PROTEINS OF INDIAN VEGETABLES. DRUMSTICK.

(*MORINGA PTERYGOSPERMA* OR *GUILANDINA*) OR *HYPERANTHERA*.

By Y. V. S. Rau and V. Ranganathan.

Name.—(Nadkarni, K. M., *Indian Materia Medica,* 561) Sanskrit: *Shobhanjana, Dvishigru, Muringi;* English: *Horse radish, Drumstick plant;* French: *Moringa a grainestripteres;* Hindustani: *Sahinjan, Segve, Duk-Mungeka jhad;* Punjabi: *Schanjna;* Bengali: *Saijna;* Telugu: *Munaga;* Guzarathi: *Saragava;* Mahratti: *Shegat;* Tamil: *Murangai;* Kanarese: *Nugge;* Malayalam: *Murina;* Konkani: *Mashinga jhad;* Sinhalese: *Murunga;* Burmese: *Dandalobin;* Malay: *Kayloi, Ramoogie.*

Habitat.—A beautiful tree wild in the sub-Himalayan range and commonly cultivated in India and Burma.

Parts used.—The bark, root, fruit, flowers, leaves, seeds and gum.

Action.—Antispasmodic, stimulant, expectorant and diuretic. The fresh root is acrid and vesicant; internally stimulant, diuretic and antilithic. The gum is bland and mucilaginous. The seeds are stimulant. The bark is emmenagogue and even abortifacient. The flowers are stimulant, tonic and diuretic and useful to increase the flow of bile.

Uses.—Its leaves, flowers and immature capsules are eaten in curries. Grated *horse radish* (European) eaten at frequent intervals during the day and also at meals is said to banish the distressing cough that lingers after influenza. It contains sulphur and is therefore recommended for rheumatism, applied as a poultice for the neuralgia of the face. In short, the different parts of the Drumstick are abundantly used in medicine.

Preparation of the material.—Drumsticks got from the local market were cut into small pieces and dried in a hot air-oven at 45°C. The air-dry material was powdered to pass a 20-mesh sieve. The material obtained was analysed and found to contain:

TABLE I.

Results expressed as percentage on air-dry material.

Silica as SiO_2	Moisture	Protein	Ash	Phosphorus as P_2O_5	Calcium as Ca	Crude fibre	Ether extract	Carbohydrates	Sulphur
0·36	15·8	15·0	5·85	0·25	0·26	30·5	2·86	29·63	0·60

The above determinations were carried out according to standard methods of A.O.A.C.

Distribution of nitrogen in various solvents expressed as per cent. total nitrogen.

TABLE II.

Solvents	% N extracted
1. Water	37·4
2. Saline	1·8
3. 70% Alcohol	1·0
4. 0·1% Alkali	11·0

A separate estimation of the non-protein nitrogen was made as follows:—

10 g. of the air-dry material was extracted a number of times with 0.5% acetic acid until no more nitrogen was present in the extract. The combined extract was heated on a water-bath at 60°C. for 10 minutes, the precipitated protein filtered off, and the filtrate concentrated under reduced pressure at a temperature not exceeding 50°C. on a water-bath. The concentrate was made up to a known volume and aliquots taken for total nitrogen.

Per cent. non-protein = 44.4.

Isolation of Globulin.—500 g. of the material were treated with 3 litres of 10% saline, shaken for 1 hour and filtered. The residue was again extracted twice with 2 litres of 10% saline. The combined filtrates were dialysed till free from chloride. The precipitated globulin was centrifuged, redissolved in 1 litre of 10% saline and filtered. The filtrate was dialysed again running distilled water, till free from

chloride. The precipitated globulin was centrifuged, washed three times with water to free it from contaminated albumin, then with graded strengths of alcohol and finally with ether. On drying, an ash coloured solid was obtained (yield 1 g.).

The dialysate was acidified with a few drops of acetic acid and heated on a water-bath. Only traces of albumin were present.

Isolation of Prolamin.—The residue after saline treatment was treated with 3 litres of 70% alcohol and refluxed for 3 hours on a water-bath, filtered, the residue was again extracted with 2 litres of 70% alcohol. The hot suspension was filtered and the combined filtrate was concentrated at 40°C. under reduced pressure. On treating the concentrated alcoholic extract (500 c.c.) with acetone (6 litres), a copious precipitate was obtained, a large portion of which was soluble in water. The insoluble portion was separated on the centrifuge and dialysed till free from chlorides, centrifuged, washed repeatedly with distilled water and dehydrated with alcohol and ether and on drying, a light brown solid was obtained (yield 1.5 g.).

The above preparations gave on analysis the following results:—

TABLE III.

Preparation	Percentages				
	Moisture	Ash	P_2O	Ash and Moisture-free	
				Nitrogen	Sulphur*
I. Globulin ..	10·25	0·01	traces	15·6	1·58
II. Prolamin ..	9·37	0·77	0·41	14·02	1·43

* Micro method of Carius.

