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Chemical Examination of the Roots of *Cissampelos pareira* Linn.: Part III — Structure & Stereochemistry of Hayatinin*

A. K. BHATNAGAR & S. P. POPLI
Central Drug Research Institute, Lucknow

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Sodium and liquid ammonia reduction of hayatinin ethyl ether affords a non-phenolic component, identified as *dl*-4′-O-methyl-7-O-ethyl-N-methylcoclaurine, and a phenolic component identified as *dl*-N-methylcoclaurine. The structures of these two fragments have been confirmed by nuclear magnetic resonance and mass spectral data as well as the synthesis of the former by usual methods. This establishes the structure of hayatinin as 4″-O-methyl-bebeerine proposed earlier by Bhattacharji *et al.* [*J. scient. ind. Res.*, 21B (1962), 428]. The optical inactivity of hayatinin as well as of phenolic and non-phenolic fragments suggests that hayatinin isolated from this plant is a racemic mixture.

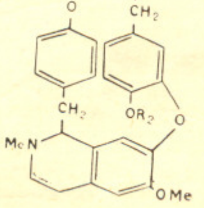
IN earlier studies on the alkaloids of *Cissampelos pareira* Linn. (Menispermaceae), the isolation of hayatin, hayatinin, hayatidin, cycleanine, *l*-bebeerine (curine) and an orange base was reported[1,2] from this laboratory. Kupchan and coworkers[3] confirmed the presence of curine and hayatin in this plant and also obtained *d*-isochondrodendrine from it. Recently, three water-soluble quaternary bases[4] and a bis-base containing a dihydroisoquinoline nucleus[6] have also been obtained from the same plant.

Of these alkaloids, curine, isochondrodendrine and cycleanine have been shown to have the L-(— —)-configuration. The structure of hayatinin[5] as 4″-O-methylbebeerine (I; $R=R_2=Me$; $R_1=H$) and that of hayatin[3] as chondrocurine (I; $R=R_2=H$; $R_1=Me$) was proposed by Bhattacharji *et al.* mainly on the basis of Hofmann degradation work. The stereochemistry at the two asymmetric centres in these two compounds, however, was not determined.

In the present study, sodium and liquid ammonia fission of hayatinin ethyl ether was undertaken to confirm, from the fragments obtained, the proposed structure of hayatinin as well as to determine the stereochemistry at the two centres mentioned.

Hayatinin ethyl ether obtained by the ethylation of hayatinin with triethylanilinium chloride, when subjected to sodium and liquid ammonia reduction gave a non-phenolic compound, m.p. 89°, $[\alpha]_D 0°$ (chloroform), mass spectrum: $M^+=341$ ($C_{21}H_{27}NO_3$) and a phenolic compound, $[\alpha]_D 0°$ (methanol), which could not be crystallized, but had the same R_f value on TLC on silica gel (benzene-ethyl acetate-diethylamine, 7: 2: 1) as N-methylcoclaurine. The O,O′-dimethylmethiodide of this compound, m.p. 131°, was identical with an authentic sample of O,O′,N-trimethylcoclaurine methiodide in respect of m.p., m.m.p. and infrared spectra. This establishes the structure of the phenolic fragment as N-methylcoclaurine (II).

The NMR spectrum of the non-phenolic fission product showed six protons in the aromatic region, two singlets for three protons each at τ 6·12 and τ 6·18 (2-OCH_3), another singlet of three protons at τ 7·43 (1-NCH_3) and a triplet of three protons at τ 8·67 ($J=7·0$ cps) (OCH_2CH_3). The absence of any signal at τ 6·4-6·5 indicated the 0-ethylation



(I)

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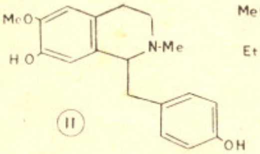
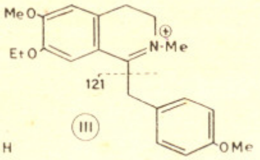
*Part II, *see* Bhattacharji, S., Roy, A. C. & Dhar, M. L., *J. scient. ind. Res.*, **21B** (1962), 428.

