Metabolites of Proteaceae. Part VI.[1] The Stereochemistry of Reflexin and Conocarpin

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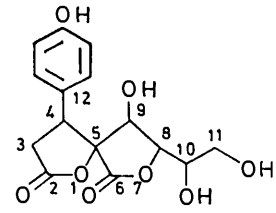
The stereochemistry of reflexin and conocarpin (from *Leucospermum* spp.) has been elucidated as 4*S*,5*S*,8*R*,9*R*,10*S*, whereas that of leucodrin (from *Leucadendron* spp.) is 4*R*,5*S*,8*R*,9*R*,10*S*. This is in accord with a mechanism of biogenesis suggested for these compounds, as well as with the greater reactivity of one lactone ring in the conocarpin series relative to that in the leucodrin series.

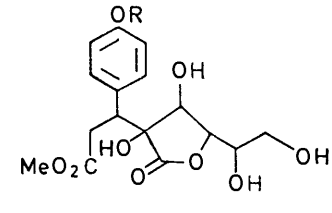
PREVIOUS reports have dealt with the isolation of conocarpin (I) from *Leucospermum conocarpodendron* [2] and of reflexin (II) from *L. reflexum*,[1] and with the elucidation of their structures. They are closely related to leucodrin, of known [3] absolute configuration (IV), and typically obtained [4] from *Leucadendron* spp.; conocarpin is indeed [2] a diastereoisomer of leucodrin. We now describe chemical results which demonstrate the stereochemistry of the title compounds.

The five chiral centres in these molecules are the carbon atoms 4, 5, 8, 9, and 10; these must furthermore, centre for centre, be the same in the two compounds, as reflexin (II) may readily be converted into conocarpin (I) by mild hydrolysis followed by closure of the two lactone rings, whereby the oxygen–carbon bonds 1,5 and 7,8 are not affected. The chemistry of each of the compounds therefore reflects their common stereochemistry.

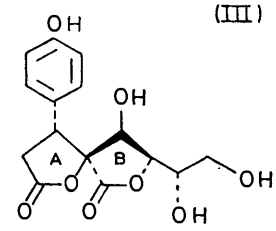
The configuration of C-4 in conocarpin has already been demonstrated [2] and is *S* in terms of specified [5] chirality. Reflexin methyl ether (III) (see Experimental section) yields the same (+)-*p*-methoxyphenylsuccinic acid (V) on oxidation in aqueous solution at 32° and pH > 5 with periodate; on carrying out a similar



(I)



(II) R = H
(III) R = Me

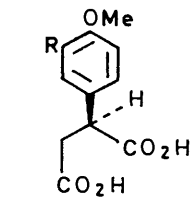


(IV)

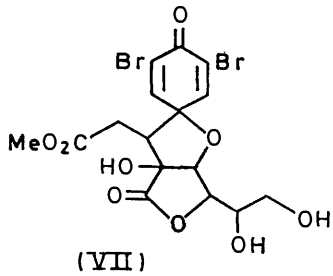
oxidation under acidic conditions, the corresponding (+)-(3-iodo-4-methoxyphenyl)succinic acid (VI) was

[1] Part V, G. W. Perold, A. J. Hodgkinson, and A. S. Howard, preceding paper.
[2] P. E. J. Kruger and G. W. Perold, *J. Chem. Soc.* (*C*), 1970, 2127.
[3] R. D. Diamand and D. Rogers, *Proc. Chem. Soc.*, 1964, 63.
[4] W. S. Rapson, *J. Chem. Soc.*, 1938, 282.
[5] R. S. Cahn, C. Ingold, and V. Prelog, *Angew. Chem. Internat. Edn.*, 1966, **5**, 385.

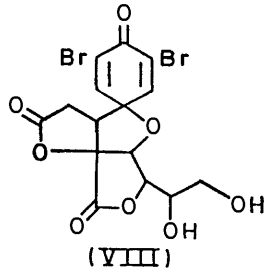
obtained, again as in the case [2] of conocarpin. The configuration of C-4 is here therefore also *S*, as in conocarpin.



