IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3610, 3420 (OH), 1725, 1650 (C=CCO$_2$R); MS *m/z* (rel. int.): 508 [M]$^+$ (0.3), 490 [M − H$_2$O]$^+$ (4), 408.178 [M − RCO$_2$H]$^+$ (6) ($C_{21}H_{28}O_8$), 83 [C_4H_7CO]$^+$ (100), 55 [83 − CO]$^+$ (41);

$$[\alpha]_{24°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\ nm}{+4.4 \quad +5.0 \quad +5.3 \quad +8.4}\ (CHCl_3;\ c = 1.0).$$

Saponification (MeOH–H$_2$O–KOH, 70°) afforded L-inositol.

L-Inositol-2, 3, 5, 6-tetra angelate and myoinositol-1, 3, 4, 6-tetra angelate. (**6** *and* **7**). Colourless gum, which could not be separated into its constituents, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3610, 3480 (OH), 1720, 1650 (C=CCO$_2$R); MS *m/z* (rel. int.): 508 [M]$^+$ (0.3), 490 [M–H$_2$O]$^+$ (2), 408.178 [M–RCO$_2$H]$^+$ (5) ($C_{14}H_{28}O_8$) 83 [C_4H_7CO]$^+$ (100), 55 [83 − CO]$^+$ (44).

Myoinositol-2, 4, 5, 6-tetra angelate (**8a**). Colourless gum, IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3610, 3440 (OH), 1730, 1250 (C=CCO$_2$R); MS *m/z* (rel. int.): 508 [M]$^+$ (0.3), 490 (4), 408.178 [M − RCO$_2$H]$^+$ (6) ($C_{21}H_{28}O_8$), 83 [C_4H_7CO]$^+$ (100), 55 [83 − CO]$^+$ (40);

$$[\alpha]_{24°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\ nm}{+3.4 \quad +3.8 \quad +4.0 \quad +6.0}\ (CHCl_3;\ c\ 0.74).$$

8 mg **8a** were heated with 0.1 ml Ac_2O for 1 hr at 70°. TLC (CH_2Cl_2–C_6H_6–Et_2O, 2:2:1) afforded 4 mg **8b**, colourless gum, MS *m/z* (rel. int.): 592 [M]$^+$ (0.2), 532 [M − HOAc]$^+$ (4), 492 [M − RCO$_2$H]$^+$ (10), 83 [C_4H_7CO]$^+$ (100).

Acknowledgements—We thank Dr. R. Gupta, University of Delhi, for plant material and the Deutsche Forschungsgemeinschaft for financial support.

REFERENCES

1. Baruah, N. C., Sharma, R. P., Thyagarajan, G. and Herz, W. (1979) *Phytochemistry* **18**, 2003.
2. Plouvier, V. (1963) in *Chemical Plant Taxonomy* (Swain, T., ed.) p. 323. Academic Press, London.
3. Bohlmann, F. and Zdero, C. (1977) *Phytochemistry* **16**, 1243
4. Bohlmann, F., Mahanta, P. K., Jakupovic, J., Rastogi, R. C. and Natu, A. A. (1978) *Phytochemistry* **17**, 1165.

Phytochemistry, Vol. 21, No. 3, pp. 782–784, 1982.
Printed in Great Britain.

0031–9422/82/030782–03$03.00/0

THREE SQUALENE DERIVATIVES FROM *CAULERPA PROLIFERA*

LORENZO DE NAPOLI, ERNESTO FATTORUSSO*, SILVANA MAGNO and LUCIANO MAYOL

Istituto di Chimica Organica e Biologica dell'Università di Napoli, Via Mezzocannone 16, 80134 Napoli, Italy; *Istituto di Chimica Biorganica dell'Università di Napoli, Via L. Rodinò 22, 80138 Napoli, Italy

(*Revised received* 24 *July* 1981)

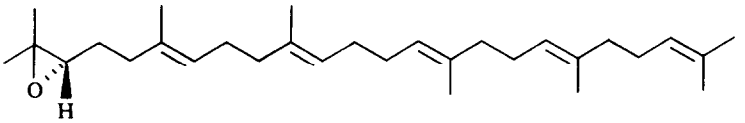
Key Word Index—*Caulerpa prolifera*; Caulerpaceae; (6*S*, 7*S*)-squalene-6,7-epoxide; (10*S*, 11*S*)-squalene-10, 11-epoxide; all-*trans*-(3*S*)-2,6,10,15,19,23-hexamethyltetracosa-1,6,10,14,18,22-hexaen-3-ol.

