438. *Alkaloids of* Daphnandra *Species. Part I. Repandine.*

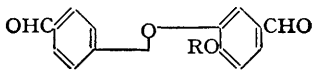
By I. R. C. Bick and A. R. Todd.

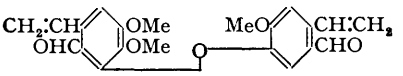
Repandine, $C_{37}H_{40}O_6N_2$, an alkaloid from *D. repandula*, is shown to have structure (III; R = H) or (IV; R = H) and to be an optical isomer of oxyacanthine. Bisbenzyl*iso*quinoline alkaloids have not hitherto been reported in plants of the family *Monimiaceæ*. Repandine is also shown to be identical with a base obtained in small amount by v. Bruchhausen and Schulze (*Arch. Pharm.*, 1929, **267**, 623) from a specimen of oxyacanthine and believed by them to be an artefact. It is considered that, in fact, traces of repandine occur together with oxyacanthine in at least one *Berberis* species.

Trees belonging to the genus *Daphnandra* (family *Monimiaceæ*) are native to Australia and occur in coastal regions of Queensland and northern New South Wales. The presence of alkaloids in the bark of *Daphnandra* species was first reported by Bancroft (*J. Proc. Roy. Soc. N.S.W.*, 1886, **20**, 69; *Proc. Roy. Soc. Queensland*, 1887, **4**, 13; *Austr. J. Pharm.*, 1887, **2**, 103), and much later Pyman (*J.*, 1914, **105**, 1679) isolated three alkaloids daphnandrine, $C_{36}H_{38}O_6N_2$, daphnoline, $C_{35}H_{36}O_6N_2$ or $C_{34}H_{34}O_6N_2$, and micranthine, $C_{36}H_{32}O_6N_2$, from *D. micrantha*. Beyond characterisation, analysis and a description of certain colour reactions, no chemical investigation of these substances appears to have been made, although Pyman (*loc. cit.*) records a pharmacological examination by Dale which showed them to have relatively low toxicity. Recently, the remaining three species have been examined by Bick and Whalley (Univ. of Queensland Papers, Dept. of Chemistry, 1946, **1**, No. 28; 1947, **1**, No. 30; 1948, **1**, No. 32, in the press). These workers isolated from *D. repandula* two new alkaloids, repanduline, $C_{40}H_{46}O_8N_2$, and repandine, $C_{38}H_{42}O_6N_2$, from *D. Dielsii* repanduline and from *D. aromatica* daphnoline, and a third new alkaloid of formula $C_{36}H_{38}O_6N_2$ which they named aromoline. Some years ago one of us (A. R. T.) in collaboration with Dr. E. S. Ewen commenced a study of

the alkaloids of *D. micrantha* using the material isolated by the late Dr. Pyman and kindly supplied by him. This study was left unfinished as a result of war circumstances, but material for a more comprehensive investigation being now available the present authors have taken it up again and extended it to cover in addition the new alkaloids isolated by Bick and Whalley (*loc. cit.*). The earlier study above mentioned had revealed that daphnandrine and daphnoline were almost certainly members of the bisbenzyl*iso*quinoline group; from the published molecular formulæ it seemed very probable that, apart perhaps from repanduline, the other *Daphnandra* bases were also members of this group, and this determined from the outset the degradative methods applied to repandine, whose structure forms the subject of the present memoir. The results of investigations on daphnandrine and the other alkaloids from *Daphnandra* species will be reported later.

As a result of further analyses, the molecular formula of repandine given by Bick and Whalley (*loc. cit.*) has been modified to $C_{37}H_{40}O_6N_2$. The presence of three methoxy- and two methylimino-groups was confirmed, and it was found that repandine has weak phenolic properties. Methylation of repandine with methyl iodide in methanolic sodium methoxide yielded O-*methylrepandine dimethiodide*; Hofmann degradation of this product gave *O*-methylrepandinemethine, isolated as its dimethiodide. When ozonised, the methine base, which was optically inactive, yielded 2-methoxydiphenyl ether-5 : 4′-dialdehyde (I; R = Me) and an amino-aldehyde, which