The two preparations (globulin and prolamin) were analysed by the method of Van Slyke (*Biochem. J.*, 1925, 19, 1004) as modified by Plimmer and his co-workers. Tyrosine, tryptophane and cystine were estimated separately on the whole protein according to Folin and Marenzi (*Jour. Biol. Chem.*, 1929, 83, 89). The distribution of nitrogen is given in the following table:—

TABLE IV.

Form of Nitrogen	Percentage of Total Nitrogen			
	Preparation			
	Prolamin		Globulin	
Acid insol. Humin	3·1		2·9	
„ sol. „	1·8		0·9	
Amide	8·1		10·7	
Total basic	25·9		26·9	
Arginine		22·6		15·6
Histidine		0·1		6·7
Cystine		1·0		0·9
Lysine		2·2		3·7
Total non-basic	60·9		58·9	
Amino		52·9		51·8
Non-amino		8·0		7·1
TOTAL ..	99·8		100·3	

In Table V are given the percentages of the more important amino-acids as estimated for the whole protein.

TABLE V.

Amino-acid	Prolamin	Globulin
Tyrosine	4·16	5·27
Tryptophane	3·17	1·71
Cystine	3·20	3·42

From the results it is evident that the globulin compares with any of the known vegetable globulins in its amino-acid make-up. But the prolamin differs from the other vegetable prolamins in that it characterises itself by a low amide and a high basic nitrogen content. It is striking that the amino-acid make-up of the prolamin and

globulin of the drumsticks is almost the same. However, the prolamin from drumsticks bears a resemblance to the prolamin from rice, as shown in the following table. The corresponding values for the prolamin from Fenugreek are given for comparison.

TABLE VI.

Percentage of Total Nitrogen in the form of		Prolamin from		
		Rice[1,2]	Drumstick	Fenugreek[3]
Arginine	1	13·2	..	3·1
	2	18·2	22·6	0·7
Histidine	1	6·2	..	..
	2	8·1	0·1	..
Lysine	1	3·4	..	..
	2	1·7	2·67	0·7
Cystine	1	1·3	..	..
	2	0·9	2·2	2·5
Tyrosine	.. .. ..	..	2·3	2·38
Tryptophane	.. .. ..	..	3·11	2·35

1. Hoffman, W. (*Journ. Biol. Chem.*, 1925, **56**, 501).
2. Tadokoro, T., and others [*Coll. Agric. Hokkaido, Imp. Univ.* (Sapparo, Japan), 1926, **18**, 175].
3. Rau, Y. V. S., and others (*Journ. Ind. Inst. Sci.*, 1933, **16A**, Part VIII, 85).

To assess the relative nutritive value of the prolamin of drumsticks, its amino-acid make-up is compared with the essential amino-acid make-up of a few animal proteins on the one hand and plant proteins on the other.

TABLE VII.

Amino-acid	Meat[4]	Casein[5]	Lactal-bumin[6]	Prolamin from Drum-stick	Corn[7]	Potato[8]
	Percentage part of nitrogen of the amino-acid on Total N					
Arginine	12·6	7·4	7·2	22·6	7·6	8·3
Histidine	10·4	6·2	4·6	0·1	2·1	3·8
Lysine	7·5	10·3	7·2	2·67	1·2	3·9
Cystine	1·5	0·2	7·3	2·20	0·2	3·1
Tryptophane ..	?	5·3	?	2·3	1·2	2·0
Tyrosine	1·2	1·5	1·5	3·11	0·9	?

4. Rottgers (*Nahrungsmittelchemie*, 1, 5 Aufl, 1926).
5. Van Slyke, D. D. (*Jour. Biol. Chem.*, 1913–14, 16, 531).
6. Osborne, Th. B., Van Slyke, D. D., Leavenworth, C. S., and Vinograd, U. M. (*Jour. Biol. Chem.*, 1925, 22, 259).
7. Osborne, Th. B. (*Ergebn. d. Physiol.*, 1910, 10, 47).
8. Sjollema, B., Rinkes, U. J. J. (*Zeitsch. fur. Physiol. Chem.*, 1911–12, 76, 369).

From the above table, it is clear that the prolamin from drum-stick contains about two times the quantity of arginine present in meat and nearly three times as that present in casein, lactalbumin and other vegetable proteins given for comparison, but it is deficient in lysine and histidine as compared to animal proteins. It contains a good percentage of tyrosine, tryptophane and cystine. Since milk contains a low percentage of cystine, it can supplement casein in diet.

The high arginine N as determined by Van Slyke's method is very striking and requires further confirmation by other methods especially that of Kossel (*Zeitsch. für. Physiol. Chem.*, 1911–12, 76, 369) as modified by Block (*Jour. Biol. Chem.*, 1935, 106, 457) which involves the isolation of amino-acid as flavianate. Work in this direction is in progress.

Our thanks are due to Prof. V. Subrahmanyan for his keen interest in the work and to the Madras Government for the award of a research scholarship to one of us (V. R.).

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[Received, 11–10–1937.]

1824-37.—Printed at The Bangalore Press, Bangalore City, by G. Srinivasa Rao, Superintendent.