Free Amino Acids of *Plumbago zeylanica*

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PLUMBAGO zeylanica Linn[1]. (Bengali : chitta ; Fam. : *Plumbaginaceae*) occurs as a wild species in Tripura, West Bengal, Southern India, Ceylon, *etc.* The plant has been in use in Indian medicine since the period of Charaka in the treatment[1,2] against inflammations, piles, krimi, ulcer and kustha. The plant has been previously subjected to chemical investigations and the presence of 1,4-naphthaquinones, naphthalenones, flavonoids, aromatic hydroxy acid and sitosterol has been reported[2,3]. As a part of our programme of phytochemical screening on Indian medicinal and related plants, we have investigated the alcohol extract of the aerial parts of this plant and report herein the isolation, identification and relative occurrence of nine amino acids.

The aerial parts of the plant, *P. zeylanica* were collected from the wasteland area of Hooghly, West Bengal in February, 1985. The air-dried aerial parts (1 kg) were powdered and extracted in a Soxhlet with petrol (b.p. 60–80°) and 90% ethanol successively. The alcoholic extract was concentrated and decolourised by refluxing with activated charcoal. The light greenish filtrate was evaporated under reduced pressure to a gummy residue (60 g). The residue (1.5 g) was demineralised in the usual manner[4] by passing through a column of Amberlite IR 120(H). The column was washed with water till the eluate gave negative test with Molisch reagent for carbohydrates. The amino acids were then eluted with 2*N* HCl. The total eluate was concentrated under reduced pressure and chromatographed by descending technique on Whatman no. 1 paper in two directions[5] using solvent system n-BuOH—AcOH—H_2O (12 : 3 : 5, solvent I) in one direction, and PhOH—H_2O—NH_4OH (5 : 1.25 : 0.9, solvent II) in another direction. Amino acids were located on chromatogram by spraying ninhydrin reagent followed by heating the paper at 100° for a few minutes. The following amino acids were identified (co-pc with authentic samples) : glycine, alanine, tyrosine, threonine, methionine, hydroxyproline and histidine.

A part of the crude residue (2 g) was also chromatographed through Amberlite IRA 400(OH) for acidic and neutral amino acids. The chromatogram was desalted by washing with water and then amino acids were eluted with 1*N* acetic acid. The eluate was concentrated under reduced pressure and paper chromatographed in solvent systems I II. Three amino acids tryptophan, tyrosine and aspartic acid, were identified by co-pc with authentic samples. Tyrosine was also detected in the eluate of the cation-exchange resin, Amberlite IR 120(H).

All these amino acids from their mixture were isolated as nindydrin complexes by preparative pc[6] on Whatman no. 3 using solvent system I and were estimated colorimetrically by a Bausch & Lomb Spectronic 20 spectrophotometer, in the usual manner[7].

The relative occurrence of the amino acids (in μg per g of air-dried aerial parts of the plant) was found to be aspartic acid, 9.9 ; tryptophan, 9.7 ; tyrosine, 4.5 ; threonine, 4.1 ; alanine, 3.5 ; histidine, 3.4 ; glycine, 1.7 ; methionine, 0.5 ; and hydroxyproline, 0.2.

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