



Study of Amino acids of seed extract of Bitter Gourd (Cucurbitaceae)

Suresh Narayan Baitha¹ and V K Prabhat²

1.Department of Botany, S N S, College, Motihari, B R A B U Muzaffarpur, INDIA.

2.P G Department of Botany, Dr A H Rizvi College ,Karari, Kaushambi, U P,INDIA.

ABSTRACT

The present paper deals with the detection of amino acids in the seed extracts of Bitter Gourd. Bitter Gourd is a good source of medicines and proteins. Bitter gourd has been used in various traditional medicine systems for a long time, for preventing and treating various diseases. Its fruits and pulp are used in treating asthma, constipation, colic, diabetes, inflammation, skin diseases etc. 5 g of germinated seeds were grinded in 25ml. of 80% alcohol in mortar and pestle. The tissue homogenate was centrifuged at 7000rpm for 15min. After centrifugation the supernatant was filtered and evaporated on water bath followed by addition of 1ml of distilled water to the dried filtrate. Now a drop of extract was spotted slowly to the chromatographic paper which was placed in chromatographic jar containing solvent (n-butanol: acetic acid: water-4:1:5) for its movement. When the solvent reached near the top, the paper was taken out from the jar, dried and sprayed uniformly 0.1% ninhydrin with the help of atomizer. The paper was dried in an oven at 90°C for 5 minutes. Coloured spots of amino acids appeared on the chromatogram and the identification of the amino acids was confirmed with their R_f values. On the basis of R_f value and colour, five amino acids namely Leucine, valine, ornithin, Arginine and phenylalanine were detected in the seed extract of Bitter Gourd.

Keywords: Bitter Gourd, Ninhydrin, n-butanol, acetic acid.

INTRODUCTION

Cucurbitaceae is one of the largest valuable families from agricultural and pharmacological point of view. The Cucurbitaceous plants are cultivated throughout the world. They are distributed in plains as well as hills. Bitter Gourd is a good source of medicines and proteins. Bitter gourd has been used in various traditional medicine systems for a long time, for preventing and treating various diseases. Its fruits and pulp are used in treating asthma, constipation, colic, diabetes, inflammation, skin diseases etc.

The amino acids are colourless amphoteric compounds with at least one amino (-NH₂) and one carboxylic acid (-COOH) functional groups that form the basic building blocks of proteins (Dayal and Lal, 1984). In nature a large number of amino acids occur in free or conjugated form. The sequence of amino acids in the peptide chain is specific for each protein and potentially capable of enormous diversity of proteins (Pandey and Sinha, 1997). Amino acids occupy a key role in nitrogen metabolism. Some of the amino acids such as tyrosine is converted into hormones thyroxin and adrenaline, glycine into the formation of haemoglobin and tryptophan in the formation of vitamin nicotinic amide and hormone IAA (Srivastava *et al.*, 2011).

Very often in plants during disease conditions, the free amino acid composition exhibits a change, hence the measurement of total free amino acids gives the physiological and health status of plants

MATERIAL AND METHODS

5g. of germinated seeds were grinded in 25ml. of 80% alcohol in mortar and pestle. The tissue homogenate was centrifuged at 7000rpm for 15min. After centrifugation the supernatant was filtered and evaporated on water bath followed by addition of 1ml of distilled water to the dried filtrate. Now a drop of the extract was spotted slowly to the chromatographic paper (whatmann's filter paper no 1) which was placed in chromatographic jar containing the solvent (n-butanol: acetic acid: water-4:1:5) for its upward movement. When the solvent reached near the top, the paper was taken out from the jar, dried and sprayed uniformly 0.1% ninhydrin with the help of atomizer (Moore and Stein, 1948). The paper was dried in an oven at 90°C for 5 minutes. Coloured spots of amino acids appeared on the chromatogram and the identification of the amino acids was confirmed with their Rf values.

RESULTS AND DISCUSSION

The content of amino acids ranged from 0.23 to 0.56 µg/g of sample weight. On the basis of the different Rf value and colour, five amino acids namely valine, leucine, ornithine, arginine and phenylalanine were detected from the sample extract of Bitter Gourd (table 1). Interestingly the pH value of amino acids (valine, leucine and arginine) were noted as 4 while that of phenylalanine and ornithine were 5.

Table 1: Amino acids in seed extracts of Bitter Gourd.

S.No	Amino acids	Sample weight	pH Level	Colours
1	Leucine	0.24	04	Violet
2	Valine	0.16	04	Medium violet
3	Ornithine	0.46	05	Bluish gray
4	Phenylalanine	0.54	05	Deep violet
5	Arginine	0.44	04	Deepbluishgray





The amino acids are the essential constituents of biological system and play an important role in the metabolism of living organisms. A change in the quantity and number

of amino acids can alter the quality of proteins resulting biochemical abnormalities. Srivastava *et al.* (2011) reported nutritional evaluation and amino acids composition in *Talinumtriangulare* while Roy (1981) identified toxic amino acids and proteins in leguminous plants species. Roy and Rao (1971) worked on the isolation and purification of trypsin inhibitor in *Lathyrus*. Singh and Singh (2010) reported the impact of sporozoites on free amino acids in *Pricini*. Chourasia (1992) analyzed amino acids in radishes while Kumar and Anandhiwal (2010) determined protein in Kalmegh.

The study on the quality of amino acids in the seed extract reveals that seed protein of Bitter Gourd might have been composed of five amino acids (leucine, valine, ornithine, phenylalanine and arginine), however the molecular analysis of seed protein may confirm.

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