SIMPLE INDOLE BASES OF *DESMODIUM GANGETICUM* (LEGUMINOSAE)

By P. K. Banerjee* and S. Ghosal*

[Manuscript received August 16, 1968]

Previously, we reported[1–3] the occurrence of seven indole-3-alkylamine bases in the above-ground portions of *Desmodium pulchellum* Benth. ex Baker. We also appraised the significance of the co-presence of these and related bases, in several plant species, in the light of tryptophan metabolism in plants. To extend the knowledge of content and distribution of simple indole bases in the genus *Desmodium*, we have now investigated the alkaloidal constituents of *Desmodium gangeticum* DC. The present communication is an account of the isolation and characterization of four known indole-3-alkylamine bases, viz. 5-methoxy-*N*,*N*-dimethyltryptamine,[3] *N*,*N*-dimethyltryptamine,[3] and their *N*-oxides,[3] and two β-carboline alkaloids, viz. N_b-methyltetrahydroharman[4] and 6-methoxy-2-methyl-β-carbolinium cation, from the above-ground portions of *D. gangeticum*. Incidentally, this is the first report of isolation of the last-named alkaloid from a plant source.

The quaternary nature of the β-carboline alkaloid was revealed from its isolation as reineckate salt from alkaline solution, its high pK_a value ($\simeq 10{\cdot}8$), and its u.v. absorption spectra in neutral ethanol and ethanolic alkali (λ_{max} (EtOH) 240, 250, 272–275sh, 290, and 342 mμ; a bathochromic shift of the major bands by about 10 mμ was observed in 0·05N ethanolic alkali). The u.v. absorption spectra of the alkaloid indicated its similarity with N_b-methyl-β-carbolinium cation.[5] Chemical proof for its structure was gained from its reduction with sodium and ethanol to 1,2,3,4-tetrahydro-6-methoxy-2-methyl-β-carboline.[3]

Although tetrahydroharman and N_b-methyltetrahydroharman bases are known[4] to occur in the Leguminosae, a quaternary β-carbolinium alkaloid of the type mentioned here appears to have been isolated only from the Loganiaceae.[6]

D. gangeticum DC. which grows abundantly in India has been known as a drug in the Ayurvedic system of medicine.[7] Its roots are used as astringent, in diarrhoea, in chronic fever, biliousness, snake-bite, and poisoning.

Assays in this laboratory indicate that air-dried samples of this plant normally contain 0·01–0·03% total alkaloid, but that fresh material contains more than three times the amount present in dry and preserved samples. In addition, fresh

* Department of Pharmaceutics, Banaras Hindu University, Varanasi-5, India.

[1] Ghosal, S., and Mukherjee, B., *Chemy Ind.*, 1964, 1800.
[2] Ghosal, S., and Mukherjee, B., *Chemy Ind.*, 1965, 793.
[3] Ghosal, S., and Mukherjee, B., *J. org. Chem.*, 1966, **31**, 2284.
[4] Johns, S. R., Lamberton, J. A., and Sioumis, A. A., *Aust. J. Chem.*, 1966, **19**, 1539.
[5] Bickel, H., Schmid, H., and Karrer, P., *Helv. chim. Acta.*, 1955, **38**, 649.
[6] Bächli, E., Vamvacas, C., and Schmid, H., *Helv. chim. Acta*, 1957, **40**, 1167.
[7] Chopra, R. N., Nayar, S. L., and Chopra, I. C., "Glossary of Indian Medicinal Plants." p. 94. (C.S.I.R.: New Delhi 1956.)

plant materials contain a large number of un-characterized indole and 5-oxyindole (positive test with α-nitroso-β-naphthol reagent) bases of low R_F values. However, the dry plant materials contain a higher proportion of 5-methoxy-*N*,*N*-dimethyltryptamine.

Experimental

The general procedure for the separation and identification of the individual alkaloids involved column chromatographic resolution over Brockmann neutral alumina; paper chromatography of the different fractions eluted out of the alumina column,' in presence of marker compounds; determination of u.v. and i.r. absorption spectra of pure components; and preparation of picrate where possible. Descending paper chromatography was used on Whatman No. 1 paper and three solvent systems, viz., n-butyl acetate–n-butanol–acetic acid–water (85 : 15 : 40 : 22, solvent 1), n-butanol–acetic acid–water (4 : 1 : 2, solvent 2), and ethanol–ammonia (4 : 1, solvent 3) were taken.

Chloroform-soluble Strong Bases

Green plant materials (1 kg, wet wt.) were macerated in a Waring blender with a mixture of chloroform (3 l.) and ammonia (15N, 50 ml) and kept at room temperature, with occasional shaking, for 1 week. The extract was filtered and the two phases separated. The chloroform layer was extracted with aqueous acetic acid (2N, 100 ml) and the organic layer was preserved for further treatment of the chloroform-soluble acetates. The aqueous acidic extract was cooled in ice, basified with ammonia (pH 9), and the liberated bases were extracted with chloroform. Removal of solvent from the chloroform extract at reduced pressure afforded a crude basic gum (2·5 g) which showed several Ehrlich- and Dragendorff-positive spots on paper chromatograms. The following two indole-3-alkylamines and their N_b-oxides were isolated from the crude mixture of bases by chromatographic resolution on Brockmann neutral alumina.

