of the free aldehyde **8** (*Scheme 2*). An unaltered ratio of products on repeated HPLC elution suggests equilibrium conditions. UV Irradiation of rarisetenolide (**3**) in MeOH, by removing lactone conjugation, provided further structural varieties: compounds **9** and **10**, obtained by MeOH addition in a 85:15 molar ratio (*Scheme 2*). Addition of MeOH rather than deconjugation [6] under UV irradiation is in accordance with the proposed lactone structure[5]).

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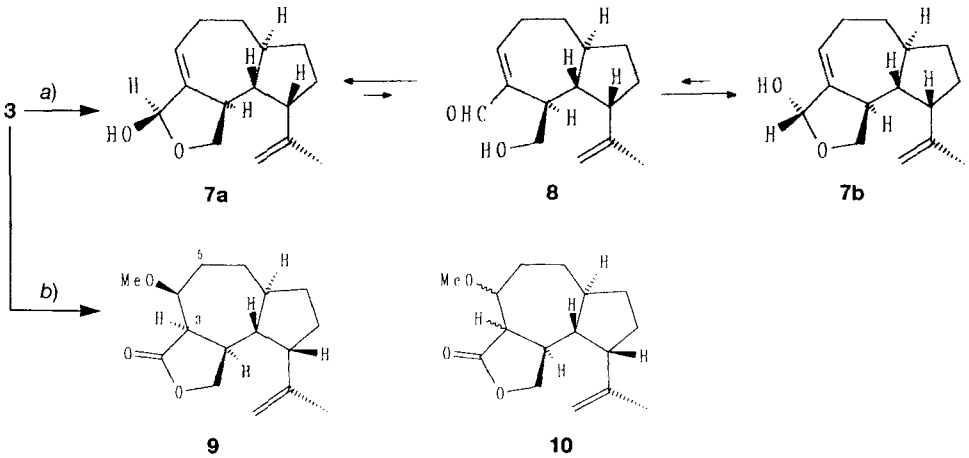
[5]) For **9**, the pseudo-axial position of MeO rests on small *J* coupling constants for H–C(4), while the α-position for H–C(3) rests on NOE enhancement on irradiation of H_α–C(5). Shortage of material prevented similar experiments for **10**, whose configuration at C(3) and C(4) remains undefined.

Scheme 2



a) DIBAL, hexane, 0°, 2 h. *b*) *hν* (254 nm), MeOH, quartz cuvette, r.t., 40 min.

2.3. *The Biological Role.* Rarisetenolide (**3**), epirarisetenolide (**5**), and epoxyrarisetenolide (**4**) are among the weakest bioactive terpenoids isolated from the ciliates examined so far, the latter being the weakest of the three. Derivative **9** ranks with terpenoids of low cytotoxicity as **3**, while the equilibrating derivative **7a**/**7b**/**8** has even weaker toxicity, like **4**. Cytotoxicity of these terpenoids against representatives of the marine interstitial ciliate community, comprising *E. rariseta* itself, showed up at only a relatively high dose, 20 μg/ml; the only exception was noticed towards the ciliate *Litonotus lamella* (O. F. MÜLLER, 1773), which proved sensitive to the cytotoxic agents at lower doses (*Table 2*).

The cytotoxicity level expressed on the unbiased sample of the interstitial ciliate diversity (*Table 2*) showed consistency for each one of the terpenoids, largely differing for different terpenoids, however, in the order of decreasing effectiveness **5** > **3** = **9** > **4** = **7a**/**7b**/**8**. Diminished cytotoxicity on either epoxidation of **3** to **4** or aldehyde/protected-aldehyde liberation from **3** (→**7a**/**7b**/**8**), *i.e.*, lower cytotoxicity of the chemically more reactive species, gives some hint that interactions with membrane receptors [1a] of the *Euplotes* morphospecies are noncovalent in nature.

As mentioned above, the strongest cytotoxicity of these terpenoids was recorded upon *L. lamella*. It is worth noticing that *L. lamella* differs from all other ciliates involved in our bioassays for its behavior as a predator. It can be maintained that the effectiveness of rarisetenolite (**3**) against *L. lamella* could make part of a defensive strategy of *E. rariseta* to avoid predation. The evidence is as follows. Individuals of the strain 'Li' of *L. lamella* prey *E. rariseta* cells, whatever their strain membership be, and increase in number, suggesting that *E. rariseta* representatives were edible by the predator. However, when *E. rariseta* individuals were presented to *L. lamella* alongside other representatives of the marine interstitial ciliate community, these last were clearly preferred. This is the case of the strains in *Table 2* of *E. vannus* (O. F. MÜLLER, 1786), *E. crassus* (DUJARDIN, 1841), and *E. minuta* YOCUM, 1930. Unlike *E. vannus* and *E. crassus*, which are of relatively large size

Table 2. *Terpenoids* **3**–**5** *of the Ciliate* Euplotes rariseta *and Their Semisynthetic Derivatives* **7**–**9**: *Toxicity towards Ciliate Strains Representing an Unbiased Sample of the Marine Interstitial Ciliate Diversity*