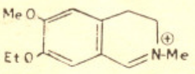
at the 7-position[7,8]. This meant that the free phenolic hydroxyl group in hayatinin was at the 7-position. Deuteration studies with hayatinin also showed that there were no protons *ortho* or *para* to the phenolic group. The assignment of the phenolic hydroxyl at the 7-position was also supported by the mass spectrum of the non-phenolic compound which showed, apart from a weak molecular ion peak (M^+) at 341 and a medium sized peak at m/e 340, abundant ions at m/e 220, 190, 177 and 121. The M-1 peak at m/e 340 corresponded to the ion (III) and the base peak (m/e 220) to (IV) which are characteristic of the fragmentation of benzyltetrahydroisoquinoline alkaloids[9]. The m/e 190 ion could presumably be formed by a further loss of -OMe with a proton transfer in ion (IV) and the ion m/e 121 by a fission of ion (III) as indicated by dotted lines.



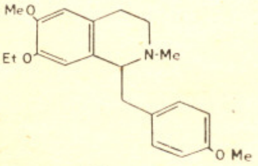
(II)



(III)



(IV)



(V)

All this evidence led to the formulation of (V) for the non-phenolic fragment. This was confirmed by its synthesis in an unambiguous manner as shown in the Chart 1.

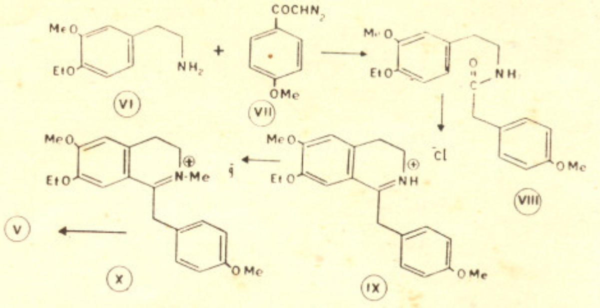


Chart 1 — Synthesis of non-phenolic component identified as (V)

β-(3-Methoxy-4-ethoxy) phenylethylamine[10] was condensed with 4-methoxyphenyldiazoketone[11] according to the method of Jeffreys[12] to yield the amide (VIII). The cyclization of the amide, methylation and subsequent reduction of the $>C{=}N-$ with sodium borohydride to yield (V) was carried out by the procedure of Jain[13].

The confirmation of the phenolic and non-phenolic fragments as (II) and (V) respectively clearly establishes the structure of hayatinin as 4″-O-methylbebeerine as proposed by Bhattacharji *et al.*[1]. This structure is fully supported by the NMR spectra ($CDCl_3$) of hayatinin and its ethyl ether.

The main features of the mass spectrum of hayatinin are ions at m/e 608 (M^+), 312, 298, 296, 191, 190, 174, 162, 146 and 145 and that from hayatinin ethyl ether at m/e 636 (M^+), 326, 312, 296, 204, 191, 190, 174, 162, 146 and 145. These are in agreement with the structure (I; $R=R_2=\text{Me}$; $R_1=\text{H}$) now established for hayatinin.

An interesting finding of the sodium and liquid ammonia fission of hayatinin ethyl ether was the optical inactivity of the phenolic as well as the non-phenolic fragments. Uptil now, the optical inactivity of hayatinin itself was assumed to be due to the (+ −) nature of the two coclaurine fragments respectively. The inactivity of the fragments also now suggests that hayatinin isolated by us from this plant is a racemic mixture.

The work described in this paper was nearly complete when Prof. Haynes of West Indies University, Jamaica, kindly sent us an advance copy of a communication[14] on the work done in his laboratories on a strongly optically active compound from *Cissampelos pareira* L. of Jamaican origin, $[\alpha]_D$ +273° (*c*, 0·7 in chloroform), m.p. 164°, which they have identified as (+ +)-4″-*O*-methylcurine. Their work is, in general, on the same lines as ours although both their phenolic as well as the non-phenolic fission products from the sodium and liquid ammonia reduction differ, in their physical characteristics, from the ones which we obtained. Such differences were, however, to be expected since both the fragments in our degradation experiment are *dl*-mixtures. Further, there are minor differences in the mass spectra of the non-phenolic fragments obtained in the two cases. We have confirmed the structures of our fission products by unambiguous synthesis and comparison with authentic samples and the evidence adduced by them in the case of their optically active compound provides fresh evidence for the confirmation of the structure of hayatinin.