(V) R = H
(VI) R = I



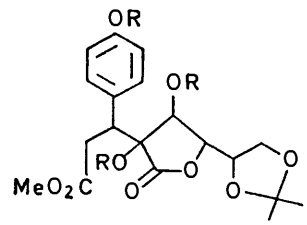
(VII)



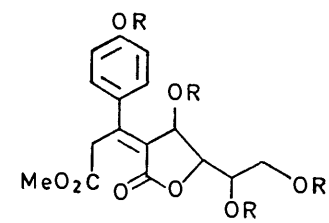
(VIII)

This finding allows of the determination of the configuration of C-5 and C-9 together, in the light of the ready formation of the dibromoquinolides (VII) and (VIII) from reflexin and conocarpin respectively (see previous paper) and the absence of any such reaction in the case of leucodrin. This reaction involves intramolecular nucleophilic attack by O-9 on the point of attachment of the aromatic ring (C-12). It does not occur in the case of leucodrin (IV), where the established[3] configuration of the molecule shows that the aromatic ring is held below the plane of lactone ring A by the 4*R* carbon atom: furthermore the internuclear distance (measured on Dreiding models with assumption of low degrees of puckering of the two lactone rings) between C-12 and O-9 in leucodrin is 3·8 Å. In conocarpin with 4*S*,5*S*,9*R* configuration, on the contrary, O-9 is held at 1·9 Å (nearly within bonding distance) from C-12, so that intramolecular nucleophilic attack by O-9 on C-12 must be particularly strongly favoured when the phenol–dienone conversion here concerned is made possible by reaction with bromine water. All other 5,9 configurations make direct intramolecular nucleophilic attack of O-9 on C-12 unlikely. Conocarpin (and reflexin) therefore can be assigned 4*S*,5*S*,9*R* configurations.

The configuration of C-8 has not yet proved amenable to a purely chemical approach. Many attempts were, for instance, made to prepare derivatives of conocarpin and reflexin in which only the hydroxy-groups in positions 5 and 9 would be available for glycol cleavage with periodate (or lead tetra-acetate). Reflexin 10,11-dimethyl acetal (IX) is clearly also such a substrate, but cleavage of the 5,9 bond of (IX) could not be attained in buffered aqueous solution or in pyridine without further degradation; this reflects the difficulty of cleaving a *trans*-diol of this kind. Treatment of the acetal (IX) with lead tetra-acetate in pyridine solution, followed by treatment of the crude product with N-sulphuric acid at 80° for 1 h, afforded only mixed phenolic products which did not react with dimedone. Attempts



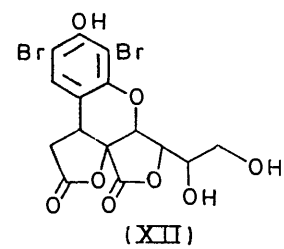
(IX) R = H
(X) R = Ac



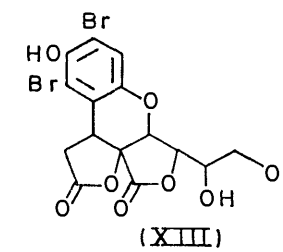
(XI) R = H, Ac, or Bz

to obtain partial hydrolysis of reflexin 10,11-dimethyl acetal triacetate (X) showed (by i.r. spectra) that the lactone group was hydrolysed at the same rate as the other acyl groups. Fruitless attempts were made to prepare a benzyl ether derivative of (IX), a pivalate ester derivative of reflexin dihydroxy-diamide, or a dihydroxy-diamide of dibromoreflexin quinolide. Attempted introduction of a 4,5-double bond by pyrolysis of reflexin penta-acetate or pentabenzoate, or by treatment of reflexin with phosphoryl chloride in pyridine solution, led to no satisfactory results: this may reflect some difficulty in introducing a double bond exocyclic to the γ-lactone ring in these instances to afford a structure such as (XI).