Abstract—(6*S*, 7*S*)-Squalene-6,7-epoxide, (10*S*, 11*S*)-squalene-10,11-epoxide and all-*trans*-(3*S*)-2,6,10,15,19,23-hexamethyltetracosa-1,6,10,14,18,22-hexaen-3-ol have been isolated from the marine alga *Caulerpa prolifera*.

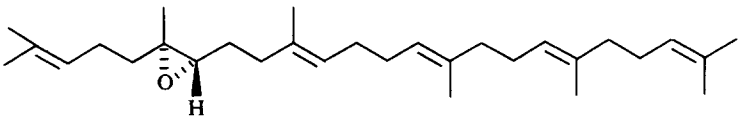
Investigations of marine green algae of the family Caulerpaceae have resulted in the isolation of di- and sesquiterpenes [1–4]. From a member of this family, *Caulerpa prolifera*, we recently isolated (*S*)-(−)-squalene-2,3-epoxide (**1**) [5], in addition to two acetylenic sesquiterpenes [1, 2]. This is the first report of the occurrence in nature of **1**, which is known to play an important role in the biogenesis of triterpenes and sterols. We wish to report here that this alga produces, in minor amounts, three structurally related compounds, the epoxides **2**, **3** and the allylic alcohol **4**. Only **3** was previously isolated from a natural source (mycelia of *Sclerotinia fructicola*), but its stereochemistry was not determined [6].

The alga was freeze-dried and the chloroform extracts were purified by Si gel column chromatography. Individual components were then obtained by rechromatography on Si gel and by preparative $AgNO_3$–Si gel TLC.

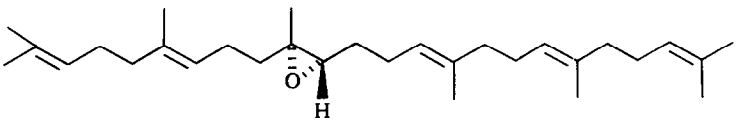
Compounds **2**, $[\alpha]_D^{20}$ − 11.2°, and **3**, $[\alpha]_D^{20}$ − 3.8°, on



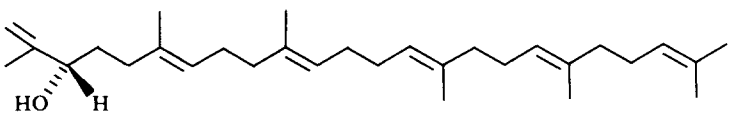
1



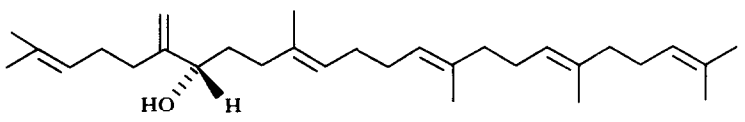
2



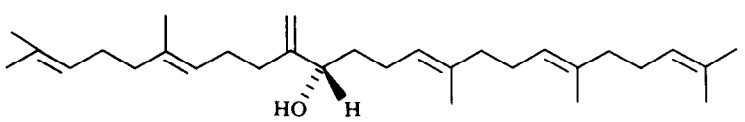
3



4



5



6

the basis of their mass spectra [6] and ^{1}H NMR spectra, were tentatively identified as squalene-6,7-epoxide and squalene-10,11-epoxide, respectively. These identifications were confirmed by comparison of their spectroscopic and chromatographic properties with those of two samples synthesized from all-*trans*-squalene with *m*-chloroperbenzoic acid according to Katayama and Marumo [6].

