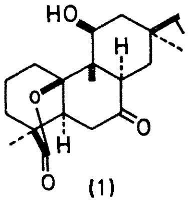
Structure of Rosein III: *X*-Ray Analysis Without a Heavy Atom

By R. Guttormson and P. Main*
(*Physics Department, University of York, Heslington, York* YO1 5DD)
and A. J. Allison and K. H. Overton
(*Chemistry Department, University of Glasgow, Glasgow, W.2*)

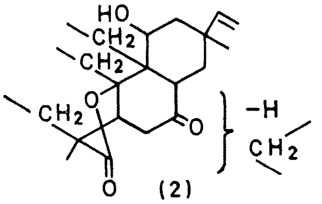
Summary The structure (**1**) of rosein III, indicated by chemical and spectroscopic evidence, has been confirmed by an *X*-ray analysis of the metabolite itself, using direct methods of phase determination.

Rosein III, a metabolite of *Trichothecium roseum*, was first described[1] by Freeman and Morrison in 1948. We have re-isolated this compound [m.p. 221°, $[\alpha]_D$ ($CHCl_3$) $-124°$] in minor amount, and on the basis of the limited evidence set out below were led to formulate it as 11-β-hydroxy rosenonolactone (**1**). More extensive investigations,[2,3] also terminated by lack of material, established the part structure (**2**), but further suggested[3] that this was incorporated in a rearranged rosane skeleton.

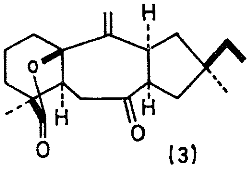
The mould's capricious refusal to produce rosein III dependably, turned our thoughts to *X*-ray analysis. Difficulties encountered in obtaining a suitable heavy-atom derivative in turn led to the structure analysis without a heavy atom detailed below.



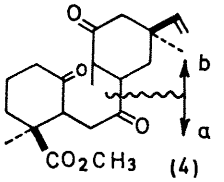
(1)



(2)



(3)



(4)

The i.r. and n.m.r. spectra of rosein III showed the close correspondence with rosenonolactone to be expected from formulation (**1**) [with the additional ν_{max} (CCl_4) 3610 cm^{-1}

and 1H, q (after D_2O-exchange) (J = 10·6 Hz) at τ 5·9]. The o.r.d. curves of the two compounds (MeOH) were, apart from amplitude, indistinguishable.

Dehydration ($POCl_3$–pyridine; 95°; 0·5 hr) of dihydrorosein III,[2] afforded the rearranged[4] (**3**), m.p. 193—194°, ν_{max} (CCl_4) 1715, 1719 cm^{-1}; 1H, s at τ 4·64 and 5·11. Rosein III diketone[2] with KOH–MeOH (5%; 20°; 10 min) gave the triketo-ester (**4**), m.p. 121° [ν_{max} (CCl_4) 1740, 1790 cm^{-1}; 3H, s, τ 6·34, 8·66, 8·87; 3H, d (J = 6 Hz) at τ 9·02. Mass spectrum: $C_{21}H_{30}O_5^+$ (M^+) 362·20984, $C_{11}H_{15}O_4^+$ (a) 211·09703 (%Σ_{130} 45; m^* 123), $C_9H_{11}O_2^+$ (a — H·COOMe) 151·07590 (%Σ_{130} 40; m^*108), $C_{10}H_{15}O^+$ (b) 151·11228 (%Σ_{130} 4; m^*63)].

The following X-ray analysis confirmed in every detail the suggested structure (**1**) for rosein III, except that it did not establish the absolute configuration.

Crystal data. $C_{20}H_{24}O_4$, M = 332. Orthorhombic, a = 14·63, b = 13·83, c = 8·77 Å, U = 1774 Å^3. Z = 4, D_c = 1·24. Space group $P2_12_12_1$ (No. 19). Intensities were measured, using an automatic diffractometer (with Mo-K_α-radiation), for 1436 independent reflexions, to θ = 23°, of which 874 were significantly above background.

The structure was solved by direct methods of phase determination using the tangent formula of Karle and Hauptman.[5] The starting point for phase determination was obtained as follows: four reflexions from principal zones, which had many Σ_2 contributors in the tangent formula, were given arbitrary phases to define the origin and enantiomorph; three other reflexions which, together with the first four, appeared to lead to good phase determination, were also chosen and, in the manner of Germain and Woolfson,[6] were given all combinations of phases in quadrants. This led to sixteen separate sets of phases for the seven reflexions. Each starting point was developed into a complete set of phases using a weighted tangent formula,

$$\tan \phi_h = \frac{\sum_{h'} W_{hh'} \left| E_h E_{h'} E_{h-h'} \right| \sin(\phi_{h'} + \phi_{h-h'})}{\sum_{h'} W_{hh'} \left| E_h E_{h'} E_{h-h'} \right| \cos(\phi_{h'} + \phi_{h-h'})} = \frac{T}{B}$$

where the weight $W_{hh'}$ is inversely proportional to the variance of $\phi_{h'} + \phi_{h-h'}$. The variance in the value of the phase ϕ_h is calculated from the equation (3·33) of Karle and Karle,[7] the value of α in the equation being given by $(T^2 + B^2)^{\frac{1}{2}}$.

A figure of merit for each set of phases was calculated as $\Sigma\alpha$ [$= \Sigma(T^2 + B^2)^{\frac{1}{2}}$] the sum being taken over all known phases. The E-map computed from the set of phases with the highest figure of merit revealed a partial structure of the molecule in which nine atoms could be placed with confidence. The remainder of the structure was obtained using the tangent formula recycling technique of Karle.[8] Refinement of the structure was carried out using block-diagonal least-squares and the present R factor is 11%.

(*Received, April* 1*st*, 1970; *Com.* 453.)

---

[1] G. G. Freeman and R. I. Morrison, *Biochem. J.*, 1948, **43**, XXIII; G. G. Freeman, R. I. Morrison, and S. E. Michael, *ibid.*, 1949, **45**, 191.

[2] J. Winter, Ph.D. Thesis, Manchester, 1961.

[3] A. R. Jones, Ph.D. Thesis, Manchester, 1966.

[4] J. D. Connolly, D. M. Gunn, R. McCrindle, R. D. H. Murray, and K. H. Overton, *J. Chem. Soc.* (*C*), 1967, 668.

[5] J. Karle and H. Hauptman, *Acta Cryst.*, 1956, **9**, 635.

[6] G. Germain and M. M. Woolfson, *Acta Cryst.*, 1968, B**24**, 91.

[7] J. Karle and I. L. Karle, *Acta Cryst.*, 1966, **21**, 849.

[8] J. Karle, *Acta Cryst.*, 1968, B**24**, 182.