

GRIMALDONE, A TRICYCLIC SESQUITERPENOID FROM *MANNIA FRAGRANS*. CRYSTAL STRUCTURE ANALYSIS.

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Abstract—The structure of grimaldone, a tricyclic sesquiterpenoid from the liverwort *Mannia fragrans*, has been established by crystal structure analysis. Its absolute configuration was assigned by comparison of its CD spectrum with that of (+)-*S*- α -cuparenone.

INTRODUCTION

In Central Europe the genus *Mannia* (Marchantiales) comprises three species, *M. pilosa*, *M. triandra* and, the most common, *M. fragrans* (Balbis) Frye et Clark. *M. fragrans* grows on both siliceous and calcareous soils or sunny cliffs. It occurs in only a few locations in the G.D.R., Czechoslovakia and Poland but is abundant in Hungary, especially near Lake Balaton and north Budapest. As its name suggests *M. fragrans* emits a pleasant and intense odour when the thallus is wet and a sensitive nose can detect it at a distance of many metres. In contrast, in its dry state the plant has a very weak odour and is difficult to distinguish from the surrounding soil [1, 2].

From *M. fragrans* collected at Balatonfüred in Hungary Huneck and Schreiber [3] isolated the odoriferous principle, C₁₅H₂₂O, mp 91–92° and named it grimaldone after the old genus name *Grimaldia*. A year later Benesová [4] found (–)-*R*- α -cuparenone in *M. fragrans* collected in Southern Moravia but could not isolate grimaldone. Our supply (10 mg) of grimaldone disappeared from a vial with a plastic stopper in the course of a year because of the high vapour pressure of this sesquiterpenoid—it sublimes readily at room temperature. The next opportunity to collect larger amounts of the liverwort came to one of us (S.H.) during botanical expeditions to Mongolia in 1978 and 1983. Extraction of the plant material followed by chromatography afforded grimaldone.

RESULTS AND DISCUSSION

Grimaldone (1) has resonances in its ¹H and ¹³C NMR spectra for three tertiary methyls [δ_H 0.96 (*d*, *J* 0.9 Hz), 1.01 and 1.05; δ_C 19.5, 21.1 and 21.2], a trisubstituted cyclopropane [δ_H 0.75 (*dd*, *J* = 4.0, 5.5 Hz), 1.62 (*dd*, *J* = 4.0, 8.4 Hz), 1.23 (*ddd*, *J* = 2.0, 5.5, 8.4 Hz); δ_C 16.1 (*t*), 27.2 (*d*), 37.1 (*s*)] and an exomethylene group [δ_H 4.66 and 4.85; δ_C 153.2 (*s*), 102.4 (*t*)]. The molecule also contains four methylenes [δ_C 27.9, 28.9, 29.2, 33.7], two quaternary carbons [δ_C 45.4, 53.4] and a cyclopentanone [δ_C 223.2, ν_{max} (CCl₄) 1742 cm^{–1}]. Thus grimaldone is

tricyclic. The UV spectrum [λ_{max}^{MeOH} 222 nm (ϵ 2630)] suggests that the cyclopropane and the exomethylene group are conjugated. Decoupling studies at 360 MHz enabled some of the proton connectivities to be established despite considerable overlap. Thus there is a methylene group [δ_H 2.35 (*ddd*, *J* = 2.7, 10.0, 19.7 Hz), 2.25 (*ddd*, *J* = 9.0, 9.8, 19.7 Hz)] α to the carbonyl group with a neighbouring isolated methylene group [δ_H 1.78 (*ddd*, *J* 9.8, 10.0, 12.6 Hz), 1.22 (*ddd*, *J* = 2.7, 9.0, 12.6 Hz)]. The cyclopropyl methine proton is coupled (*J* = 2.0 Hz) to one proton [δ_H 2.01 (*m*, clearly including *d*, *J* = 2.0 Hz)] of a further –CH₂CH₂–system [δ_H 2.18 (*m*), 2.09 (*m*, clearly including allylic couplings *t*, *J* = 2.4 Hz) and 1.75 (*m*, clearly including weak unresolved allylic couplings)]. Initially we interpreted this as a vicinal coupling and consequently we were unable to derive a rational structure for grimaldone. The problem was solved by a single crystal X-ray analysis which revealed the structure and relative stereochemistry of grimaldone as in 1. An ORTEP diagram of the molecule is shown in Fig. 1. Both five-membered rings adopt twisted conformations and the bicyclohexane group is pseudoequatorial to the cyclopentanone. With hindsight it is clear that the coupling between the cyclopropyl methine and the methylene proton is a ⁴*J* interaction.