In order to clarify the chirality of C-6 and C-7 in **2** and C-10 and C-11 in **3**, these compounds were treated with aluminium isopropoxide in boiling toluene. This highly regioselective reaction [7] afforded the two allylic alcohols **5** and **6**. Application of the GC modification of the Horeau method according to Brooks and Gilbert [8] allowed the chirality of C-7 in **5** and C-11 in **6** to be determined as *S*, thus revealing the stereochemistry of C-7 in **2** and C-11 in **3**.

The absolute configurations of the tertiary carbon atoms of the oxirane rings in **2** and **3** were established by the following observation. The synthetic squalene-6,7-epoxide and squalene-10,11-epoxide were identical chromatographically and spectroscopically to **2** and **3**, and on the basis of the stereochemistry of the epoxidation with peracids [9], these must be racemic mixtures of (6*R*,7*R*)- and (6*S*,7*S*)-squalene-6,7-epoxide and (10*R*,11*R*)- and (10*S*,11*S*)-squalene-10,11-epoxide. Considering that the stereochemistry of C-6 in **2** and C-10 in **3** was established as *S*, **2** and **3** must be (6*S*,7*S*)-squalene-6,7-epoxide and (10*S*,11*S*)-squalene-10,11-epoxide, respectively.

Compound **4**, isolated in a yield of 0.003%, $[\alpha]_D^{20}$ −10.0°, had the molecular formula $C_{30}H_{50}O$ (precise mass measurement).

Its ^{1}H NMR spectrum indicated the presence of seven vinylic methyl groups (a set of seven overlap-

ping signals from δ 1.73 and 1.60, 21 H), seven olefinic protons (5.15, 5H, complex signals; 4.94, 1H, *m*; $W_{1/2} = 6$ Hz; 4.81, 1H, *m*; $W_{1/2} = 7$ Hz) and one allylic $\rangle$CHOH group (4.05, 1H, *t*, $J = 6$ Hz).

On the basis of these data, structure **4** was tentatively assigned to this compound. This assignment was confirmed by comparison with an authentic sample prepared from (*S*)-(−)-all-*trans*-squalene-2,3-epoxide, isolated from the same source, by treatment with aluminium isopropoxide in boiling toluene.

EXPERIMENTAL

General procedures. The ^{1}H NMR spectra were measured at 270 MHz with TMS as internal standard; $CDCl_3$ was used as solvent. Mass spectra were taken with a direct inlet system.

Extraction and isolation of constituents. The alga (700 g fr. wt) collected in June 1980 in the Bay of Salerno, Italy, was freeze-dried and ground to a fine powder with a blender. The dried alga was extracted ×3 with $CHCl_3$ with continuous stirring. The extracts were combined and evapd to give a dark-green oil (15 g). The crude extract was applied to a column (5 × 120 cm) of Si gel. The column was eluted with a solvent gradient from hexane to hexane–Et_2O (9:1). Fractions of 200 ml were collected. The following compounds were isolated in order of increasing polarity.

(6*S*,7*S*)-*Squalene*-6,7-*epoxide* (**2**) *and* (10S,11S)-*squalene*-10,11-*epoxide* (**3**). Fractions 20–22 were pooled and subjected to column chromatography (Si gel, 1 × 60 cm) using as eluant a gradient system of hexane–C_6H_6 from 8:2 to 6:4. Fractions of 20 ml were collected. Fractions 19–21, evapd to dryness, afforded 25 mg of an oily material which was further fractionated by prep. TLC ($AgNO_3$–Si gel, C_6H_6–EtOAc, 7:3) to give pure **2** (6.5 mg) and **3** (8 mg).

2, $[\alpha]_D^{20} - 11.2°$ ($CHCl_3$; *c* 0.100), M^+ 426 *m/z*, δ 5.12 (5H, *br*, vinyl protons), 2.71 (1H, *t*, J = 6 Hz, H-7) and 1.26 (3H, *s*, Me-6).