5-Methoxy-N,N-dimethyltryptamine.—5-Methoxy-*N*,*N*-dimethyltryptamine (0·57 g) was eluted out of the column with light petroleum–benzene (50 : 50) and was crystallized from the same solvent as colourless plates, m.p. and mixed m.p. 69°, R_F 0·51, 0·72, and 0·90 (solvents 1–3, respectively); pale yellow fluorescence under u.v. light on paper; λ_{max} 224, 277, and 294 mμ (log ε 4·45, 3·82, and 3·77, respectively); orange picrate from ethanol, m.p. 168°. The mixture of the picrate with an authentic sample,[3] m.p. 172°, had m.p. 168°.

N,N-Dimethyltryptamine.—Chloroform eluates from the column gave *N*,*N*-dimethyltryptamine as colourless thick oil, R_F 0·53, 0·74, and 0·93; co-chromatography with the marker compound[3] showed single spots having the same R_F values (solvents 1–3, respectively); no appreciable fluorescence under u.v. light; λ_{max} 222–224, 274, and 294 mμ; yellow picrate from ethanol, m.p. and mixed m.p. 168°.

N,N-Dimethyltryptamine N_b*-oxide.*—This oxide migrated out of the column with chloroform–methanol (9 : 1) as a hygroscopic solid (0·21 g); R_F 0·82 and 0·63; co-chromatography with the marker[3] showed single spots having same R_F values (solvents 2 and 3, respectively); yellow picrate from ethanol, m.p. 178–180°; the mixed m.p. with an authentic picrate[3] of m.p. 178–180° remained undepressed.

5-Methoxy-N,N-dimethyltryptamine N_b*-oxide.*—Methanol eluates from the column afforded a pale brown gum (0·18 g); R_F 0·89 and 0·59; co-chromatography with the marker[3] showed single spots having same R_F values (solvents 2 and 3, respectively); dull red colour under u.v. light; red picrate from ethanol, m.p. and mixed m.p. 158–159°.

Chloroform-soluble Acetates

The crude mixture of bases (three spots on paper chromatograms) obtained from the chloroform-soluble acetates by removal of the solvent at reduced pressure was chromatographed on Brockmann alumina; N_b-methyltetrahydroharman (0·03 g), *N*,*N*-dimethyltryptamine (0·41 g), and *N*,*N*-dimethyltryptamine N_b-oxide (0·12 g) were obtained as follows.

N_b*-Methyltetrahydroharman.*—Chloroform eluates from the alumina column afforded almost crystalline N_b-methyltetrahydroharman, m.p. and mixed m.p. with an authentic synthetic

sample 106–108° (lit.[4] 109–110°); R_F 0·58, co-chromatography with the synthetic sample showed the same R_F value (solvent 1); λ_{max} 226, 277, 292 mμ; λ_{max} (KBr) 2·95 (NH) and 3·55 μ (N_b-Me), superimposable i.r. spectrum with the authentic N_b-methyltetrahydroharman; yellow picrate from ethanol, m.p. 183–184° (lit.[4] 182–184°) (Found: N, 16·5. Calc. for $C_{13}H_{16}N_2,C_6H_3N_3O_7$: N, 16·3%).

N,*N*-Dimethyltryptamine and its N_b-oxide migrated out of the column with chloroform–methanol and were identified as described above.

Water-soluble Bases

An aliquot of the aqueous mother liquor, from the separation of the aforementioned indole-3-alkylamines, was acidified with 4N sulphuric acid and was then treated with a saturated aqueous solution of ammonium reineckate. The light pink reineckate complex had m.p. 155–157° (dec.). The precipitation of the reineckate salt was also carried out with the aqueous alkaline solution which afforded the same complex salt. 6-Methoxy-2-methyl-β-carbolinium cation was regenerated from the reineckate salt and was identified in the following way.

6-Methoxy-2-methyl-β-carbolinium cation.—The reineckate salt (0·21 g) was taken up in acetone and was passed through a column of De-acidite FF (pH 8) using ethanol as the eluent. Fractions of 5 ml were collected. The first three fractions on evaporation gave a light brown amorphous material (0·07 g); R_F 0·41 (as the major area of intensity with the Dragendorff's spray) plus three other faint spots at 0·33, 0·51, and 0·92 (solvent 2). The major component, purified by t.l.c., was subjected to alkaloidal colour reactions. It did not give any colour with Ehrlich reagent, gave a dull violet colour with ceric ammonium sulphate, and a pink colour with Dragendorff reagent. It showed a blue-violet fluorescence under u.v. light on papers. The amorphous material (0·04 g) was reduced with sodium (0·22 g) and absolute alcohol (10 ml). The reduced product showed an intense blue colour with Hopkin–Cole glyoxalic reagent. It was converted into the picrate, m.p. 183–185°; mixed m.p. with the authentic picrate[3] of 1,2,3,4-tetrahydro-6-methoxy-2-methyl-β-carboline, m.p. 188–191°, remained undepressed (Found: N, 15·7. Calc. for $C_{13}H_{16}N_2O,C_6H_3N_3O_7$: N, 15·7%).

Acknowledgments

The authors are grateful to Dr J. N. Chatterjea, Professor and Head of the Department of Chemistry, Patna University, Patna, for the i.r. spectra and to the C.S.I.R., New Delhi, for awarding a Junior Research Fellowship to one of them (P.K.B.).