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APPLICATIONS OF COSY AND HOMONUCLEAR RELAY 2D-NMR IN THE DETERMINATION OF THE STRUCTURE OF A NEW CEMBRANE ISOLATED FROM THE MOLLUSC PLANAXIS SULCATUS[1]

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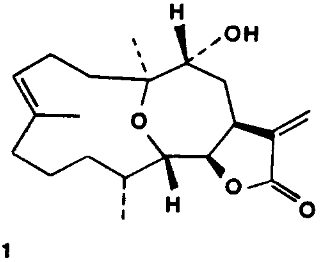
Abstract. Proton-proton homonuclear chemical shift correlation (COSY) and homonuclear relayed coherence transfer (RELAY) have been used to establish the structure of 3,11-diacetoxy-15,16-dehydrocembran-6-one.

Numerous cembranoid diterpenes have been isolated from a variety of marine and terrestrial sources.[2] Several recent publications have reported the isolation of cembranoid compounds from various species of mollusc. Coll and co-workers[3,4] have isolated a metabolite of sarcophytoxide, 7,8-deoxysarcophytoxide from the mollusc Ovula ovum. Previous work in these laboratories has led to the isolation of the oxa-bridged cembranoid diterpene jeunicin[5] (1) and more recently the novel hydroperoxide planaxool[6] (2). A less polar fraction from P. sulcatus has now been investigated and found to contain a further cembranoid diterpene which has been identified as 3,11-diacetoxy-15,16-dehydrocembran-6-one (3) on the basis of two-dimensional nmr experiments.



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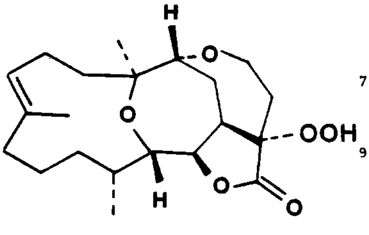

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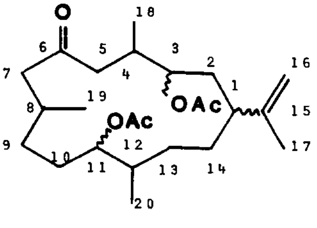

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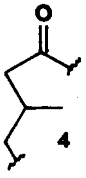
The new compound, 3, $C_{24}H_{40}O_5$,[7] was isolated in a yield of 25 mg from an i-PrOH extract of *P. sulcatus*.[8] The i-PrOH extract (9.7g) was triturated with hexanes followed by $CHCl_3$. The residue from the chloroform extract was chromatographed over an LH-20 column (eluting solvent: CH_2Cl_2/MeOH (9/1 v/v). Fractions (30-38, 4ml each) containing desired compounds were combined and evaporated to dryness; recrystallization from $CHCl_3$ gave colorless needles, mp 145°, $|\alpha|_D^{22} = -4.0°$. Analysis of the spectral data confirmed that 3 contained a ketone and exomethylene group (^{13}C-nmr, 209.7 and 109.4(t, APT)δ $CDCl_3$)[7]; and two aceoxyl groups ($^{13}C/^{1}H\delta$ - methine resonances at 76.0/4.76 and 77.2/4.69δ; acetyl methyls 21.1/2.04 and 20.9/2.02δ).

The COSY[9] and RELAY[10,11] 2D-nmr spectra of 3 in $CDCl_3$ are shown in Figure 1. Proton connectivities shown in 4 and 5 were deduced from the COSY spectrum. Similarities in the proton chemical shifts in the C9-C14 region of the structure precluded the exclusive use of the COSY experiment for the entire structure elucidation. Instead, it was necessary to supplement the data obtained from the COSY spectrum by homonuclear relay[10] with fixed delays for magnetiztion transfer optimized at 3 and 5 Hz. In this fashion, the structure of fragment 6 was established. It should also be noted that a single RELAY spectrum alone was insufficient to completely elucidate the proton-proton connectivity network. However, the combined usage of the 3 and 5 Hz optimized RELAY spectra did successfully establish all of the connectivities. Finally, the RELAY spectra also allowed the orientation of fragment 6 relative to fragments 4 and 5 thereby completing the structure elucidation.



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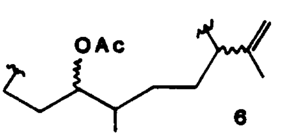

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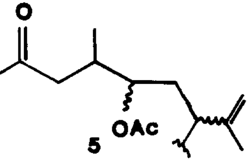

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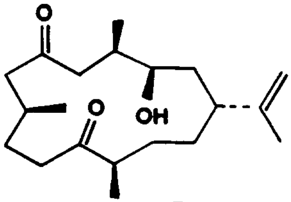

7

The positions of oxygen functions in 3 are identical with those of plexaurolone (7)[12] which was isolated from a *Plexaura* gorgonian collected in the Caribbean. It is probable that the mollusc acquired 3 or a precursor such as 7 from a dietary source. Similarly, the presence of jeunicin (1), obtained previously only from Caribbean gorgonians, and its apparent metabolite 2 in the same mollusc, suggests that there may be greater similarity in the cembranoids present in Australian soft corals and Caribbean gorgonians than previously appreciated. Further work on the corals themselves will, of course, be necessary to determine the extent of similarities in the diterpenes produced by Australian and Caribbean species.