The dibromoquinolide of conocarpin (VIII), however, did prove useful in determining the stereochemistry at C-8. It underwent a dienone–phenol rearrangement in the presence of aluminium chloride to yield a chroman derivative (see Experimental section) which we formulate as (XII) or (XIII). The difference between the two possibilities is only one of positional isomerism with regard to the aromatic ring; this will not significantly affect the preferred conformation of the dihydropyran ring and hence is not of direct importance in the further discussion.



(XII)

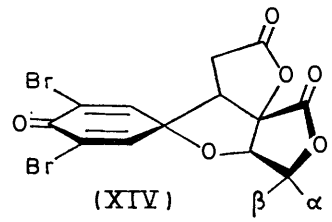


(XIII)

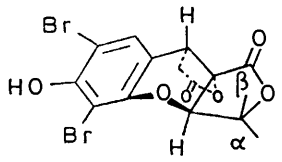
Conocarpin dibromoquinolide (VIII) possesses a rigid spiro-locked dilactone structure; thus relationships of the dienone system with regard to the remainder of the molecule may be assessed with some firmness. Calculation of anisotropic effects to be expected (according to Johnson and Bovey [6]) from the nearest carbon–carbon double bond show, however, that essentially no effect is to be expected for H-8 in either of the possible configurations of C-8: this is represented in perspective [see (XIV)].

[6] L. M. Jackman and S. Sternhell, 'Applications of Nuclear Magnetic Resonance Spectroscopy in Organic Chemistry,' Pergamon, Oxford, 1969, pp. 83—88.

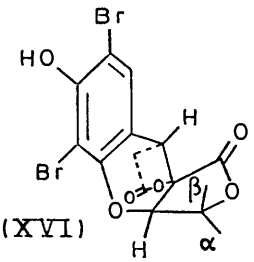
The chroman derivative [(XII) or (XIII)] does however allow more useful prediction. The two conformations available for the dihydropyran ring have marked differences in the spatial relation between the aromatic ring and C-8. In the one case [such as (XV)] the aromatic ring is held back from lactone ring B and roughly parallel to the mean plane of it: calculations [7] suggest that H-8 in either configuration of C-8 could experience only a small effect due to the aromatic ring. In the other case



(XIV)



(XV)



(XVI)

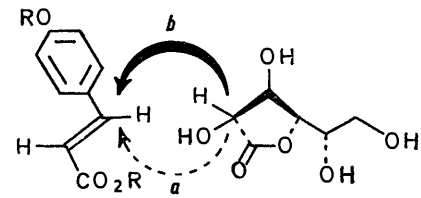
[such as (XVI)], however, the aromatic ring is held very definitely forward towards lactone ring B and roughly perpendicular to it. In this case the calculations [7] indicate strong shielding of H-8β [see (XVI)], *cis* to the dihydropyran ring, and at most weak shielding of H-8α, *trans* to the dihydropyran ring. Comparison of the δ values for conocarpin dibromoquinolide (VIII) and the corresponding chroman (see Table) shows that H-8 in the chroman compound [(XII) or (XIII)] is indeed shielded with respect to H-8 in the quinolide compound (VIII) to the extent of 0·4 p.p.m. This indicates a conformation such as (XVI) for the chroman derivative and the β-configuration for H-8, so that C-8 should be specified as *R*.

^{1}H N.m.r. signals of dibromoconocarpin quinolide hydrate (VIII) and the chroman [(XII) or (XIII)] in [$^2H_6$]-acetone [p.p.m. relative to tetramethylsilane; separations (*S*) Hz]

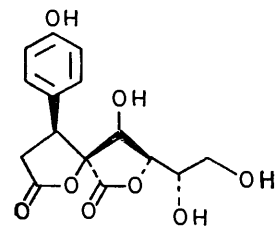
Assignment	Type	(VIII)	(XII) or (XIII)
Olefinic	AB	7·56, 7·64 (*S* 2·5)	
Aromatic	A		7·3
Lactone ring A	ABX	3·8, 3·0, 2·7 (*S* 9, 3, 19·5)	4·2, 3·4, 2·6 (*S* 10, 9, 18)
Lactone ring B	AB	5·4, 4·8 (*S* 2)	5·3, 4·4 (*S* 7)
Side-chain	A_2B	4·0, 3·6 (*S* 6)	3·9, 3·6 (*S* 6)
OH	A	4·7, 3·6	

The configuration at C-10 was unambiguously established by excision of the C-8,10,11 chain (as glyceraldehyde) by oxidation with periodate of the 10,11-dimethyl acetal (IX) [8] of reflexin, and of that of conocarpin (see Experimental section). The dimedone derivative obtained in both instances was that of L-glyceraldehyde; thus the configuration at C-10 is *S*.

