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Identification of Amino Acids Present in The Leaves and Stem of *Ipomoea carnea*

E. KHATIWORA, V.B. ADSUL† and N.R. DESHPANDE*
T.R. Ingle Research Laboratory, Department of Chemistry,
S.P. College, Pune-411 030, India
Tel: (91)(20)25440968; E-mail: anurag.deshpande@rediffmail.com

Air shade dried powdered material of stem and leaves were used for detection of amino acids from *Ipomoea carnea*. Paper chromatographic technique was used with different mobile phases. Specific amino acids were detected in specific mobile phases. The obtained amino acids were identified by comparing with standards. Total 18 amino acids in the leaves and 7 amino acids in the stem were detected. Two mobile phases were found suitable for the amino acids present in the leaves and only one for those present in the stem.

Key Words: *Ipomoea carnea*, Paper chromatography, Amino acids.

INTRODUCTION

Ipomoea carnea is a native of South America and available in plenty in all the states of India due to its adaptation to the Indian climatic conditions[1]. This non-woody plant is cheap, representing an annually renewable source of high quality fibers that can be successfully grown in temperate and tropical climatic conditions, without requiring much attention[1]. It is frequently found in planes and low lands near water sources[2]. It belongs to convolvulaceae family and fistulosa subfamily[3]. It is an ornamental plant due to its variety of flowers which appear pale rose, pink or light violet and whitish blue[4]. It is large, diffuse shrub, with milky juice, grows to a height of 5 m. The stem is thick and develops into a solid trunk over several years with many branches from base. The stem has an upper part which is green due to the chlorophyll of primary bark and a lower part which is gray due to the formation of secondary bark[4]. Flowers are large and funnel shaped, fruits are glabrous capsule, seeds are silky and flowering and fruiting season is throughout the year[5]. It is a green manure crop and also used as a folk medicine[5]. Ash of leaves is used for the treatment of skin disease in some rural areas of Chhattisgarh, India[5]. This non-woody fibrous plant has the greatest promise as a supplementary source of useful fiber species and also has good opportunities for industrial acceptance in India. It can be used to monitor SO_2 emission from power plants[6]. Several reports

†Y.M. College, Department of Chemistry, Bharati Vidyapeeth University, Pune-411 038, India.

are available on the biological activities of *Ipomoea carnea*. The cardiac effects of aqueous extract of the fresh leaves of *Ipomoea carnea* using mouse and frog hearts were studied. It was suggested from the data that the aqueous extract of *Ipomoea carnea* produces positive inotropic effect on isolated frog heart possibly by sodium extrusion or release of the intracellular calcium[7]. Antimicrobial activity of metal complexes prepared from the leaves proteins of *Ipomoea carnea* was reported[8]. However, no report on investigation of the amino acid pattern from the leaves and stem of *Ipomoea carnea* is available in literature. It plays important role in plant metabolism. Considering the fact, investigation of amino acids from the leaves and stem has been carried out.

EXPERIMENTAL

Whatman filter paper No. 1 was used for paper chromatography. Amino acid kit (CHH laboratory reagent) was used for standard amino acids. Leaves and stem of *Ipomoea carnea* were collected from river banks, Pune, Maharashtra, India, shade dried and powdered. Three extracts were prepared by using weighed quantity of powder in known volume of water, 9 % (w/v) aqueous sodium chloride solution and ethanol. Extraction procedure was repeated 3 times using chloroform to remove chlorophyll. The chlorophyll layer was separated and the remaining part was used for amino acid analysis[9]. Eighteen and seven amino acids were detected and identified from leaves and stem respectively after using ninhydrin spray. Following mobile phases were tried to screen out the best mobile phase for separating the amino acids present in the stem and leaves by paper chromatographic technique: (i) phenol:water (1:1), (ii) water:butanol:acetic acid (5:4:1), (iii) *n*-hexane:*n*-butanol: methanol:acetic acid:water (0.5:3:1:2:3.5), (iv) *n*-butanol:acetone:water (1:3:2), (v) *n*-butanol: acetone:water (1:3:1), (vi) *n*-propanol:ammonia (7:3), (vii) *n*-butanol: water:ethanol (2:1:1), (viii) *n*-butanol:ethanol:water:pyridine (2:0.5:1:1.5), (ix) *n*-butanol:ethanol: water:pyridine (3:2:3:2), (x) *n*-butanol:pyridine:water (2:0.5:2.5), (xi) pyridine: ammonia (7:3).

