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PROTEIN AND AMINO ACIDS OF *GREWIA ASIATICA*

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The chemical composition of falsa fruit *Grewia asiatica* including the seed was determined. Crude protein percentage, it's amino acid composition were evaluated. Electrophoretic pattern of proteins revealed the presence of two major proteins having molecular weights as 19,100 and 89,200 dalton. The quantity of phosphoserine was found to be higher in pulp as compared to other free amino acids (FAA), while the hydrolysate contained aspartic acid, glycine and tyrosine in excess. The specific amino acid profile may be used as an index for judgement of adulteration in pure juice. The significant quantity of sodium, potassium and calcium makes the fruit juice more palatable and nutritious.

Key words: Protein, Amino acids, *Grewia asiatica.*

INTRODUCTION

Falsa fruit *Grewia asiatica* is a small dark purple berry which is popular due to its spectacular taste and thirst quenching property. It is cultivated in South East Asia, and harvested in summer. The fruit is popular due to its unique quenching flavour and pulp is eaten as such after discarding seeds. Traditionally a sherbat is made with it by macreating the fruits water, and sugar and salt are added to taste. The fruit is readily fermentable and cannot be stored for more than 24 hours at room temperature. This fruit has not yet been commercially processed. The chemical composition of the fruit is not yet fully studied. The juice contained fructose, glucose and maltose while sucrose was completely absent [1]. Moisture content of the fruit were found to be 70-75 % [2]. Khurdiya [3] has reported in 1981 two anthocyanin pigments, delphinidine-3-glucoside and cyanidin-3-glucoside in falsa fruit. The fruit and its juice may readily perish but the fermentation process is totally controlled by the addition of Na-benzoate [4]. The immense interest in evaluation of nutritional value of food has triggered the interest of scientists to explore the structure and functionality of proteins [5]. Amino acid analysis not only determine the protein quality but has also been used to discriminate between natural and artificial fruit juices [6]. The specificity of amino acid composition has established itself as a reliable tool for judging the authenticity of a fruit juice [7]. Adulteration of certain fruit juices with the juice of other cheaper fruits may also be detected by amino acid analysis [8].

MATERIAL AND METHODS

Reagent grade chemicals and double distilled water were used throughout the experimentation. Molecular weight marker proteins were obtained from Fluka. Protein content was estimated by micro Kjeldahl method. Metals were estimated by flame analyser (Gallenkamp model FGA-330 Gallenkamp).

Amino acid analysis. Free amino acids in juice were analyzed after precipitation of proteins by adding 10 % sulfosalicylic acid to the juice in ratio of 1 : 1. The mixture was shaken vigorously and kept for 30 minutes at 4° and finally centrifuged at 3000 g. The clear supernatant (25 ul) was used for analysis using automatic amino acid analyzer LC-6001 Biotronic-GmbH.

Amino acids were also estimated after hydrolyzing the pulp and seed separately with 6N HCl in sealed glass tubes incubated at 110° for 20 hrs. Finally HCl was removed in vacuum and amino acids were analyzed by the automatic amino acid analyzer.

Electrophoresis. (i) *Page.* Polyacrylamide gel electrophoresis was performed as described by Davis (1964). Polyacrylamide gel 7.5 % were run using Tris-glycine buffer pH 8.3 as upper electrode buffer and Tris-HCl pH 8.0 as lower electrode buffer. 100 ul of sample was loaded on each tube and a current of 2 mA per tube was maintained. Gels were stained by Commassie brilliant blue R-250.

(ii) *SDS-Page.* SDS polyacrylamide gel electrophoresis was performed according to [9] using discontinuous system with 10 % resolving and 2.5 % stacking gels. 100 ul of each sample was loaded and a current density of 2 mA per tube was maintained. The molecular weights of proteins were estimated using molecular weight marker ranging from 14,300 to 71,500 daltons (Fluka). Moisture contents were estimated by Ultra X Infra-red moisture tester from Jergens & GmbH & Co.

Crude fiber were determined according to the modified method of Bidwell and Bost [10].

Total ash was estimated according to the method of Boline and Schrenk [11].

Fat was estimated in seeds by Soxhlet extraction apparatus and in juice by liquid-liquid extraction method.