Strain	Morphospecies	Lowest concentration [μg/ml] eliciting 100% kills				
		3	**4**	**5**	**7a/7b/8**	**9**
BR1	*Euplotes rariseta* CURDS, WEST *et* DORAHY, 1974	[a])	[a])	20	[a])	[a])
GRH5	*Euplotes rariseta* CURDS, WEST *et* DORAHY, 1974	[a])	[a])	[b])	[a])	[a])
PBH1	*Euplotes rariseta* CURDS, WEST *et* DORAHY, 1974	[a])	[b])	20	[b])	[b])
NZ	*Euplotes rariseta* CURDS, WEST *et* DORAHY, 1974	[a])	[b])	20	[b])	[b])
SicAA	*Euplotes* sp.	20	[b])	10	[b])	[b])
CA1	*Euplotes* sp.	[a])	[b])	20	[b])	[b])
TB6	*Euplotes vannus* (O. F. MÜLLER, 1786)	20	[a])	[b])	[a])	20
G-Lb5	*Euplotes crassus* (DUJARDIN, 1841)	20	[a])	[b])	[a])	[a])
SSt22	*Euplotes crassus* (DUJARDIN, 1841)	20	[a])	[b])	[a])	20
SL2	*Euplotes crassus* (DUJARDIN, 1841)	20	20	[b])	[a])	20
Mar11	*Euplotes minuta* YOCUM, 1930	20	[a])	[b])	[a])	20
Kling2	*Euplotes charon* (O. F. MÜLLER, 1773)	20	[a])	[b])	[a])	20
SB8	*Euplotes raikovi* AGAMALIEV, 1966	[a])	[a])	[b])	[a])	[a])
CO	*Euplotes magnicirratus* CARTER, 1972	20	[a])	[b])	[a])	20
Li	*Litonotus lamella* (O. F. MÜLLER, 1773)	10	[a])	10	5	10

[a]) No effect at concentrations up to 20 μg/ml. [b]) Not investigated.

in this order, *E. minuta* is of smaller size, much the same as *E. rariseta*. This rules out the possibility that *L. lamella* carries on a selective predatism of large-sized ciliates. Hence, the recorded detrimental effect of secondary metabolites of *E. rariseta* appears to work in rendering this species less palatable to the predator *L. lamella* with respect to the other ciliates which share the same habitat. The conclusion that the terpenoids at issue express their ecological role in improving niche exploitation by *E. rariseta* is warranted.

The four strains of *E. rariseta*, PD16, BR1, PBH1, and GRH5, which produce both rarisetenolide (**3**), and, as likely a physiologically conditioned by-product, epoxyrarisetenolide (**4**), have been collected from locations geographically widely far apart from each other. It is difficult to imagine that this is incidental. The production of secondary metabolites is related to enzymes encoded by the organism's genome, their presence reflecting the expression of functional genes. Therefore, the inference of a close phylogenetic relationship among the foregoing strains is warranted. Strain NZ produces epirarisetenolide (**4**), whose epimeric relationship with rarisetenolide (**3**) implies profound differences in metabolic pathways with respect to the other strains above, hence a genetic

differentiation of strain NZ. Neither rarisetenolide nor epirarisetenolide could be detected at the level of sensitivity of our HPLC-UV methodology in two strains, SicAA[6]) and Ca1[7]), which were tentatively classified as *E. rariseta*. The entangled taxonomy of *E. rariseta* could provide, however, a more conceivable explanation of such inconsistency. *Valbonesi* and *Luporini* refer to literature records of at least three different morphotypes comprised under the species denomination *E. rariseta*, and their newly described Antarctic strain represents the fourth such morphotype [7]. Reinvestigation of the type slides of *Euplotes algivora* AGATHA, WILBERT, SPINDLER et ELBRÄCHTER, 1190 by *Petz et al.* [8] established synonymousness between *E. algivora* and *E. rariseta*. All this suggests a highly polymorphic condition of the *E. rariseta* taxon and an unreliability of the outwardly visible characteristics to establish conspecificity. Strains SicAA and CA1, as well as strain NZ, could belong to different evolutionary units of the *E. rariseta* complex, endowed of peculiar physiological and genetic characteristics with respect to the unit comprising the above four rarisetenolide-producing strains. The results of a cross-mating analysis allows classifying the aforesaid representatives of evolutionary units into different mating groups, stressing a loosening of inter-unit affinities. All this strengthens the proposal of natural-product characters as an intrinsically better tool than traditional morphocharacters in inferring phylogenetic relationships for *E. rariseta*.

We thank Prof. *M. J. Menon*, Mrs. *D. Rossi*, and Dr. *G. Santangelo* for collecting strains BR1, PD16, and NZ, respectively, as well as Drs. *A. Donati* and *C. Bonechi* (Dipartimento di Chimica, Università di Siena) for the 600-MHz ^{1}H-NMR spectra, and, for financial support, both *MURST* (Progetti 40% and 60%) and *CNR*, Roma.

Experimental Part

1. *General.* See [2b]. Moreover, differential NOEs: irradiated proton(s)→NOE (%) on the observed proton(s). NOESY concerning **3** (mixing time 200 ms) were carried out on a *Bruker-AMX-600* spectrometer operating at 600 MHz. Photochemistry was carried out using a very-low-output *RS55* semimicro photochemical reactor from *Applied Photophysics*, London. Molecular-mechanics (MM) calculations were carried out with the PCMODEL 4.0 computer program, based on MMX force field, from *Serena Software*, Bloomington, Indiana.