(I.) 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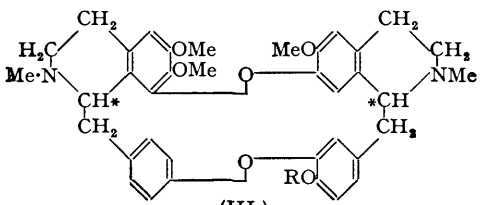
was converted into its methiodide in the usual manner and submitted to a further Hofmann degradation. The product was a crystalline aldehyde, identified as 2 : 3 : 2′-trimethoxy-5 : 4′-divinyldiphenyl ether-6 : 5′-dialdehyde (II) by direct comparison with an authentic specimen prepared from oxyacanthine by an analogous series of reactions (v. Bruchhausen and Gericke, *Arch. Pharm.*, **1931**, **269**, 115). The isolation of the two aldehydes above mentioned shows that *O*-methylrepandine belongs to the bisbenzyl*iso*quinoline group of bases and must have structure (III; R = Me) or (IV; R = Me), whilst *O*-methylrepandinemethine dimethiodide must be correspondingly represented by (V) or (VI).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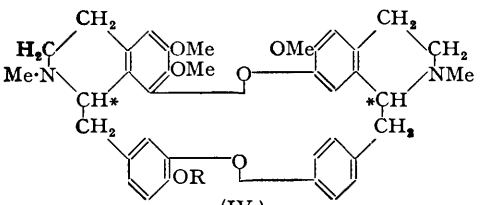
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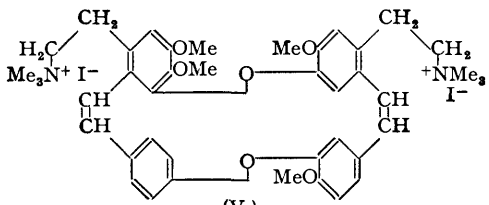
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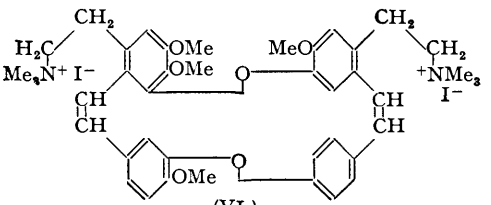
(VI.)

Repandine is isomeric with oxyacanthine and with berbamine, both alkaloids isolated from various *Berberidaceæ*. Of formulæ (III; R = Me) and (IV; R = Me), one also represents *O*-methyloxyacanthine and the other *O*-methylberbamine (v. Bruchhausen and Gericke, *loc. cit.*).

In this series, mixed melting points are not wholly satisfactory as a means of establishing the identity of compounds derived from different sources and recourse was had to the method of *X*-ray powder photography. It was found that the powder photographs of *O*-methylrepandinemethine dimethiodide (V or VI) and *O*-methyloxyacanthinemethine dimethiodide (V or VI; v. Bruchhausen and Gericke, *loc. cit.*) were identical. It follows therefore that repandine corresponds to oxyacanthine rather than to berbamine in its structure. Examination of *X*-ray powder photographs and optical rotations, however, showed that *O*-methylrepandine dimeth-

iodide was not identical with *O*-methyloxyacanthine dimethiodide; clearly *O*-methylrepandine (III; R = Me or IV; R = Me) must differ from *O*-methyloxyacanthine in the stereochemical arrangement about one or other of the asymmetric centres marked with asterisks.

In order to determine the location of the phenolic hydroxyl in repandine, the alkaloid, which could not be satisfactorily ethylated with diazoethane, was converted into its resinous dimethiodide and the latter ethylated to give O-*ethylrepandine dimethiodide*. When this product was submitted to Hofmann degradation, and the resulting methine base ozonised, an aldehyde was isolated which gave the expected analytical values for *2-ethoxydiphenyl ether-5 : 4′-dialdehyde* (I; R = Et) and when oxidised gave an acid corresponding in its properties to 2-ethoxydiphenyl ether-5 : 4′-dicarboxylic acid previously obtained by Späth and Pikl (*Ber.*, 1929, **62**, 2251) in similar fashion from *O*-ethyloxyacanthine.