3, $[\alpha]_D^{20} - 3.8°$($CHCl_3$; *c* 0.100), M^+ 426 *m/z*, δ 5.11 (5H, *br*, vinyl protons), 2.74 (1H, *t*, $J = 6$ Hz, H-11) and 1.26 (3H, *s*, Me-10).

All - trans - (3S) - 2,6,10,15,19,23 - *hexamethyltetracosa* - 1,6,10,14,18,22-*hexaen*-3-*ol* (**4**). Fractions 28–30 were rechromatographed on prep. TLC (Si gel hexane–Et_2O, 9:1) to give **4** as an oily residue (21 mg), $[\alpha]_D^{20} - 10.0°$ ($CHCl_3$; *c* 0.100); MS *m/z* 426.3841 (M^+, calc. for $C_{30}H_{50}O$, 426.3860).

Treatment of **1**, **2** *and* **3** *with aluminium isopropoxide.* To a soln of **1** (20 mg) in 3 ml dry toluene, $Al(OCHMe_2)_3$ (15 mg) was added and the mixture refluxed for 10 hr. After addition of H_2O the organic phase was separated and taken to dryness. The crude product was purified by prep. TLC (Si gel, hexane–EtOAc, 17:3) to give 14 mg of **4**.

2 (3 mg) and **3** (3 mg) were treated with $Al(OCHMe_2)_3$ and the reaction mixtures purified in the same experimental conditions described above to give **5** (2 mg; M^+ *m/z* 426; δ 5.13, 1H, *m*, $W_{1/2} = 6$ Hz and δ 4.94, 1H, *m*, $W_{1/2} = 6$ Hz, $=CH_2$; δ 4.04, 1H, *t*, $J = 7$ Hz, $\rangle$CHOH) and **6** (2 mg, M^+ *m/z* 426; δ 5.00, 1H, *m*, $W_{1/2} = 6$ Hz and 4.93, 1H, *m*, $W_{1/2} = 6$ Hz, $=CH_2$; 4.04, 1H, *t*, $J = 7$ Hz, $\rangle$CHOH).

Application of the GC modification of the Horeau method to **4** *and* **5**. Compound **4** (2 mg) in dry pyridine (4 μl) was treated with an excess (±)-α-phenylbutyric anhydride and was kept at 40° for 1.5 hr. Conventional work-up [8] led to the isolation of a preponderance of (*R*)-α-phenylbutyric acid.

Compound **5** (2 mg) in the same experimental conditions described above also gave a preponderance of (*R*)-α-phenylbutyric acid.

Acknowledgements—This work is a result of research sponsored by the Consiglio Nazionale delle Ricerche (Rome) in the Programme of the 'Progetto Finalizzato per l'Oceanografia e i Fondi Marini'. Thanks are due to the Centro di Metodologie Chimico-fisiche of the University of Naples (Mr. I. Giudicianni) for the determination of ^{1}H NMR spectra.

REFERENCES

1. Blackman, A. J. and Wells, R. J. (1976) *Tetrahedron Letters* 2729.
2. Blackman, A. J. and Wells, R. J. (1978) *Tetrahedron Letters* 3063.
3. Amico, V., Oriente, G., Piattelli, M., Tringali, C., Fattorusso, E., Magno, S. and Mayol, L. (1978) *Tetrahedron Letters* 3593.
4. De Napoli, L., Fattorusso, E., Magno, S. and Mayol, L. *Experientia* (in press).
5. De Napoli, L., Fattorusso, E., Magno, S. and Mayol, L. (1980) *Tetrahedron Letters* 2917.
6. Katayama, M. and Marumo, S. (1979) *Tetrahedron Letters* 1293.
7. Terao, S., Shiraishi, M. and Kato, K. (1979) *Jpn. Kokai Tokkyo Koho* 7, **76**, 507.
8. Brooks, C. J. W. and Gilbert, J. D. (1973) *J. Chem. Soc. Chem. Commun.* 194.
9. Gould, E. S. (1959) *Mechanism and Structure in Organic Chemistry*, p. 534. Holt, Rinehart & Winston, New York.