STUDIES IN THE PROTEINS OF INDIAN FOODSTUFFS. PART V. The Alcohol Soluble Protein of Fenugreek (*Trigonella Fœnum Græcum*).

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In a previous communication Sreenivasa Rao and Sreenivasaya (*Jour. Indian Inst. Science*, 1932, **15A**, 122) have reported their findings on the albumin and the globulin of fenugreek. The present paper deals with the prolamin from the same source.*

EXPERIMENTAL.

The flour was first extracted with ether in a Soxhlet and the residue (3 Kg.) stirred up with 70 per cent. alcohol (9 L) in the cold for six hours. The mixture was then warmed on the water bath and maintained under reflux for 1 hour at the boiling temperature. The hot suspension was then filtered and the extract on cooling in the ice chamber deposits a white solid. After filtration, the extract was concentrated at 40° under reduced pressure. The concentrate (2 L) was allowed to stand overnight at room temperature when a white solid separated. The precipitate was washed consecutively with distilled water, 90 per cent. alcohol and ether. It was then dried, powdered to pass through a 100-mesh sieve and preserved in a desiccator over calcium chloride (Preparation I ; yield 5 g.)

On treating the concentrated alcoholic extract (1.5 L) with acetone (8 L), a copious precipitate was obtained a large portion of which was found to be soluble in water. The insoluble portion was separated on the centrifuge, washed repeatedly with distilled water and dehydrated with alcohol and ether. On drying, a light, white solid was obtained (Preparation II ; yield, 4 g.)

Both the above-mentioned preparations gave all the reactions for proteins and contained tyrosine, tryptophan, sulphur and phosphorus. On analysis they gave the following results. (Table I.)

* Since writing this paper a preliminary note on the prolamin of fenugreek prepared by a different method by A. Hassan and M. K. A. Basha has appeared in the *Biochemical Journal* (1932, 26, 1843.) That publication does not, however, relate to the composition of the prolamin.

TABLE I

Preparation	PERCENTAGES				
	Moisture	Ash	P_2O_5 [1]	Ash and moisture free	
				Nitrogen	Sulphur [2]
I	3.0	0.99	0.50	14.03	1.12
II	9.6	1.10	0.41	14.08	0.99

[1] Micro method of Pregl.
[2] Micro-combustion method of Carius.

The two preparations were then analysed by the method of Van Slyke as modified by Plimmer and his co-workers (*Biochem. J.*, 1925, **19**, 1004; 1927, **21**, 247). Tyrosine, tryptophan and cystine were estimated separately on the whole protein according to Folin and Marenzi (*J. Biol. Chem.*, 1929, **83**, 89).

The distribution of nitrogen is given in the following table.—

TABLE II

Form of Nitrogen	Percentages of total nitrogen			
	Preparation			
	I		II	
Acid insoluble Melanin ...	0.9		1.1	
Acid soluble Melanin ...	0.4		0.5	
Amide ...	20.2		20.0	
Basic ...	5.8		5.6	
Arginine ...		3.1		2.9
Histidine ...		0.7		0.7
Cystine ...		1.4		1.3
Lysine ...		0.7		0.7
Non-basic: ...	72.7		72.8	
Amino ...		70.2		70.4
Non-amino ...		2.5		2.4

A noteworthy feature of the prolamin from fenugreek is its low basic nitrogen content in which respect it is comparable only with eleucine from ragi (Hoffman and Gortner, Coll. Symp. Monograph, 1925, Vol. 2, 246; Niyogi, Narayana and Desai, private communication).

In Table III are given the percentages of the more important amino acids as estimated for the whole protein. The corresponding figures for ragi prolamin are cited for comparison.

TABLE III

	Source of prolamin		Method
	Fenugreek	Ragi	
Arginine ...	2.3	2.5	Direct
Histidine ...	0.4	1.4	Van Slyke
Lysine ...	0.5	0.5	,,
Cystine ...	3.0	2.5	Folin and Marenzi
Tyrosine ...	4·3	5·3	,,
Tryptophan ...	2.4	1.6	,,

SUMMARY.

The two preparations of the prolamin from fenugreek obtained by different methods were found to have very nearly the same composition. The prolamin is characterised by a low basic nitrogen content and high percentages of cystine and tryptophan and thus resembles the alcohol soluble protein of ragi isolated by one of us.

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