2. *Collection, Culture, and Isolation. Euplotes rariseta* CURDS, WEST et DORAHY, 1974, strain PD16, was collected in October 1992 near Port Douglas along the north-eastern coasts of Australia. Strain PBH1 was collected under the lighthouse of Palm Beach in extreme southern Sydney in April 1995. Strain BR1 was collected at Ubatuba beach, along the southern coasts of Brazil, in January 1995. Strain GRH5 was collected along the beach of Garachico, Teneriffe, Canary Islands, in September 1995. Strain NZ was collected along the shore in Omaha Bay, New Zealand. Strain PD16 was repeatedly mass cultured by the methodology described for *Euplotes crassus* [1], collecting by centrifugation a total of 2.5 ml of closely packed cells (*ca.* $2.2 \cdot 10^8$ cells), which were soaken in little abs. EtOH and stored at −80°. At culture completion, after some weeks, the solvent was recovered by filtration on sintered glass, thoroughly washing the cells with fresh abs. EtOH. The filtrate was evaporated and the residue partitioned between hexane/AcOEt 9:1 and H_2O. The aq. phase, evaporated, gave 110 mg, of a residue mainly constituted of nucleoside. The org. phase was separated and evaporated yielding 98 mg of residue that was subjected to FC (*Si60*, hexane/AcOEt gradient elution, collection of 11 fractions of 40 ml each). *Fr. 6–7* were subjected to reversed-phase FC (*RP-18*, MeCN) yielding from the first two fractions *rarisetenolide* (**3**), which was further purified by HPLC (*Si60*, hexane/i-PrOH 97:3; t_R 10.0 min): 5.5 mg of **3**.

Cultures of *E. rariseta*, strain BR1, led to 8.1 ml of closely packed cells (*ca.* $7 \cdot 10^8$ cells) which were worked out as above (17 fractions of 40 ml each). *Fr. 5–7*, obtained as above, led to **3** (10.2 mg). *Fr. 14*, subjected to FC (*RP-18*) as above, followed by HPLC (*Si60*, hexane/i-PrOH 95:5), led to *epoxyrarisetenolide* (**4**; t_R 10 min; 1.2 mg).

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[6]) Found by *P. Luporini* in a sand sample of the southern Thyrrhenian shores near Milazzo, Italy.

[7]) Collected from a not recorded location of the Caribbean Sea.

Cultures of *E. rariseta*, strain NZ, led to 0.5 ml of closely packed cells (*ca.* $4 \cdot 10^7$ cells), which were worked out as above yielding 15 mg of org. residue that was subjected to reversed-phase FC (*RP-18*, MeCN, collection of 2-ml fractions). *Fr. 2* was further purified by HPLC (*Si60*, hexane/i-PrOH 97:3) to give *epirarisetenolide* (**5**; t_R 9.8 min; *ca.* 0.4 mg).

3. *Cytotoxicity Assays.* Details of the experimental protocol followed to carry out biological assays of the various terpenoids in this work were already reported [1], except that DMSO, instead of EtOH, was used in this work as a solvent to prepare both the stock solns. of the terpenoids and their artificial-seawater/DMSO test solns. used to run through the cytotoxicity bioassays the series of ciliate strains employed for the tests. The latter are reported in *Table 2*. Controls consisting of both DMSO at the highest concentration of 2% occurring in the test solns. and pure, sterilized seawater were run simultaneously with experimental treatments. The toxic effect of each terpenoid on each ciliate strain tested is expressed as the lowest terpenoid concentration in μg/ml eliciting 100% kills (lethal dose, LD_{100}) (*Table 2*). Reasons for choosing this parameter for ecological work were already given [1b].

