Tetrahedron Letters, Vol. 34, No. 12, pp. 1985-1986, 1993
Printed in Great Britain

0040-4039/93 $6.00 + .00
Pergamon Press Ltd

New Antiproliferative 9,11-Secosterol from Soft Coral *Gersemia fruticosa*

Reet Koljak, Tõnis Pehk[a], Ivar Järving, Milana Liiv, Annika Lopp, Külliki Varvas, Aino Vahemets, Ülo Lille, Nigulas Samel*

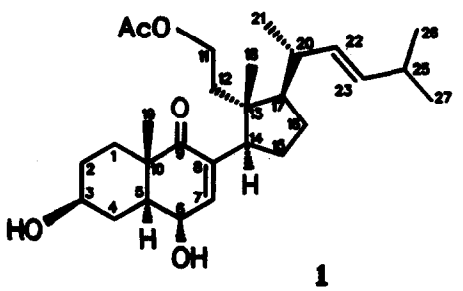
Institute of Chemistry, Estonian Academy of Sciences, Akadeemia tee 15, EE0026 Tallinn, ESTONIA
[a]Institute of Chemical Physics and Biophysics, Estonian Academy of Sciences, Rävala pst. 10, EE0100 Tallinn, ESTONIA

Abstract: A new highly antiproliferative 9,11-secosterol 1 was isolated from the White Sea soft coral *Gersemia fruticosa*. The structure was established by spectroscopic data.

In the course of our screening program for bioactive compounds from cold sea marine organisms a new 9,11-secosterol 1 with antiproliferative activity was isolated from *Gersemia fruticosa (Nephdeidae, Alcynacea)*. The growth inhibitory activity (IC_{50}) estimated on two different tumor cell lines *in vitro* was 1 and 3 μg/ml for Ehrlich carcinoma and human erythroleukemia K-562 cells, respectively.

The EtOAc extract of freeze-dried animals (collected at the Gulf of Kandalaksha, White Sea, Arctic Circle, at a depth of 20-25m and temperature about 0°C) was partitioned between hexane and EtOH/0.2N $KHCO_3$ in water (1:1, v/v). The basic layer, extracted after acidification with EtOAc, was repeatedly chromatographed on silica gel (EtOAc/Hex, Acetone/$CHCl_3$, i-PrOH/Hex) followed by RP-HPLC (CH_3CN/H_2O) to give pure 24-nor-9,11-seco-11-acetoxy-3β,6α-dihydroxycholesta-7,22(E)-dien-9-one 1 as colourless oil, $[\alpha]_D^{22}$ +23° (c 0.17, MeOH), UV (EtOH) λ_{max} 239 (ϵ 4,000) nm.

The molecular formula of 1 was established as $C_{28}H_{44}O_5$ by HRMS (m/z 460.3192; calcd. 460.3189). Its structure was elucidated by ^1H and ^{13}C NMR 1D and 2D NMR spectroscopy on a Bruker AMX-500 instrument. ^1H-^1H COSY, ^1H-^{13}C COSY and inverse detected long-range ^1H-^{13}C 2D correlations were measured using standard pulse programs. The inverse detected long-range correlation was crucial to complete the structure via the quaternary carbons. The chemical shifts of 1 in $CDCl_3$ and C_6D_6 solutions are reported in Table.



1

The trans junction of A/B rings was determined by the ^{13}C chemical shifts of C-5, C-10 and C-19. An equatorial β-orientation of OH at C-3 follows from a comparison with other sterols[1] and the recently found 9,11-secosterol (3β,6α-dihydroxy-9-oxo-

9,11-secocolest-7-en-11-al) $[\alpha]_D$ +5.0° (c=0.2, $CHCl_3$)[2]. The hydroxyl group at C-6 is α-oriented due to 9.8 Hz coupling between H-6 and H-5. The side chain double bond has E configuration on the basis of ^{13}C chemical shifts of model olefins[3] and 15.4 Hz vicinal coupling between the olefinic protons. The orientation of substituents at C-13, C-14 and C-17 is the same as in related 9,11-secosterol[2] and herbasterol[4] due to the close values of carbon chemical shifts around these carbon atoms. The ichthyotoxic herbasterol (9,11-seco-2β,3α,6β,11,19-pentahydroxy-5β-cholesta-9-one)[4] is the only marine 9,11-secosterol so far isolated (see also[5]) whose biological activity has been reported.

Table. ^{13}C and ^{1}H Chemical Shifts of 24-nor-9,11-seco-11-acetoxy-3β,6α-dihydroxycholesta-7,22(E)-dien-9-one

	in $CDCl_3$(a		in C_6D_6(b	
Atoms	δ ^{13}C	δ ^{1}H	δ ^{13}C	δ ^{1}H
1	31.74	1.89 / 1.47	32.34	1.91 / 1.41
2	30.41	1.92 / 1.46	30.92	1.60 / 1.23
3	69.96	3.59	69.86	3.11
4	32.71	2.30 / 1.44	33.07	2.07 / 1.11
5	48.57	1.77	48.66	1.36
6	69.37	4.27	69.19	3.68
7	146.18	6.52	146.60	6.25
8	136.72	-	136.49	-
9	203.69	-	203.13	-
10	44.70	-	44.77	-
11	61.47	4.17	61.65	4.44 / 4.48
12	36.90	1.62 / 1.24	37.44	1.85 / 1.49
13	45.86	-	46.23	-
14	42.63	3.25	42.92	3.40
15	25.47	1.72 / 1.46	25.75	1.57 / 1.40
16	26.74	1.57	26.94	1.58
17	50.69	1.71	50.96	1.72
18	17.31	0.67	17.60	0.61
19	16.08	1.12	15.98	0.85
20	38.35	2.16	38.72	2.17
21	21.41	1.04	21.74	1.10
22	131.97	5.25	132.82	5.27
23	136.00	5.29	136.08	5.31
25	31.02	2.19	31.47	2.23
26	22.66	0.95	22.89	1.00
27	22.55	0.95	22.80	1.01
OAc	21.17	2.02	20.71	1.71
	171.24	-	171.93	-

a) Solvent at 77.0 and 7.27 ppm
b) Solvent at 128.0 and 7.15 ppm

Acknowledgements

The coral samples were collected and identified by Drs. Vladimir Buryakov and Aleksei Sukhotin, Kartesh White Sea Biological Station. We also thank Dr. Jorma Matikainen, Helsinki University, for HRMS study.

References

1. Blunt, J. W.; Stothers, J. B. *Org. Magn. Reson.*, **1977**, 9, 439.
2. Migliuolo, A.; Piccialli, V.; Sica, D. *Tetrahedron*, **1991**, 47, 7937.
3. De Haan, J. W.; Van de Ven, L. J. M. *Org. Magn. Reson.*, **1973**, 5, 147.
4. Capon, R. J.; Faulkner, D. J. *J. Org. Chem.*, **1985**, 50, 4771.
5. (a) Enwall, E. L.; Van der Helm, D.; Nan Hsu, I.; Pattabhiraman, T.; Schmitz, F. J.; Spraggins, R. L.; Weinheimer, A.J. *J. Chem. Soc. Chem. Commun.*, **1972**, 215. (b) Kazlauskas, R.; Murphy, P. T.; Ravi, B. N.; Sanders, R. L.; Wells, R. J. *Aust. J. Chem.*, **1982**, 35, 69. (c) Bonini, C.; Cooper, C. B.; Kazlauskas, R.; Wells, R. J.; Djerassi, C. *J. Org. Chem.*, **1983**, 48, 2108.

(Received in UK 13 November 1992)