It thus appears that leucodrin (4*R*,5*S*,8*R*,9*R*,10*S*) is related to the pair of compounds conocarpin and reflexin (both 4*S*,5*S*,8*R*,9*R*,10*S*), in a way which is in full agreement with the view [3] that the biogenesis of leucodrin may involve a Michael-type condensation of *p*-hydroxycinnamic acid and L-γ-galactonolactone * (or their equivalents). If leucodrin arises by attack of the lactone system (through the carbanion in the position α to its carbonyl group) from above [see arrow *b* in (XVII)] to give structure (IV), then the conocarpin series of compounds would arise by the alternative mode of attack [arrow *a* in (XVII)] to give conocarpin (XVIII) or its congeners.



(XVII)



(XVIII)

The stability of lactone ring A in leucodrin, as contrasted with the demonstrated lability of lactone A in the conocarpin series, is furthermore then easily understood in terms of the strong steric interaction between the aromatic ring and the 9-hydroxy-group in the latter series. This must cause strong stresses in lactone ring A in that case, whereas in the case of leucodrin no such interaction occurs.

EXPERIMENTAL

Experimental procedures and spectral techniques are described in the preceding paper. Furthermore, c.d. spectra were run for solutions in methanol on a Jasco ORD/UV-5 instrument with a c.d. attachment.

Oxidation of Reflexin Methyl Ether (III) *with Periodate.*—(a) *At pH* > 5. Reflexin methyl ether [1] (120 mg) in N-potassium hydroxide (3 ml) and water (25 ml) was treated with sodium periodate (950 mg) in water (10 ml) for 4 h at 32° while the pH was kept >5 by adding 0·1N-sodium hydroxide. Excess of oxidant was destroyed with sulphur dioxide gas and the solution was continuously extracted (18 h) with ether to yield a brown solid (80 mg), which

* The occurrence of L-galactose derivatives in these higher plants is striking. We postulate a 1,6-reduction–oxidation sequence, based on D-galactose as the starting point, which would afford this 'unnatural sugar' derivative, and we are developing an experiment to test this hypothesis.

[7] Ref. 6. pp. 94—98.

[8] G. W. Perold and H. K. L. Hundt, *Chem. Comm.*, 1970, 712.

crystallised from water to give (+)-*p*-methoxyphenylsuccinic acid (V), m.p. 201—202·5° (Found: C, 58·7; H, 5·4. Calc. for $C_{11}H_{12}O_5$: C, 58·9; H, 5·4%), $[\alpha]_D$ +127° (*c* 0·90); c.d. $\lambda_{max.}$ 228, 273, and 288 nm ($\Delta\varepsilon$ +8·4, +0·4, and 0), identical with the compound obtained [2] from conocarpin (m.p. and i.r. comparison).

(b) *Under acid conditions.* Reflexin methyl ether (175 mg) was kept in N-potassium hydroxide (2·5 ml) for 30 min, then treated with orthoperiodic acid (1·17 g) in water (10 ml) and 2N-sulphuric acid (2·5 ml) at 30° for 90 min. Excess of oxidant was removed by passing sulphur dioxide gas. Continuous extraction (20 h) with ether afforded a brown mixture (168 mg) showing a major acidic component at R_F 0·43 (benzene–acetic acid, 9 : 1 v/v). Crystals of (+)-(3-iodo-4-methoxyphenyl)succinic acid (VI) [2] were obtained directly from water (Found: C, 37·7; H, 3·3. Calc. for $C_{11}H_{11}IO_5$: C, 37·7; H, 3·2%), m.p. 199—200°, $[\alpha]_D$ +102° (*c* 1·01), as before.[2]