4. *Rarisetenolide* (= *(6aR*,9R*,9aS*,9bS*)-5,6,6a,7,8,9,9a,9b-Octahydro-9-(1-methylethenyl)azuleno[4,5-c]furan-3(1H)-one*; **3**). $[\alpha]_D^{20} = -54$ (*c* = 0.28, MeOH). CD (MeOH; $\Delta\varepsilon_{max}(\lambda)$): −0.23 (226). UV (MeOH): 225 (8200). ^{1}H- and ^{13}C-NMR (C_6D_6): *Table 1*. ^{1}H-NMR ($CDCl_3$)[3]): 1.47 (*dt*, *J*(1,10) = 8.3, *J*(1,2) ≈ *J*(1,7) = 11.2, H–C(1)); 3.05 (*tddd*, *J*(2,4) ≈ *J*(2,5α) = 3.7, *J*(2,15α) = 9.1, *J*(2,15β) = 8.1, *J*(2,1) = 11.2, H–C(2)); 7.13 (*td*, *J*(4,2) ≈ *J*(4,5α) = 3.7, *J*(4,5β) = 9.1, H–C(4)); 2.12 (*qdd*, *J*(5α,2) ≈ *J*(5α,6α) ≈ *J*(5α,4) = 3.7, *J*(5α,6β) = 11.7, J_{gem} = 16.2, H_α–C(5)); 2.50 (*dddd*, *J*(5β,6β) = 2.5, *J*(5β,6α) = 4.7, *J*(5β,4) = 9.1, J_{gem} = 16.2, H_β–C(5)); 1.96 (*m*, H_α–C(6)); 1.12 (*dddd*, *J*(6β,5β) = 2.5, *J*(6β,7) = 10.6, *J*(6β,5α) = 11.7, J_{gem} = 13.7, H_β–C(6)); 1.98 (*m*, H–C(7)); 2.00 (*m*, H_α–C(8)); 1.32 (*dddd*, *J*(8β,9α) = 7.5, *J*(8β,7) = 9.2, *J*(8β,9β) = 9.8, J_{gem} = 13.9, H_β–C(8)); 1.56 (*m*, H_α–C(9)); 1.92 (*dddd*, *J*(9β,8α) = 1.0, *J*(9β,8β) = 9.8, *J*(9β,10) = 8.3, J_{gem} = 13.9, H_β–C(9)); 2.90 (*dt*, *J*(10,9α) = 3.7, *J*(10,9β) ≈ *J*(10,1) = 8.3, H–C(10)); 4.77 (*qd*, *J*(12a,Me) = 1.4, J_{gem} = 2,2, H_a–C(12)); 4.72 (*qd*, *J*(12b,Me) = 0.6, J_{gem} = 2.2, H_b–C(12)); 1.78 (*dd*, *J*(Me,12b) = 0.8, *J*(Me,12a) = 1.4, 3 H–C(13)); 4.58 (*t*, *J*(15α,2) = J_{gem} = 9.1, H_α–C(15)); 3.91 (*dd*, *J*(15β,2) = 8.1, J_{gem} = 9.1, H_β–C(15)). NOE (C_6D_6): H–C(1)→2.33 (5%), 3.41 (2%); H–C(2)→4.41 (4%), 4.15 (8%), 1.39 (6%); H–C(4)→1.90 (5%); H–C(10)→4.41 (8%), 3.41 (5%), 0.73 (8%); H_a–C(12)→2.33 (8%); 3 H–C(13)→2.36 (2%), 4.54 (7%); H_α–C(15)→2.36 (4%), 1.39 (1%); H_β–C(15)→0.73 (3%). ^{13}C-NMR ($CDCl_3$)[3]): 52.88 (*d*, C(1)); 40.77 (*d*, C(2)); 131.37 (*s*, C(3)); 141.87 (*d*, C(4)); 27.87 (*t*, C(5)); 32.72 (*t*, C(6)); 47.75 (*d*, C(7)); 33.95 (*t*, C(8)); 28.10 (*t*, C(9)); 49.51 (*d*, C(10)); 147.16 ((*s*, C(11)); 114.31 (*t*, C(12)); 21.56 (*q*, C(13)); 171.57 (*s*, C(14)); 70.23 (*t*, C(15)). MS: 232 (36, $M^{+\cdot}$), 217 (17, $[M - Me]^+$), 204 (7, $[M - CO]^{+\cdot}$), 190 (32, $[M - C_3H_6]^{+\cdot}$), 189 (43), 188 (11, $[M - CO_2]^{+\cdot}$), 187 (19), 164 (45), 162 (54), 148 (31), 133 (47), 119 (41), 105 (58), 91 (82), 79 (68), 69 (86), 41 (100). HR-MS: 232.1455 ± 0.002 ($C_{15}H_{20}O_2^{+\cdot}$; calc. 232.1463); 190.0989 ± 0.004 ($C_{12}H_{14}O_2^+$; calc. 190.0994); 189.0922 ± 0.004 ($C_{12}H_{13}O_2^+$; calc. 189.0915); 188.1532 ± 0.004 ($C_{14}H_{20}^{+\cdot}$; calc. 188.1565); 188.0840 ± 0.003 ($C_{12}H_{12}O_2^{+\cdot}$; calc. 188.0837); 187.1483 ± 0.003 ($C_{14}H_{19}^+$; calc. 187.1487).