It follows, therefore, that repandine has structure (III; R = H) or (IV; R = H) and is an optical isomer of oxyacanthine. Four isomers are possible for each of the structures (III) and (IV), and if we assume that their optical rotation is due to the summation of two components A and B corresponding to each of the two asymmetric centres, then of the four possibilities A + B, A − B, −A + B, and −A − B (where A is assumed to be greater than B), the first evidently represents oxyacanthine ($[\alpha]_D$ = + 279°) and the third repandine ($[\alpha]_D^{15°}$ = − 106°). On this basis A would be *ca.* 190° and B *ca.* 85° in the oxyacanthine group. It is interesting to note that, of the four isomers possible in the berbamine series, berbamine itself is the only one known to occur naturally; of the corresponding methyl ethers, however, three occur (Kondo and Keimatsu, *Ber.*, 1935, **68**, 1503), *viz.*, phaeanthine ($[\alpha]_D^{30°}$ = − 278°), its enantiomorph tetrandrine ($[\alpha]_D^{24°}$ = + 263°) and *iso*tetrandrine (*O*-methylberbamine) ($[\alpha]_D^{17°}$ = + 146°). The fact that repandine is a bisbenzyl*iso*quinoline alkaloid is of some botanical interest since no alkaloid of this group has hitherto been reported either in the *Monimiaceæ* or in the related *Lauraceæ*.

The properties of repandine agree closely with those recorded for a base, m. p. 255° ($[\alpha]_D^{20°}$ = − 95°), isolated from a sample of oxyacanthine by v. Bruchhausen and Schulze (*loc. cit.*) during an attempt to prepare a monohydrochloride of the latter substance; the new base was considered by these workers to be an artefact produced from oxyacanthine during treatment with hydrogen chloride. Through the kindness of Prof. v. Bruchhausen we have been able to establish the identity of repandine with this base by direct comparison; the two materials showed no m. p. depression on mixing and their Debye–Scherrer diagrams were identical. In view of the relationship between repandine and oxyacanthine established in the present investigation, we consider it very improbable that it could be produced from oxyacanthine by treatment with hydrogen chloride, and indeed we could obtain no evidence of its formation from purified oxyacanthine in these circumstances. We therefore conclude that it did not arise as an artefact in the experiments of v. Bruchhausen and Schulze (*loc. cit.*) and that repandine actually occurs in small amount along with oxyacanthine in at least one species of *Berberis*.

Experimental.

Repandine.—A specimen of the alkaloid isolated from *D. repandula* (Bick and Whalley, *loc. cit.*) was purified by repeated recrystallisation from acetone and ethanol and formed colourless needles, m. p. 255°, $[\alpha]_D^{15°}$ −106° (*c*, 1·2 in chloroform). It gave no coloration with ferric chloride but gave a pink colour with Millon's reagent. On addition of excess of alkali to a solution of the base in dilute hydrochloric acid (5%) the precipitate first formed redissolved to a clear solution; on passing carbon dioxide through this solution a white precipitate was produced which after recrystallisation from ethanol had m. p. 255°, undepressed by the original repandine (Found : C, 72·9; H, 6·9; N, 4·7; MeO, 15·2; MeN, 9·3. Calc. for $C_{37}H_{40}O_6N_2$: C, 73·0; H, 6·6; N, 4·6; 3MeO, 15·3; 2MeN, 9·5%).

O-*Methylrepandine Dimethiodide.*—Repandine (5 g.) was dissolved in dry methanol (200 c.c.) and methyl iodide (7·5 c.c.) was added, followed by methanolic sodium methoxide (0·32 g. of sodium in 5 c.c. of methanol). The mixture was refluxed for 6 hours, a similar amount of sodium methoxide added, and heating continued for a further 6 hours; this process was repeated until in all five such additions of sodium methoxide had been made. The solution was then evaporated under reduced pressure, and the residue taken up in hot water. The aqueous solution on cooling deposited an amorphous solid which was collected and redissolved in hot water, and the solution was boiled with a little copper powder during ten minutes, filtered, and set aside. The *dimethiodide* slowly crystallised as colourless needles (6·1 g.) which decomposed at 255—260° without melting, $[\alpha]_D^{15°}$ −95° (*c*, 0·3 in 50% aqueous ethanol) (Found : C, 48·4; H, 5·8; N, 2·6; I, 26·0; loss at 110°/0·1 mm., 9·3. $C_{40}H_{48}O_6N_2I_2,5H_2O$ requires C, 48·2; H, 5·9; N, 2·8; I, 25·5; $5H_2O$, 9·0%). *O*-Methyloxyacanthine dimethiodide had $[\alpha]_D^{14°}$ = +40° (*c*, 0·4 in 50% aqueous ethanol) (Gadamer and v. Bruchhausen, *Arch. Pharm.*, 1926, **264**, 193, give $[\alpha]_D$ = + 42°).

