Chemical Investigation of *Alhagi pseudalhagi* (Bieb.) Desv.: β-Phenethylamine and Tetrahydroisoquinoline Alkaloids

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Abstract □ From the stems of *Alhagi pseudalhagi* (Bieb.) Desv. (Leguminosae), five β-phenethylamine and one tetrahydroisoquinoline alkaloids were isolated and characterized. On the basis of chemical transformation and spectral (UV, IR, PMR, and mass) evidence, the alkaloids were identified as β-phenethylamine (I), *N*-methyl-β-phenethylamine (II), hordenine (III), 3,4-dihydroxy-β-phenethyltrimethylammonium hydroxide (IV), *N*-methylmescaline (V), and salsolidine (VI). This is the first time that the occurrence of a 3,4,5-trioxygenated β-phenethylamine such as V has been demonstrated outside the family Cactaceae. The roots of this plant contain essentially the same alkaloids but in poorer yields. Preliminary pharmacological studies of the total alkaloids indicated that some curative properties of the plant extract are due to these entities.

Keyphrases □ *Alhagi pseudalhagi*—isolation, identification of five β-phenethylamine and one tetrahydroisoquinoline alkaloids □ 3,4-Dihydroxy-β-phenethyltrimethylammonium hydroxide—isolation, identification from *Alhagi pseudalhagi* □ Salsolidine—isolation, identification from *Alhagi pseudalhagi* □ Medicinal plants—*Alhagi pseudalhagi*, chemical investigation and pharmacological screening

Alhagi pseudalhagi (Bieb.) Desv. (syn. *A. camelorum* Fisch., *A. maurorum* Baker) (Leguminosae: Lotoidae) is a low erect shrub, widely distributed in the gangetic plains in India. The plant is bitter and acrid with a distinct flavor and finds uses in the indigenous system of medicine for a variety of purposes (1, 2). Pharmacological screening with a 50% alcoholic extract of the whole plant showed (3) antiprotozoal, spasmolytic, cardiotonic, and anticancer (sarcoma 180 in mice) activities. Previous reports (4, 5) indicated only the presence of an alkaloid in this plant. A recent investigation (6) in the authors' laboratory resulted in the isolation of a considerable amount of an alkaloid mixture from its fresh stems. Separation of these alkaloids into individual entities and identification of the major alkaloids have now been achieved.

RESULTS AND DISCUSSION

The stems and roots of *A. pseudalhagi* were separately processed for alkaloids. Gradient pH extraction (7), fractionation into phenolic and nonphenolic bases over a resin[1] column (8), precipitation of the reineckate salts under basic and acidic conditions, and their regeneration over ion-exchange resins (7, 8) resulted in the isolation of five β-phenethylamine and one tetrahydroisoquinoline alkaloids from the stems of *A. pseudalhagi*. The alkaloids are β-phenethylamine (I), *N*-methyl-β-phenethylamine (II), hordenine (III), 3,4-dihydroxy-β-phenethyltrimethylammonium hydroxide (IV), *N*-methylmescaline (V), and salsolidine (VI). This is the first demonstration of the occurrence of a 3,4,5-trioxygenated β-phenethylamine (V) outside the family Cactaceae.

In addition to the mentioned alkaloids, two partially characterized β-phenethylamine bases (one secondary and one quaternary) and a liberal amount of choline were obtained from the stems of this plant. The roots contained essentially the same alkaloids but in poorer yields.

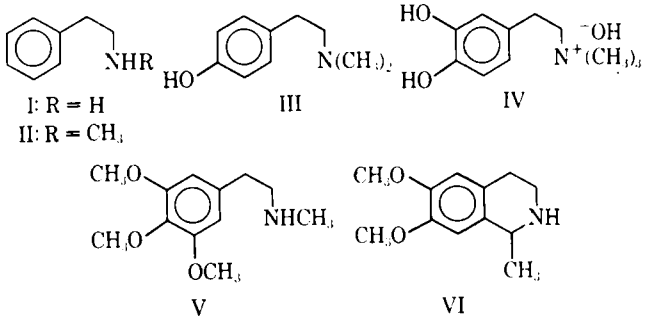
Cooccurrence of aryl-unsubstituted β-phenethylamines (I and II), the oxygenated analogs (III–V), and the cyclized compound (VI) has not been encountered before in nature. In the members of the Cactaceae, which are known to elaborate liberally 3,4,5-trioxygenated β-phenethylamines and their cyclized analogs, occurrence of mono- and dioxygenated compounds is a rare event (9). Also, aryl-unsubstituted compounds (I and II) have not been reported in the Cactaceae. These observations seem to indicate that *A. pseudalhagi* is a phylogenetically complex species, and further studies with related species in this genus are warranted.

Preliminary pharmacological studies with the total alkaloids of *A. pseudalhagi* indicated that the stimulant and expectorant properties of the plant extracts (1, 2) are due to these entities. Details of these findings will be reported soon.