5. *Epoxyrarisetenolide* (= *(6aR*,9R*,9aS*,9bS*,1'S*)-5,6,6a,7,8,9,9a,9b-Octahydro-9-(1-methyloxiran-2-yl)azuleno[4,5-c]furan-3(1H)-one*; **4**). $[\alpha]_D^{20} = -52$ (*c* = 0.24, MeOH). UV (MeOH): 226 (8000). ^{1}H-NMR ($CDCl_3$)[3]): 1.46 (*dt*, *J*(1,10) = 7.5, *J*(1,2) ≈ *J*(1,7) = 11.3, H–C(1)); 3.29 (*tddd*, *J*(2,4) ≈ *J*(2,5α) = 3.4, *J*(2,15α) = 9.0, *J*(2,15β) = 7.8, *J*(2,1) = 11.3, H–C(2)); 7.17 (*td*, *J*(4,5) ≈ *J*(4,2) = 3.4, *J*(4,5β) = 9.0, H–C(4)); 2.14 (*qdd*, *J*(5α,2) ≈ *J*(5α,6α) ≈ *J*(5α,4) = 3.4, *J*(5α,6β) = 12.7, J_{gem} = 16.2, H_α–C(5)); 2.52 (*dddd*, *J*(5β,6β) = 2.2, *J*(5β,6α) = 4.6, *J*(5β,4) = 9.0, J_{gem} = 16.2, H_β–C(5)); 1.99 (*m*, H_α–C(6)); 1.14 (*ddt*, *J*(6β,5β) = 2.2, *J*(6β,7) = 10.3, *J*(6β,5α) ≈ J_{gem} = 12.7, H_β–C(6)); 1.91 (*m*, H–C(7)); 1.85 (*m*, H_α–C(8)); 1.30 (*m*, H_β–C(8)); 2.05 (*m*, H_α–C(9)); 1.85 (*dtd*, *J*(9β,8α) = 3.5, *J*(9β,8β) ≈ *J*(9β,10) = 8.9, J_{gem} = 13.0, H_β–C(9)); 1.90 (*dt*, *J*(10,9α) = 3.7, *J*(10,9β) ≈ *J*(10,1) = 8.6, H–C(10)); 2.69 (br. *d*, J_{gem} = 4.8, H_a–C(12)); 2.61 (*qd*, *J*(12b,Me) = 0.8, J_{gem} = 4.8, H_b–C(12)); 1.35 (*d*, *J*(Me,12b) = 0.8, 3 H–C(13)); 4.57 (*t*, *J*(15α,2) = J_{gem} = 9.0, H_α–C(15)); 3.98 (*dd*, *J*(15β,2) = 7.8, J_{gem} = 9.0, H_β–C(15)). NOE ($CDCl_3$): H–C(2)→4.57 (4%), 1.35 (2%); 3 H–C(13)→3.29 (5%), 2.69 (7%). ^{13}C-NMR ($CDCl_3$)[3]): 50.52 (*d*, C(1)); 40.33 (*d*, C(2)); 131.00 (*s*, C(3)); 142.66 (*d*, C(4)); 27.80 (*t*, C(5)); 33.05 (*t*, C(6)); 47.03 (*d*, C(7)); 33.19 (*t*, C(8)); 25.89 (*t*, C(9)); 48.13 (*d*, C(10)); 58.40 (*s*, C(11)); 55.92 (*t*, C(12)); 21.18 (*q*, C(13)); 171.57 (*s*, C(14)); 70.18 (*t*, C(15)). MS: 248 (5, $M^{+\cdot}$), 233 (4, $[M - Me]^+$), 230 (9, $[M - H_2O]^{+\cdot}$), 215 (7, $[M - Me - H_2O]^+$), 202 (15), 189 (19), 187 (17), 173 (16), 162 (36), 145 (21), 137 (32), 119 (33), 105 (51), 91 (74), 85 (97), 79 (61), 69 (23), 43 (100). HR-MS: 248.1458 ± 0.002 ($C_{15}H_{20}O_3^{+\cdot}$; calc. 248.1451).

6. *Epirarisetenolide* (= *(6aS*,9R*,9aR*,9bR*)-5,6,6a,7,8,9,9a,9b-Octahydro-9-(1-methylethenyl)azuleno-[4,5-c]furan-3(1H)-one*; **5**). UV (MeOH): 226 (9000). ^{1}H-NMR ($CDCl_3$)[3]): 1.42 (*td*, *J*(1,10) ≈ *J*(1,7) = 10.1, *J*(1,2) = 12.9, H–C(1)); 2.95 (*ddtd*, *J*(2,4) = 3.2, *J*(2,5α) = 4.0, *J*(2,15α) ≈ *J*(2,15β) = 9.2, *J*(2,1) = 12.9, H–C(2)); 7.13 (*td*, *J*(4,2) ≈ *J*(4,5α) = 3.2, *J*(4,5β) = 9.0, H–C(4)); 2.18 (*qdd*, *J*(5α,2) ≈ *J*(5α,6α) ≈ *J*(5α,4) = 3.7,

$J(5\alpha,6\beta) = 12.5$, $J_{gem} = 16.4$, H_α–C(5)); 2.54 (*dddd*, $J(5\beta,6\beta) = 2.6$, $J(5\beta,6\alpha) = 4.4$, $J(5\beta,4) = 9.0$, $J_{gem} = 16.4$, H_β–C(5)); 1.96 (*m*, H_α–C(6)); 1.17 (*m*, H_β–C(6)); 1.86 (*m*, H–C(7)); 1.20–2.00 (series of *m*, 2 H–C(8), 2 H–C(9)); 2.49 (*dt*, $J(10,9\beta) = 6.5$, $J(10,9\alpha) \approx J(10,1) = 10.1$, H–C(10)); 4.80 (*qd*, $J(12a,Me) = 1.4$, $J_{gem} = 2.2$, H_a–C(12)); 4.69 (*qd*, $J(12b,Me) = 0.6$, $J_{gem} = 2.2$, H_b–C(12)); 1.70 (*dd*, $J(Me,12b) = 0.6$, $J(Me,12a) = 1.4$, 3 H–C(13)); 4.48 (*t*, $J(15\alpha,2) = J_{gem} = 9.3$, H_α–C(15)); 3.87 (*dd*, $J(15\beta,2) = 9.1$, $J_{gem} = 9.3$, H_β–C(15)). MS: 232 (21, $M^{+\cdot}$), 217 (6), 204 (100), 189 (14), 188 (6, $[M - CO_2]^{+\cdot}$), 187 (8), 170 (40), 161 (44), 91 (69), 79 (58), 69 (41), 41 (82). HR-MS: 232.1450 ± 0.002 ($C_{15}H_{20}O_2^{+\cdot}$; calc. 232.1463).