O-*Methylrepandinemethine Dimethiodide.*—*O*-Methylrepandine dimethiodide (5 g.) was dissolved in water (1 l.) and shaken with freshly prepared silver oxide (from 5 g. of silver nitrate). After 1 hour the solution, which was free from iodide ions, was filtered and concentrated to 100 c.c. under reduced pressure. Aqueous potassium hydroxide (50 c.c. of 50%) was now added, and the mixture heated on

the steam-bath for 2 hours during which time a yellowish resin slowly separated. The cooled mixture was extracted with chloroform, and the combined extracts were washed with water and dried (Na_2SO_4) On evaporation of the chloroform solution the methine base was obtained as a yellowish resin (2·5 g.) which could not be crystallised. For characterisation a portion of the resin (1 g.) was dissolved in methanol (50 c.c.) and refluxed with methyl iodide (1 c.c.) during 15 minutes. On concentrating the resulting solution to small bulk *O*-methylrepandinemethine dimethiodide separated and was recrystallised from methanol. Colourless needles, m. p. 255—260° (decomp.), identical (Debye–Scherrer diagram) with *O*-methyloxyacanthinemethine dimethiodide (m. p. 255—260°, decomp.) (Found : C, 52·2; H, 6·1; N, 3·1; loss at 110°/0·1 mm., 4·6. Calc. for $C_{42}H_{52}O_6N_2I_2,2H_2O$: C, 52·0; H, 5·8; N, 2·9; $2H_2O$, 3·9%).

Ozonolysis of O-*Methylrepandinemethine.*—The above resinous methine (3·1 g.) was dissolved in dilute sulphuric acid (20 c.c. of 5%), and a stream of ozone passed through the ice-cold filtered solution for 30 minutes. The yellowish resin which separated was extracted with ether; further passage of ozone through the aqueous solution left after extraction yielded only a small additional quantity of product, which was likewise extracted with ether. The combined ether extracts were washed, dried (Na_2SO_4), and evaporated. Recrystallisation of the residue from light petroleum (b. p. 40—60°) gave colourless prismatic needles (0·9 g.), m. p. 77—78° (Found : C, 70·2; H, 4·7; MeO, 11·4. Calc. for $C_{15}H_{12}O_4$: C, 70·3; H, 4·7; MeO, 12·1%). A mixed m. p. with 2-methoxydiphenyl ether-5 : 4′-dialdehyde (m. p. 77—78°) prepared by a similar series of reactions from oxyacanthine showed no depression.

The acid aqueous solution obtained after ozonolysis and extraction with ether was shaken for 15 minutes with palladised charcoal (0·1 g.) to remove peroxides, then shaken with hydrogen for a further 15 minutes and made alkaline with sodium hydroxide. The precipitated amino-aldehyde was extracted with ether, and the extract dried and evaporated. The residue was dissolved in methanol (10 c.c.), methyl iodide (2 c.c.) added, and the solution refluxed for 15 minutes. On concentration of the solution the amino-aldehyde dimethiodide separated as colourless needles (2·8 g.) and was recrystallised from aqueous methanol. On heating it melted with decomposition between 230° and 240° (Found : C, 43·0; H, 5·8; N, 3·8; loss at 110°/0·1 mm., 3·0. Calc. for $C_{27}H_{38}O_6N_2I_2,H_2O$: C, 42·6; H, 5·6; N, 3·7; H_2O, 2·4%).