EXPERIMENTAL

Extraction of Alkaloids—The principle of isolation and separation of the alkaloids involved utilization of differential solubility in solvents of graded polarity (petroleum ether, chloroform, and ethanol) in the presence and absence of fat, difference in base strengths, and phenolic and nonphenolic characters. The petroleum ether extract was kept aside for further examination for less basic constituents. The alkaloid mixtures from the ethanol extract were broadly divided into three groups (7): chloroform-soluble acetates, chloroform-soluble strong bases, and water-soluble bases. Subsequently, the alkaloids were separated into phenolic and nonphenolic bases in the usual way (8). The individual alkaloids were obtained by chromatography of the mixtures over neutral alumina[2] and preparative TLC[3] (2-mm. thickness). Monitoring of the column chromatographic runs was accomplished at each stage of purification by TLC. Two solvent systems were used as developers: Solvent 1, chloroform–methanol–concentrated ammonia (80:20:1); and Solvent 2, chloroform–methanol–acetic acid (75:15:10). Dragendorff's, tetrazotized *o*-dianisidine (8), and α-nitroso-β-naphthol–nitrous acid (10) reagents were used for staining. The identity of the homogeneous alkaloids was established by their co-TLC behavior with authentic markers (10–12), by chemical transformation into suitable derivatives where possible, and from correspondence of melting point, optical rotation, and UV, IR, PMR, and mass spectra of the alkaloids with those reported in the literature (9, 13, 14). Satisfactory combustion analyses were obtained for all reported compounds.

In a typical experiment, air-dried and milled stems of *A. pseudal-*



[1] Amberlite IRA-400.

[2] Brockmann, activity grade about III.

[3] Silica gel G, E. Merck.

hagi[4] (10.3 kg.) were processed for alkaloids. The description of the alkaloids is given here in the order of their isolation.

β-Phenethylamine (I)—Column chromatography and preparative TLC (Solvent 2) of the chloroform-soluble acetates afforded (from the upper preparative TLC zone) β-phenethylamine as a pale-brown liquid (0.18 g.) (identified by co-TLC, IR, melting point, and mixed melting point of the picrate).

***N*-Methyl-β-phenethylamine (II)**—The lower zone of the previously mentioned preparative chromatogram gave *N*-methyl-β-phenethylamine as a brown oil (72 mg.) (identified by co-TLC, IR, mass spectrometry, melting point, and mixed melting point of the picrate).

Salsolidine (VI)—Preparative TLC (Solvent 1) of the nonphenolic fraction from the chloroform-soluble strong bases furnished (from the middle zone) salsolidine as a semisolid mass (42 mg.) (identified by co-TLC, UV, IR, mass spectrometry, PMR, melting point, and mixed melting point of the base hydrochloride; $[\alpha]_D^{25}$ 0°.

***N*-Methylmescaline (V)**—From the preparative chromatogram, the lower zone of the nonphenolic alkaloid fraction afforded *N*-methylmescaline as a brown liquid (9 mg.) (identified by co-TLC, UV, and mass spectrometry). No solid derivative could be prepared due to the lack of sample. However, correspondence of spectral data (9, 13) and co-TLC behavior (Solvents 1 and 2) with an authentic synthetic sample, prepared from mescaline by treatment with formaldehyde and reduction of the anil with sodium borohydride in methanol (15), established its identity as *N*-methylmescaline.

Hordenine (III)—Column chromatography and preparative TLC (Solvent 1) of the phenolic alkaloid fraction afforded hordenine as light-brown needles (38 mg.) (identified by melting point, mixed melting point, co-TLC, UV, and melting point and mixed melting point of the methiodide).

3,4-Dihydroxy-β-phenethyltrimethylammonium Chloride/Hydroxide (IV)—The water-soluble bases, after separation of choline in the usual way (7, 16), gave two phenolic quaternary chlorides: R_f 0.11 (major) and 0.20 (minor) (Solvent 2). These were separated by preparative TLC using the same solvent system. The hydroxide of the major component, prepared by passing an alcoholic solution of the chloride through De-Acidite FF (7), readily suffered aerial oxidation. The quaternary chloride (18 mg.) showed UV λ_{max} 218, 230 (sh), and 283 nm., and $\lambda_{max}^{C_2H_5OH-NaOC^1OCH_2\text{-}H_2BO_3}$ 242 and 310 nm., characteristic of *o*-dihydroxy-β-phenethylamine chromophore (14, 17). The PMR spectrum of the quaternary chloride (in D_2O) showed signals at δ 7.1 6.3 (3H, m, unsymmetrical aromatic protons), 3.5–2.7 (4H, m, methylenes), and 3.2 (9H, s, quaternary *N*-methyl) p.p.m., which were consistent with the assigned structure. Co-TLC with an authentic sample of 3,4-dihydroxy-β-phenethyltrimethylammonium chloride, prepared from homoveratrylamine (18), established the correctness of this conclusion.

[4] The identity of the plant material was confirmed by Dr. C. S. P. Rao, Department of Botany, Banaras Hindu University. A voucher specimen has been kept at the Department of Pharmaceutics, Banaras Hindu University.

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