The absolute configuration of grimaldone as in 1 follows from comparison of its CD spectrum (Fig. 2) with that of (+)- α -cuparenone (2) which is assumed to have its *p*-tolyl group pseudoequatorial to the cyclopentanone ring. Such a twisted conformation would give rise [5] to a positive CD for 2 in agreement with the observed sign. Similarly the conformation of grimaldone 1 based on the X-ray crystal structure would give rise to a negative CD. Thus grimaldone is (–)-6S, 8R-cyclo-5R-cupar-9(15)-en-2-one, a derivative of *ent*-cuparane, and provides a further example of the capacity of liverworts to produce sesquiterpenoids enantiomerically related to those of higher plants [6]. Grimaldone is the first example of a sesquiterpenoid with this tricyclic carbon skeleton [7]. Recently Asakawa and his colleagues [8] have isolated the alcohol 3 from *Marchantia paleacea* Bertol. var *dip-tera* (Mont) Hatt.

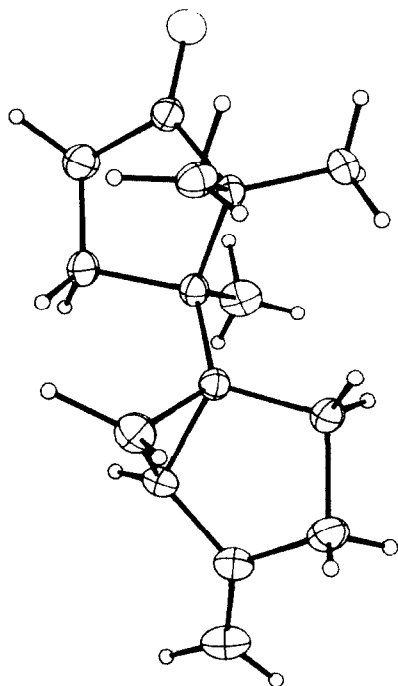
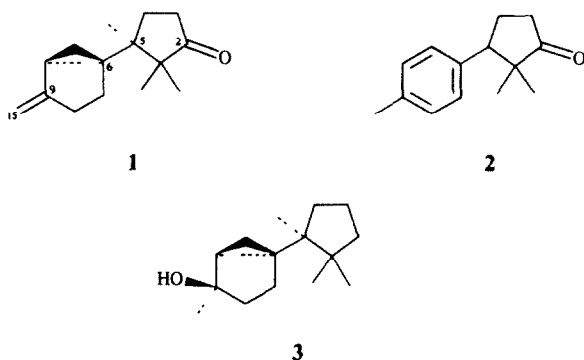


Fig 1 ORTEP diagram of grimaldone



EXPERIMENTAL

Isolation of grimaldone from M. fragrans collected (i) near Ulan-Bator and (ii) in North Mongolia (i) The moist plant material (~150 g) (Mongolian Peoples Republic, Ulan-Bator, Zajsan Valley, 3 km S of Ulan-Bator, on soil in the mountain steppe [9], leg. S Huneck, 26 July 1978, det. R. Grolle) was separated as well as possible from the adhering soil, minced in a beaker with an ultra-Turrax after the addition of a mixture of MeOH–Et₂O (4 l, 150 ml) and the slurry filtered. The extract was evapd to dryness, the green waxy residue (1.4 g) redissolved in C₆H₆, the insoluble fraction removed and the soln chromatographed on silica gel (60 g, 5% H₂O), after *n*-hexane (750 ml) *n*-hexane–Et₂O [49:1 (1 l)] eluted a yellow oil which crystallized after the addition of a few drops of MeOH. The crystals were filtered, sublimed *in vacuo* at 120–140° and recrystallized from *n*-pentane to yield grimaldone (1) (0.022 g, 0.014%), C₁₅H₂₂O (218), in prisms of mp 91–92° and *R_f* 0.58 [silica gel PF 254 + 366, C₆H₆–Et₂O (9:1), 10% H₂SO₄, 150° → violet spot] MS, *m/z* 218.165 (M⁺, 11%, calc. for C₁₅H₂₂O 218.167), 203.144 ([M