Out of these, mobile phases 5 and 9 were found suitable for the leaves and 11 was found suitable for stem. The experimental extracts were spotted on the chromatographic paper along with standard samples. The mobile phase was allowed to run to a certain height and the chromatogram was dried at room temperature. Ninhydrin solution was sprayed on the paper and again dried at room temperature. The R_f values of the amino acids of the experimental samples were determined and compared with the standards. Tables 1-3 below enlist the amino acids present in the leaves and stem of the plant.

RESULTS AND DISCUSSION

Amino acids present in the leaves: Both the mobile phases 5 and 9 showed the presence of DL-2 amino-N-butyric acid, L-cysteine monohydrochloride, L-histidine monohydrochloride, DL-serine, DL-threnonine and DL-valine.

TABLE-1
AMINO ACIDS DETECTED IN MOBILE PHASE 5:
BUTANOL:ACETONE:WATER (1:3:1)

Name of the amino acids	R_f values for standard amino acids	R_f values for amino acids of the leaves
DL-2 Amino-N-butyric acid	0.416	0.401
L-Cystein mono hydrochloride	0.489	0.496
L-Glutamic acid	0.146	0.146
L-Histidine mono hydrochloride	0.206	0.212
DL-Norleucine	0.343	0.328
DL-Iso-leucine	0.569	0.577
L-Leucine	0.212	0.212
L-Ornithine-hydrochloride	0.058	0.051
DL-Serine	0.226	0.240
DL-Threonine	0.292	0.292
DL-Valine	0.474	0.474
L-Proline	0.372	0.387
L-Hydroxy proline	0.278	0.278

TABLE-2
AMINO ACIDS DETECTED IN MOBILE PHASE 9:
BUTANOL:ETHANOL:WATER:PYRIDINE (3:2:3:2)

Name of the amino acids	R_f values for standard amino acids	R_f values for amino acids of the leaves
DL-2 Amino-N-butyric acid	0.386	0.357
L-Arginine monohydrocloride	0.071	0.064
Aspartic acid	0.193	0.193
L-Cysteine mono hydrochloride	0.328	0.343
L-Histidine mono hydrochloride	0.164	0.142
L-Lysine mono hydrochloride	0.078	0.107
DL-Methionine	0.235	0.230
DL-Serine	0.200	0.200
DL-Threonine	0.285	0.285
L-Tyrosine	0.250	0.250
DL-Valine	0.486	0.500

TABLE-3
AMINO ACIDS DETECTED IN MOBILE PHASE 11: PYRIDINE:AMMONIA (7:3)

Name of the amino acids	R_f values for standard amino acids	R_f values for amino acids of the leaves
Aspartic acid	0.026	0.026
Cysteine	0.019	0.019
L-Proline	0.192	0.215
L-Hydroxy proline	0.111	0.107
2-Amino-N-butyric acid	0.230	0.230
Iso leucine	0.430	0.415
L-Tyrosine	0.384	0.353

According to the concentration of compounds, showed by the chromatographic spots, the amino acids were classified approximately as major, minor and negligible amounts. In the mobile phase 5, 13 amino acids were detected. L-Glutamic acid, L-histidine mono hydrochloride, DL-norleucine, DL-serine, L-hydroxy proline were found in major amount while cystein mono hydrochloride, DL-iso-leucine, DL-valine, L-proline were found in minor amount. The remaining DL-2 amino-N-butyric acid, DL-threonine, L-orithine-hydrochloride, L-leucine were found in negligible amount.

The presences of 11 amino acids were detected in mobile phase 9. Aspartic acid, L-histidine mono hydrochloride, DL-threonine and L-tyrosine were found in major amount. DL-2 Amino-N-butyric acid, L-lysine mono hydrochloride, DL-methionine and DL-serine were found in minor amount. L-Arginine monohydrochloride, L-lysine monohydrochloride and DL-valine were found in negligible amount. Total 18 amino acids were present.

Amino acids present in the stem: Mobile phase 11 was used for stem extract. It was pyridine:ammonia (7:3). It indicated the presence of seven amino acids in the stem in paper chromatogram. They were aspartic acid, cysteine, L-proline, L-hydroxyproline, 2-amino-N-butyric acid, iso leucine and L-tyrosine. Among them, L-proline, L-hydroxyproline, 2-amino-N-butyric acid and L-tyrosine were found in major, iso leucine was found in minor and aspartic acid and cysteine were found in negligible amount.

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