7. *Epoxidation of Rarisetenolide.* To **3** (4 mg, 0.017 mmol) in dry CH_2Cl_2 (0.5 ml), was added 65% 3-$ClC_6H_4CO_3H$ (4 mg) and 1 mol-equiv. of $NaHCO_3$, and the mixture was stirred overnight at r.t. To this mixture were then added 5% aq. $NaHCO_3$ soln. (1 ml) and hexane (3 ml), and the mixture was subjected to *Whatman* phase separation, recovering the org. phase which was evaporated. The residue contained **4**/**6** 92:8 (^{1}H-NMR), while **3** had disappeared. HPLC (*Si60*, hexane/i-PrOH 95:5) gave pure **6** (t_R 9 min; 0.3 mg, 7%) and **4** (t_R 15 min; 3.4 mg, 80%). NMR and MS of **4**: superimposable to those for nature-isolated epoxyrarisetenolide.

Data of **6** (only data significantly different from those of **4**): ^{1}H-NMR ($CDCl_3$)[3]: 3.41 (*tddd*, $J(2,4) \approx J(2,5\alpha) = 3.9$, $J(2,15\alpha) = 8.0$, $J(2,15\beta) = 9.5$, $J(2,1) = 11.3$, H–C(2)); 7.16 (*td*, $J(4,2) \approx J(4,5\alpha) = 3.9$, $J(4,5\beta) = 9.1$, H–C(4)); 2.44 (*d*, $J_{gem} = 5.1$, H_a–C(12)); 2.40 (br. *d*, $J_{gem} = 5.1$, H_b–C(12)); 4.98 (*dd*, $J(15\beta,2) = 9.5$, $J_{gem} = 9.5$, H_β–C(15)); 3.87 (*dd*, $J(15\alpha,2) = 8.0$, $J_{gem} = 9.5$, H_α–C(15)). EI-MS: 248 (5, $M^{+\cdot}$), 233 (2, $[M - Me]^+$), 230 (10, $[M - H_2O]^{+\cdot}$), 215 (8, $[M - Me - H_2O]^+$), 202 (15), 189 (23), 187 (15), 173 (16), 162 (31), 145 (24), 137 (21), 119 (31), 105 (49), 91 (68), 85 (56), 79 (55), 69 (28), 43 (100).

8. *Treatment of Rarisetenolide* (**3**) *with DIBAL: (3*R*,*6a*S*,*9*S*,*9a*R*,*9b*R*)*- and (3*R*,*6a*R*,*9*R*,*9a*S*,*9b*S*)*-1,3,5,6,6a,7,8,9,9a,9b-Decahydro-9-(1-methylethenyl)azuleno[4,5-c]furan-3-ol* (**7a** and **7b**, resp.) *and (3*R*,*3a*S*,*4*S*,*8a*R*)*-1,2,3,3a,4,7,8,8a-Octahydro-4-(hydroxymethyl)-3-(1-methylethenyl)azulen-5-carbaldehyde* (**8**). To **3** (5 mg, 0.021 mmol) in hexane (0.7 ml) was added 1M DIBAL in THF (3 mol-equiv.). The mixture was stirred at 0° for 2 h and then quenched with H_2O. The org. phase was recovered through a *Whatman* phase-separation filter and evaporated and the residue examined by ^{1}H-NMR, revealing the presence of 30% of unreacted **3**, 60% of **7a**/**7b** 7:3, and 7% of **8** as main products. This mixture was subjected to HPLC (*Si60*, hexane/i-PrOH 97:3) yielding **3** (t_R 5.3 min; 1.4 mg) and **7a**/**7b**/**8** (t_R 9.2 min; 2.0 mg, 60%) in the proportions stated above, as determined by ^{1}H-NMR. **7a**/**7b**/**8**: MS: 234 (50, $M^{+\cdot}$), 219 (7, $[M - Me]^+$), 216 (21, $[M - H_2O]^{+\cdot}$), 201 (38, $[216 - Me]^+$), 191 (76), 187 (22), 173 (49), 170 (30), 163 (28), 161 (16), 147 (29), 145 (32), 141 (31), 133 (33), 118 (54), 105 (53), 91 (79), 77 (69), 67 (60), 41 (100).