Experimental Procedure

Hayatinin ethyl ether — Hayatinin was ethylated with triethylanilinium chloride in the usual way; m.p. 174°; yield 80 per cent (Found: C, 73·71; H, 7·40; N, 4·60. $C_{39}H_{44}N_2O_6$ requires C, 73·59; H, 6·89; N, 4·40%).

Sodium and liquid ammonia fission of hayatinin ethyl ether — A solution of hayatinin ethyl ether (500 mg.) in benzene (10 ml.) and toluene (10 ml.) was added dropwise to stirred liquid ammonia (500 ml.) containing sodium (500 mg.). More sodium was added (total, 1·5 g.) until a permanent blue colour persisted. The stirring was continued for 2 hr, the mixture kept overnight and ammonia allowed to evaporate. Water (50 ml.) was added, the organic layer washed with 5 per cent sodium hydroxide solution (solution B), then with water and finally the solvent evaporated when the non-phenolic product crystallized out in plates. It was recrystallized from methanol and purified by sublimation under vacuum; m.p. 89°; yield 220 mg. (Found: C, 73·62; H, 8·21; N, 4·32. $C_{21}H_{27}NO_3$ requires C, 73·89; H, 7·92; N, 4·10%); methiodide (methanol-ether), m.p. 182° (Found: N, 2·62. $C_{22}H_{30}NO_3I$ requires N, 2·89%).

The sodium hydroxide solution (solution B) containing the phenolic fragment was saturated with ammonium chloride and extracted with chloroform thrice. The organic layer was dried and on evaporation gave a residue (240 mg.) which was chromatographed on neutral alumina. The eluate from chloroform-methanol (49: 1) fraction gave 110 mg. of the phenolic compound. This, however, could not be crystallized. It was methylated with diazomethane and the product converted into the methiodide which was crystallized (methanol-ether); m.p. 131°; $[\alpha]_D$0° (methanol); yield 60 mg.; m.p. and infrared spectrum showed it to be identical with O,O′,N-trimethylcoclaurine methiodide.

p-Methoxy-N-(4-ethoxy-3-methoxyphenylethyl)-phenylacetamide—This amide was prepared in quantitative yield by condensing β-(4-ethoxy-3-methoxy)phenylethylamine (1·2 g.) and *p*-methoxyphenyl diazoketone (1·10 g.) by Jeffreys' method using *o*-dichlorobenzene instead of *o*-dichlorotoluene; m.p. 102° (from benzene-petrol ether) (Found: C, 70·21; H, 7·52; N, 4·21. $C_{20}H_{25}NO_4$ requires C, 69·97; H, 7·28; N, 4·03%).

1-(*p-Methoxybenzyl*)-6-*methoxy*-7-*ethoxy*-3,4-*dihydroisoquinolinium chloride* — This was prepared by heating the amide (2·5 g.) in dry toluene (40 ml.) with phosphorus oxychloride (0·75 ml.) at 115-20° for 1 hr and working up to give a hygroscopic product in 50 per cent yield. This was used directly, without isolation for the preparation of the methiodide.

1-(*p-Methoxybenzyl*)-6-*methoxy*-7-*ethoxy*-3,4-*dihydroisoquinoline methiodide* — A solution of the free base obtained above in benzene was treated with methyl iodide in an atmosphere of nitrogen. On keeping overnight, a yellow deposit of the methiodide was obtained in a quantitative yield. This was crystallized from methanol-ether; m.p. 158°.

1-(*p-Methoxybenzyl*)-6-*methoxy*-7-*ethoxy-tetrahydroisoquinoline* — Sodium borohydride (0·8 g.) was added during 1 hr to a solution of the methiodide (1·0 g.) in methanol (50 ml.). The reaction was kept stirred and under nitrogen during this time. The product was worked up in the usual manner and crystallized as its hydrochloride (methanol-ether); m.p. 136° (Found: C, 63·51; H, 7·51; N, 4·08. $C_{21}H_{27}NO_3,HCl,H_2O$ requires C, 63·71; H, 7·45; N, 3·54%). The free base obtained from this was crystallized from methanol; m.p. 89° (Found: C, 73·71; H, 8·04; N, 4·45. $C_{21}H_{27}NO_3$ requires C, 73·89; H, 7·92; N, 4·10%). The infrared spectrum of this compound was identical with that of the non-phenolic fragment obtained by sodium and liquid ammonia reduction of hayatinin ethyl ether. The methiodide of the base crystallized from methanol-ether; m.p. 182°.

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