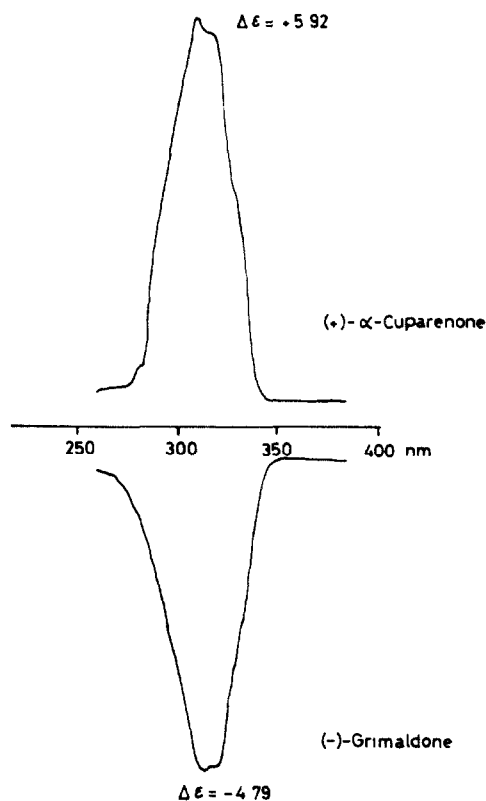


Fig 2 CD spectra of grimaldone and (+)-α-cuparenone

–Me]⁺, 3, calc. for C₁₄H₁₉O 203.143), 125.093

17, calc. for C₈H₁₃O 125.096),

97, 11), 94.065 ([α]_D²⁴ 100; calc. for

C₇H₁₀ 94.078), 83 (C₆H₁₁⁺, 100), IR ν_{max}^{KBr} cm⁻¹ 805, 866, 888, 915, 938, 958, 972, 984, 996, 1045, 1062, 1090, 1107, 1144, 1158, 1192, 1204, 1260, 1273, 1290, 1305, 1315, 1365, 1377, 1387, 1410, 1462, 1650, 1722, 2080, 3045, 3084.

	589	578	546	436	406	366
[α] _D ²⁴	-49.6	-55	-79	-206	-312	-643
	nm					

(CHCl₃, c 0.635), ORD (3.74 mg/3 ml MeOH) [φ]₄₀₀ -600, [φ]₃₁₂ -6200, [φ]₂₉₆ ± 0, [φ]₂₇₄ + 7600, [φ]₂₃₀ ± 0

(ii) The air-dried and ground liverwort (400 g) (Mongolian Peoples Republic, Chovsgol Aimak, Ulaan-uul Sum, on cliffs near the Tomyń-Brigade [10], leg. S Huneck, 17 July 1983, det. R. Grolle, voucher specimens of both samples in the herbarium of S.H.) was extracted with Et₂O, the solvent removed from the extract and the oily residue (3 g) chromatographed on silica gel (70 g, 5% H₂O), fractions 1(*n*-hexane, 750 ml) colourless oil (0.2 g, not further investigated), 2(*n*-hexane, 1 l) orange wax (0.07 g, not further investigated), 3(*n*-hexane–Et₂O, 24 l, 1:5 l) yellow oil (0.8 g), 4 (*n*-hexane–Et₂O, 4 l, 500 ml) greenish oil

and crystals (0.6 g). Fraction 3 was further separated by prep. TLC on plates with silica gel PF 254+366 [$20 \times 20 \times 0.1$ cm, *n*-hexane-Et₂O (4:1)]. After crystallization from *n*-pentane the band of *R_f* 0.39 yielded grimaldone (33 mg, 0.083%) of mp 86–88°. Fraction 4 was rechromatographed and gave a mixture of steroids (0.02 g, 0.005%) of mp 128–130°, GLC-MS (steel column, 180×0.2 cm, SE 30, 250°) showed the presence of brassicasterol, campesterol, stigmasterol and sitosterol.

Crystal data C₁₅H₂₂O, *M* = 218.3, orthorhombic, *P*2₁2₁2₁, *a* = 7.173(3), *b* = 8.833(1), *c* = 20.726(4) Å, *U* = 1313.0 Å³, *F*(000) = 480, *D_c* = 1.10 g/cm³, *Z* = 4, MoK_α radiation, *λ* = 0.71069 Å. 1055 independent observed reflections (*I* ≥ 3.0 σ_{*i*}) were collected on an Enraf-Nonius CAD-4 automatic diffractometer. The structure was solved by direct phasing techniques using MIT-HRIL [11]. Full matrix least-squares refinement of anisotropic parameters for non-hydrogen atoms and in the final two cycles the isotropic parameters of the hydrogen atom positions resulted in a final *R* of 0.047 and *wR* of 0.058 [*w* = 1/σ²(*F_o*)] [The atomic coordinates for this work are available on request from the Director of the Cambridge Crystallographic Data Centre, University Chemical Laboratories, Lensfield Road, Cambridge CB2 1EW].

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