Data of **7a** (in the mixture **7a**/**7b**/**8**): ^{1}H-NMR ($CDCl_3$)[3]: 1.42 (*dt*, $J(1,10) = 8.3$, $J(1,2) \approx J(1,7) = 11.3$, H–C(1)); 2.67 (*tddd*, $J(2,4) = J(2,5\alpha) = 3.0$, $J(2,15\alpha) = 8.3$, $J(2,15\beta) = 9.8$, $J(2,1) = 11.2$, H–C(2)); 6.05 (*td*, $J(4,5) \approx J(4,2) = 3.3$, $J(4,5\beta) = 8.9$, H–C(4)); 2.00 (*qdd*, $J(5\alpha,2) \approx J(5\alpha,6\alpha) \approx J(5\alpha,4) = 3.3$, $J(5\alpha,6\beta) = 12.6$, $J_{gem} = 15.5$, H_α–C(5)); 2.27 (*dddd*, $J(5\beta,6\beta) = 2.2$, $J(5\beta,6\alpha) = 4.6$, $J(5\beta,4) = 8.9$, $J_{gem} = 15.5$, H_β–C(5)); 1.99 (*m*, H_α–C(6)); 1.09 (*ddt*, $J(6\beta,5\beta) = 2.2$, $J(6\beta,7) = 10.3$, $J(6\beta,5\alpha) \approx J_{gem} = 12.7$, H_β–C(6)); 1.91 (*m*, H–C(7)); 1.85 (*m*, H_α–C(8)); 1.30 (*m*, H_β–C(8)); 2.05 (*m*, H_α–C(9)); 1.87 (*dtd*, $J(9\beta,8\alpha) = 3.5$, $J(9\beta,8\beta) \approx J(9\beta,10) = 8.9$, $J_{gem} = 13.0$, H_β–C(9)); 2.80 (*dt*, $J(10,9\alpha) = 3.0$, $J(10,9\beta) \approx J(10,1) = 8.3$, H–C(10)); 4.73 (*m*, 2 H–C(12)); 1.76 (*d*, $J(Me,12b) = 0.8$, 3 H–C(13)); 5.58 (*td*, $J(14,4) = J(14,5\alpha) = 1.2$, $J(14,OH) = 5.1$, H–C(14)); 4.32 (*t*, $J(15\alpha,2) = 8.2$, $J_{gem} = 8.2$, H_α–C(15)); 3.76 (*dd*, $J(15\beta,2) = 9.8$, $J_{gem} = 8.2$, H_β–C(15)); 2.42 (*d*, $J(OH,14) = 5.1$, OH). ^{13}C-NMR ($CDCl_3$)[3]: 52.35 (*d*, C(1)); 43.76 (*d*, C(2)); 126.66 (*d*, C(4)); 27.36 (*t*, C(5)); 33.46 (*t*, C(6)); 47.67 (*d*, C(7)); 33.65 (*t*, C(8)); 28.53 (*t*, C(9)); 50.26 (*d*, C(10)); 145.35 (*s*, C(11)); 113.71 (*t*, C(12)); 21.77 (*q*, C(13)); 99.45 (*d*, C(14)); 71.56 (*t*, C(15)).

Data of **7b** (in the mixture **7a**/**7b**/**8**): ^{1}H-NMR ($CDCl_3$; only data significantly different from those of **7a**)[3]: 2.88 *tddd*, $J(2,4) \approx J(2,5\alpha) = 3.0$, $J(2,15\beta) = 6.7$, $J(2,15\alpha) = 8.9$, $J(2,1) = 11.2$, H–C(2)); 6.04 (*td*, $J(4,5) \approx J(4,2) = 3.3$, $J(4,5\beta) = 8.9$, H–C(4)); 2.84 (*dt*, $J(10,9\alpha) = 3.3$, $J(10,9\beta) \approx J(10,1) = 8.3$, H–C(10)); 1.75 (*d*, $J(Me,12b) = 0.8$, 3 H–C(13)); 5.58 (*qd*, $J(14,4) = J(14,5\alpha) = J(14,5\beta) = 1.3$, $J(14,OH) = 6.1$, H–C(14)); 4.40 (*t*, $J(15\alpha,2) = J_{gem} = 8.4$, H_α–C(15)); 3.70 (*dd*, $J(15\beta,2) = 6.7$, $J_{gem} = 8.4$, H_β–C(15)); 2.43 (*d*, $J(OH,14) = 6.1$, OH). ^{13}C-NMR ($CDCl_3$)[3]: 52.49 (*d*, C(1)); 41.94 (*d*, C(2)); 126.50 (*d*, C(4)); 146.05 (*s*, C(11)); 113.74 (*t*, C(12)); 21.84 (*q*, C(13)); 100.39 (*d*, C(14)); 72.08 (*t*, C(15)).

Data of **8** (in the mixture **7a**/**7b**/**8**): ^{1}H-NMR ($CDCl_3$)[3]: 6.85 (*dd*, $J(4,5\alpha) = 6.5$, $J(4,5\beta) = 8.5$, H–C(4)); 9.37 (*s*, OH); signals for the other protons were submerged by those of the major compounds **7a** and **7b**.

9. *Photochemistry of Rarisetenolide: (3a*R*,*4*S*,*6a*R*,*9*S*,*9a*R*,*9b*R*)*- and (3a*ξ*,4*ξ*,6a*R*,*9*S*,*9a*R*,*9b*R*)*-3a,4,5,6,6a,7,8,9,9a,9b-Decahydro-4-methoxy-9-(1-methylethenyl)azuleno[4,5-c]furan-3(1*H*)-one* (**9** and **10**, resp.). A N_2-flushed soln. of **3** (4 mg) in MeOH (3 ml) was irradiated with 254-nm light in a 1-cm optical path

quartz cuvette for 40 min at r.t. Then the mixture was evaporated and the residue subjected to HPLC (*Si60*, hexane/i-PrOH 98.5:1.5): unreacted **3** (40%) and **9** (t_R 10.2 min; 2.4 mg) and **10** (t_R 12.7 min; *ca.* 0.4 mg).