Graded Hydrolysis of Reflexin 10,11-*Dimethyl Acetal Triacetate* (X).—The title compound (104 mg) was kept in methanol (2 ml) and 0·1N-sodium hydroxide (2 ml, 1 mol. equiv.) for 30 min. The solution was then evaporated and an i.r. spectrum taken of the residue. This product was then treated in the same way another three times. The product from the first treatment showed lactone (1790 cm^{-1}) and ester (1730 cm^{-1}) absorptions, together with a small carboxylate (1560 cm^{-1}) peak; after the second and third treatments the 1790 and 1730 cm^{-1} peaks were much weaker, and the final product showed only the 1560 cm^{-1} absorption.

Dienone–Phenol Rearrangement of the Conocarpin Dibromoquinolide (VIII).—The conocarpin dibromoquinolide (VIII) [1] (m.p. 215—218°; 250 mg) was kept in nitrobenzene solution (10 ml) with aluminium chloride (3 g) for 72 h; the mixture was then poured into water (30 ml) and extracted with ethyl acetate (3 × 10 ml) to yield a syrup, dried *in vacuo*, showing a major component with R_F 0·65 in benzene–ethyl acetate–methanol (6 : 3 : 1 v/v). Chromatography in the same solvent afforded the chroman derivative (XII) or (XIII) (60 mg), m.p. 228·5—230° (from aqueous methanol and ethyl acetate) (Found: C, 36·2; H, 2·7. Calc. for $C_{15}H_{12}Br_2O_8,H_2O$: C, 36·2; H, 2·8%), $\nu_{max.}$ 3535, 3500, and 3280 (OH), and 1785 and 1775sh cm^{-1} (C=O), *m/e* 478, 480(64%), and 482 (M^+); 432, 434, and 436; 359, 361, and 363; 319, 321(100%), and 323; and 266, 268, and 270; n.m.r. data in the Table.

L-*Glyceraldehyde from Reflexin.*—Reflexin 10,11-dimethyl acetal [1] (250 mg) in N-sodium hydroxide (4 ml) was treated with sodium periodate (0·9 g) in water (15 ml) for 1 h. Lead subacetate (0 1 ml of 20% solution) was added, the mixture was filtered after 10 min, and lead was removed from the solution by adding N-sulphuric acid (8 ml) and filtering again. The acidic filtrate was kept at 50° for 1 h then treated with Dowex 1-X8 resin (10 g), filtered, and concentrated *in vacuo* at 40° to 10 ml. Potassium dihydrogen phosphate (1M; 0·5 ml) and N-sodium hydroxide (0·3 ml) were added together with dimedone (300 mg) in ethanol (3 ml), and the mixture was kept for 5 h at 40° and then for 16 h at room temperature. The dimedone derivative of L-glyceraldehyde [9] which separated was obtained pure (140 mg) from ethanol; m.p. 202—203° (Found: C, 68·4; H, 7·9. Calc. for $C_{19}H_{26}O_5$: C, 68·2; H, 7·8%), $[\alpha]_D$ −210° (*c* 1·08) (lit.,[9] m.p. 197—198·5°, $[\alpha]_D$ −210°; lit.,[8] $[\alpha]_D$ −208°).

L-*Glyceraldehyde from Conocarpin.*—Conocarpin 10,11-dimethyl acetal [1] (150 mg) was treated as in the preceding experiment and in the same way afforded the dimedone derivative of L-glyceraldehyde [9] (60 mg), m.p. 195—200° (from ethanol) (Found: C, 68·3; H, 7·9. Calc. for $C_{19}H_{26}O_5$: C, 68·2; H, 7·8%), $[\alpha]_D$ −208° (*c* 0·63).

In addition to the help acknowledged in the preceding paper,[1] we thank Dr. P. R. Enslin (C.S.I.R.) for obtaining c.d. spectra.

[2/463 *Received*, 28*th February*, 1972]

[9] A. S. Perlin and C. Brice, *Canad. J. Chem.*, 1956, **34**, 85.