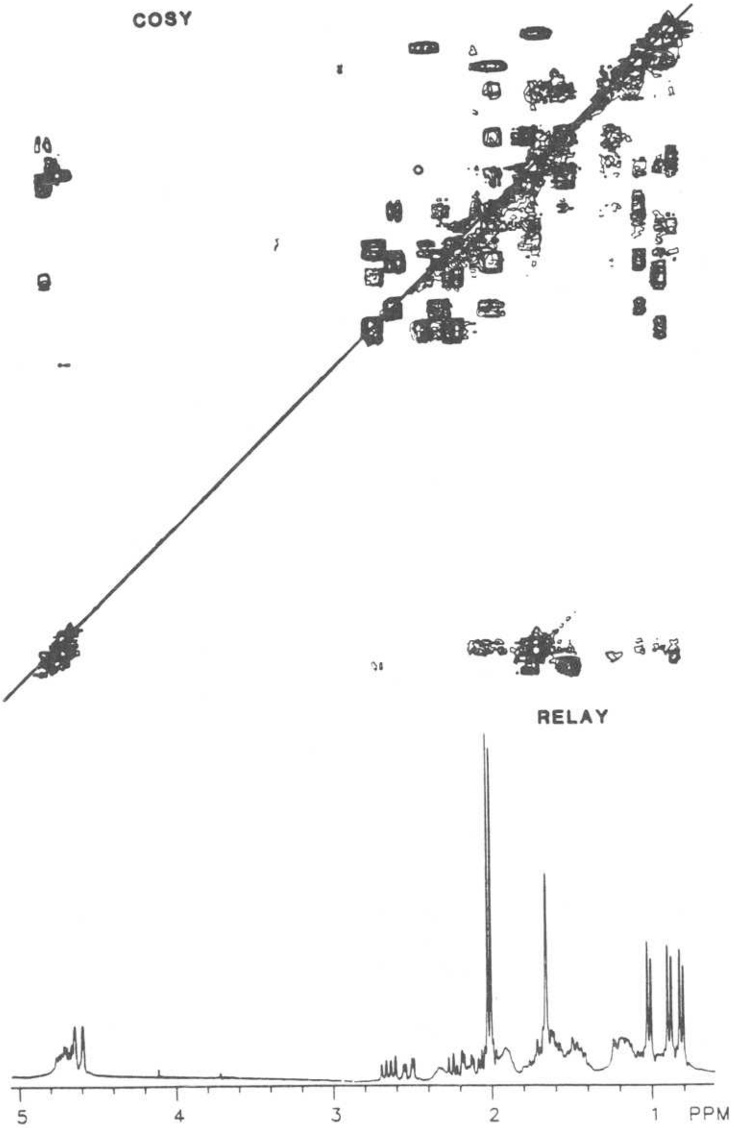

Figure 1. Composite contour plot showing the COSY and RELAY spectra of **3**. The RELAY spectrum shown was optimized for a 3 Hz magnetization transfer ($\tau=1/4J_{HH}=83.3$ msec). A second RELAY spectrum (not shown) was also performed using a 5 Hz transfer ($\tau=50$ msec) and was necessary to complete the structure elucidation because of the similarities of a number of the proton chemical shifts in the structural fragment shown by **6**.

Acknowledgements

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References

1. Part VI in the series Two-Dimensional NMR Studies of Marine Natural Products.
2. For a review, see: A.J. Weinheimer, C.W.J. Chang and J.A. Matson, *Fortschr. Chem. Org. Naturst.*, **36**, 285 (1979).
3. J.C. Coll, D.M. Tapiolas, B.F. Bowden, L. Webb and H. Marsh, *Marine Biol.*, **74**, 35 (1983).
4. J.C. Coll, B.F. Bowden, D.M. Tapiolas, R.H. Willis, P.Djura, M.Streamer and L. Trott, *Tetrahedron*, **41**, 1085.
5. R. Sanduja, G.S. Linz, E.L. Ezell, M. Alam, A.J. Weinheimer and G.E. Martin, *J. Heterocyclic Chem.*, **23**, in press (1986).
6. R. Sanduja, G.S. Linz, A.J. Weinheimer, M. Alam and G.E. Martin, *J. Org. Chem.*, submitted.
7. A total ^1H and ^{13}C nmr spectral assignment was obtained (multiplicities were determined using the APT experiment (S.L. Patt and J.N. Shoolery ,*J. Magn. Reson.*, **46**, 535 (1982)) (position-$\delta^{13}C$/mult./δ^1H): 1-43.7/d/1.91; 2-36.3/t/1.72,1.48; 3-76.0/d/4.76; 4-33.8/d/2.34; 5-47.4/t/2.67,2.16; 6-209.7/s/—; 7-49.8/t/2.54, 2.27; 8-30.0/d/1.92; 9-28.4/t/1.43, 1.20; 10-28.8/t/1.65, 1.59; 11-77.2/d/4.69; 12-35.4/d/1.65; 13-30.5/t/1.51, 1.17; 14-29.4/t/1.23, 1.07; 15-148.9/s/—; 16-20.2/q/1.67; 17-109.4/t/4.66, 4.61; 18-15.6/q/0.91; 19-20.8/q/1.03; 20-16.1/q/0.83; (acetyls were assembled using long range optimized proton-carbon chemical shift correlation but were not specifically linked to either the 3 or 12 positions) 170.4/s/— and 21.1/q/2.04; 170.7/s/— and 20.9/q/2.02.
8. The organism *Planaxis sulcatus* was collected of Heron Island, Australia in 1974.
9. R. Freeman, G.Morris, A. Bax, *J. Magn. Reson.*, **42**, 164 (1981)
10. G. Eich, G. Bodenhausen and R.R. Ernst, *J. Am. Chem.Soc.*, **104**, 3731 (1982).
11. A. Bax and G. Drobny, *J. Magn. Reson.*, **61**, 306 (1985)
12. S. E. Ealick, D. van der Helm, R. A. Gross, Jr., A. J. Weinheimer, L. S. Ciereszko and R. E. Middlebrook, *Acta Cryst.*, **B36**, 1901 (1980).

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