2 : 3 : 2′-*Trimethoxy*-5 : 4′-*divinyldiphenyl Ether*-6 : 5′-*dialdehyde.*—The above amino-aldehyde dimethiodide (2·5 g.) was dissolved in water (100 c.c.) and shaken for 3 hours with freshly prepared silver oxide (from 2 g. of silver nitrate). The mixture was filtered, the filtrate concentrated to small bulk, and aqueous potassium hydroxide (20 c.c. of 50%) added. The alkaline solution was heated on the steam-bath until no more trimethylamine was evolved. The yellowish resin which separated was extracted with chloroform, and the extract washed, dried, and evaporated to small bulk. On standing, 2 : 3 : 2′-trimethoxy-5 : 4′-divinyldiphenyl ether-6 : 5′-dialdehyde separated in almost colourless diamond-shaped crystals (0·9 g.). Recrystallised from chloroform it had m. p. 139°, undepressed in admixture with an authentic specimen (m. p. 139°) prepared from oxyacanthine. On catalytic hydrogenation the substance took up 2 mols. of hydrogen rapidly and a further 2 mols. more slowly (Found : C, 68·6; H, 5·4; MeO, 24·5. Calc. for $C_{21}H_{20}O_6$: C, 68·5; H, 5·5; 3MeO, 25·3%).

O-*Ethylrepandine Dimethiodide.*—Repandine (2 g.) was dissolved in hot methanol (100 c.c.) and refluxed with methyl iodide (2 c.c.) for 15 minutes. Removal of solvents under reduced pressure gave the dimethiodide as a yellowish resin which could not be crystallised. The resin was dissolved in hot ethanol (200 c.c.), and ethyl iodide (5 c.c.) added followed by ethanolic sodium ethoxide (0·12 g. of sodium in 4 c.c. of ethanol). The mixture was refluxed for 6 hours, when a further similar amount of sodium ethoxide was added and heating continued. The process was repeated until 5 such quantities of sodium ethoxide had been added. The resulting solution was evaporated under reduced pressure, and the residue dissolved in hot water. A brownish resin separated on cooling; it was redissolved in hot water, and the solution boiled with copper powder for 10 minutes, filtered, and cooled. O-*Ethylrepandine dimethiodide* separated as a white amorphous mass (2 g.). Purified by being allowed to separate from hot water, it melted with decomposition between 235° and 245° (Found : C, 51·3; H, 5·7; N, 3·0; loss at 110°/0·1 mm., 5·1. $C_{41}H_{50}O_6N_2I_2,2\frac{1}{2}H_2O$ requires C, 51·0; H, 5·7; N, 3·0; H_2O, 4·7%).

2-*Ethoxydiphenyl Ether*-5 : 4′-*dialdehyde.*—*O*-Ethylrepandine dimethiodide (1·5 g.) was submitted to Hofmann degradation in the manner described above for the corresponding *O*-methyl compound. The crude methine base was dissolved in ice-cold dilute sulphuric acid and ozonised during 30 minutes. The resin which separated was taken up in ether, and the extract washed, dried (Na_2SO_4), and evaporated. The residue crystallised from ether–light petroleum as colourless prismatic needles, m. p. 59—60° (Found : C, 70·8; H, 5·4; EtO, 16·3. $C_{16}H_{14}O_4$ requires C, 71·1; H, 5·2; EtO, 16·7%). Admixture with 2-methoxydiphenyl ether-5 : 4′-dialdehyde (above) gave a depression in m. p. of *ca.* 10°.

The *dialdehyde*, oxidised with potassium permanganate in acetone solution, gave an acid crystallising from glacial acetic acid in needles, m. p. 287° (Found : C, 63·2; H, 5·2. Calc. for $C_{16}H_{14}O_6$: C, 63·6; H, 4·7%). Späth and Pikl (*loc. cit.*) record m. p. 288·5—289·5° for 2-ethoxydiphenyl ether-5 : 4′-dicarboxylic acid.

We wish to express our gratitude to Prof. F. von Bruchhausen and Prof. F. von Wessely for their gifts of samples of oxyacanthine and certain of its degradation products, and to the British Council for a scholarship held by one of us (I. R. C. B.). We are also deeply indebted to Dr. P. J. G. de Vos who carried out the *X*-ray crystallographic examinations.

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