Data of **9**: $[\alpha]_D^{20} = -17$ ($c = 0.08$, MeOH). ^{1}H-NMR ($CDCl_3$)[3]: 2.12 (*dt*, $J(1,10) = 8.2$, $J(1,2) \approx J(1,7) = 10.5$, H–C(1)); 2.58 (*tddd*, $J(2,4) = 2.2$, $J(2,15\alpha) = 9.8$, $J(2,15\beta) = 8.0$, $J(2,1) = J(2,3) = 10.5$, H–C(2)); 2.52 (*dd*, $J(3,4) = 1.5$, $J(3,2) = 10.5$, H–C(3)); 4.04 (*td*, $J(4,3) \approx J(4,5\alpha) = 1.5$, $J(4,5\beta) = 6.1$, H–C(4)); 1.28 (*dddd*, $J(5\alpha,4) = 1.5$, $J(5\alpha,6\alpha) = 3.5$, $J(5\alpha,6\beta) = 11.7$, $J_{gem} = 14.6$, H_α–C(5)); 2.26 (*tdd*, $J(5\beta,6\beta) = J(5\beta,6\alpha) = 3.0$, $J(5\beta,4) = 6.1$, $J_{gem} = 14.6$, H–C(5)); 1.76 (*m*, H_α–C(6)); 1.43 (*dddd*, $J(6\beta,5\beta) = 2.5$, $J(6\beta,7) = 10.6$, $J(6\beta,5\alpha) = 11.7$, $J_{gem} = 13.4$, H_β–C(6)); 1.78 (*tddd*, $J(7,8\alpha) = J(7,6\alpha) = 1.6$, $J(7,8\beta) = 9.2$, $J(7,1) = 11.2$, $J(7,6\beta) = 11.7$, H–C(7)); 2.04 (*m*, H_α–C(8)); 1.30 (*m*, H_β–C(8)); 1.59 (*m*, H_α–C(9)); 1.87 (*ddd*, $J(9\beta,8\beta) = 9.8$, $J(9\beta,10) = 7.6$, $J_{gem} = 13.2$, H_β–C(9)); 2.70 (*dt*, $J(10,9\alpha) = 3.1$, $J(10,9\beta) \approx J(10,1) = 7.6$, H–C(10)); 4.77 (*qd*, $J(12a,Me) = 1.4$, $J_{gem} = 2.2$, H_a–C(12)); 4.67 (*qd*, $J(12b,Me) = 0.6$, $J_{gem} = 2.2$, H_b–C(12)); 1.71 (*dd*, $J(Me,12b) = 0.8$, $J(Me,12a) = 1.4$, 3 H–C(13)); 4.37 (*t*, $J(15\alpha,2) = J_{gem} = 8.0$, H_α–C(15)); 3.86 (*dd*, $J(15\beta,2) = 9.8$, $J_{gem} = 8.0$, H_β–C(15)); 3.27 (*s*, MeO). ^{13}C-NMR ($CDCl_3$)[3]: 51.29 (*d*, C(1)); 39.45 (*d*, C(2)); 49.42 (*d*, C(3)); 81.35 (*d*, C(4)); 30.13 (*t*, C(5)); 30.21 (*t*, C(6)); 44.96 (*d*, C(7)); 33.80 (*t*, C(8)); 31.76 (*t*, C(9)); 51.72 (*d*, C(10)); 146.82 (*s*, C(11)); 113.27 (*t*, C(12)); 21.93 (*q*, C(13)); 180.18 (*s*, C(14)); 74.11 (*t*, C(15)); 57.04 (*q*, MeO). MS: 264 (17, $M^{+\cdot}$), 249 (2, $[M - Me]^+$), 246 (3, $[M - H_2O]^{+\cdot}$), 232 (37, $[M - MeOH]^{+\cdot}$), 217 (14, $[232 - Me]^+$), 204 (14, $[232 - CO]^{+\cdot}$), 189 (21), 187 (19), 173 (15), 164 (15), 162 (13), 147 (22), 133 (25), 119 (33), 105 (44), 91 (50), 79 (50), 71 (100), 41 (88).

Data of **10**: ^{1}H-NMR ($CDCl_3$; only data significantly different from those of **9**)[3]: 2.36 (*m*, H–C(2)); 2.42 (*m*, H–C(3)); 3.70 (*ddd*, $J(4,3) = 7.6$, $J(4,5\alpha) = 2.9$, $J(4,5\beta) = 3.7$, H–C(4)); 1.34 (*m*, H_α–C(5)); 2.21 (*m*, H_β–C(5)); 2.81 (*dt*, $J(10,9\alpha) = 4.1$, $J(10,9\beta) \approx J(10,1) = 9.0$, H–C(10)); 4.71 (br. *s*, 2 H–C(12)); 1.75 (br. *s*, 3 H–C(13)); 4.52 (*dd*, $J(15\alpha,2) = 6.8$, $J_{gem} = 9.1$, H_α–C(15)); 3.75 (*dd*, $J(15\beta,2) = 10.6$, $J_{gem} = 9.1$, H_β–C(15)); 3.41 (*s*, MeO).

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