RESULTS AND DISCUSSION

The moisture, protein, fiber, carbohydrate, fat and ash contents of the whole fruits, pulp and seeds are reported in Table 1. The seeds are a rich source of protein, fat and fibers and they do not impart negative flavour, therefore, it suggests that in juice extraction seeds should also be included so that the amino acid content may also be increased. The high contents of phosphoserine and serine in the unhydrolysed juice and seeds in Table 2 is of special significance while evaluating the purity or dilution of the juice as has been reported by Joop [8] in assessing the quality of other fruit juices. It was also interesting to note that glutamic acid and β-alanine are completely absent in seed while only a small amount in hydrolysed juice is present. – amino butyric acid was not detected either in pulp or juice and it was present only in aqueous extract of seeds. Glycine and tyrosine were unusually very high in seed hydrolysates while they were absent in pulp hydrolysate.

The presence of certain amino acids in a specific ratio or the absence of certain amino acids is the most reliable parameter for judging the quality of a fruit juice and it appear as shown in Table 2 that the amino acid profile of Falsa may be used as a tool for quality assurance.

It was interesting to note that threonine was missing in seed hydrolysate as well as seed sap and it could be exploited in detecting the presence of pulp in seed extracts. Electrophoretic pattern of proteins revealed that two major proteins of molecular weight 19,100 and 89,200 are present as produced by SDS PAGE analysis shown in Fig. 1. However the simple PAGE analysis produced a single band (Fig. 2) showing that protein have equivalent electrical charge under the conditions mentioned.

Table 1. Composition of *Grewia asiatica.*

	Moisture (%)	Protein (%)	Fat (%)	Fiber (%)	Ash (%)
Whole fruit	75	1.5	–	18	2.08
Pulp	85	0.9	–	12.5	1.5
Seeds	20	5	7.2	55	–

Table 2. Amino acid composition of falsa fruit (m.mol/100 g)

	Pulp		Seed	
Amino acids	F.A.A.	Hydrolyzate	F.A.A.	Hydrolyzate
PHOS	8.23	–	22.82	–
TAU	5.41	–	13.38	–
THR	2.76	6.69	–	–
SER	7.33	4.45	58.18	–
PRO	3.68	–	19.92	–
GLY	3.19	–	14.82	62.51
ALA+CIT	1.40	–	27.88	–
α-NH – BA	–	–	11.62	–
MET	–	–	7.2	14.26
I.LEU	1.35	–	7.74	–
TYR	1.73	–	7.5	47.6
LYS	–	28.79	6.74	0.38
HIS	–	2.43	3.06	0.17
PHE	2.87	–	–	4.21
VAL	1.65	–	–	4.03
CYS	1.70	–	–	–
LEU	1.24	–	8.42	–
β-ALA	0.86	–	–	–
GLU	–	40.31	–	–
ASP	–	40.31	–	–

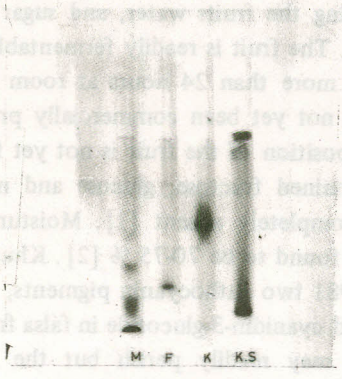

Fig. 1. (SDS – PAGE).

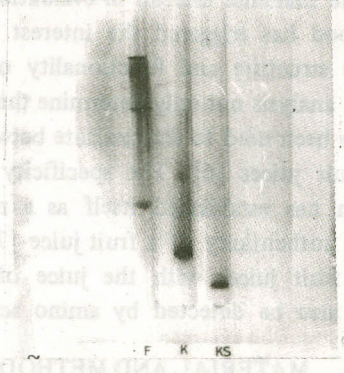

Fig. 2. (PAGE)

M = Molecular weight marker proteins (14000 to 85000 dalton); F = Falsa Juice (*Grewia asiatica*); K&K.S = Not related.

The sodium, potassium and calcium contents of fruit are given in Table 3, which shows that seeds extract is a good source of minerals.

Table 3. Analysis of falsa juice.

Sodium (ppm)	22
Potassium (ppm)	1250
Calcium (ppm)	260

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