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An Evaluation of Protein Quality of Fenugreek Seeds (*Trigonella foenumgraecum*) and their Supplementary Effects

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ABSTRACT

The chemical composition and the nutritional value of protein of whole as well as solvent-extracted fenugreek seeds were determined. Fenugreek seeds are rich in protein (25·5%), fat (7·9%), unavailable carbohydrate (48%), mucilaginous matter (20%) and saponins (4·8%). The replacement of casein diet up to 10% by fenugreek seeds (extracted) did not produce any deleterious effect on protein quality of casein as assessed by protein efficiency ratio (PER), protein and dry matter digestibilities and net protein utilization (NPU). Further increasing the level of fenugreek did reduce these parameters. The extraction of seeds improved, and cooking did not alter the quality of fenugreek seed protein.

INTRODUCTION

Fenugreek (*Trigonella foenumgraecum*) is mainly cultivated in India, Mediterranean countries and the Near East. The plant has many culinary uses. Its tender leaves are used as vegetables and its seeds as condiments. In Egypt, the seed powder is mixed with cornflour to bake bread (Taha El-Katib, 1947).

There is unanimous opinion among researchers on the causal relationship between high levels of serum cholesterol and incidence of coronary heart disease in the general population (Truswell, 1985). It has been suggested that hypercholesterolemia should be treated right from the age of three years and for this purpose long-term dietary measures have been

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advocated.[2] (Truswell, 1985). Studies carried out at our Institute showed that incorporation of fenugreek seeds at 5–10% level in the diet resulted in a significant reduction in serum cholesterol levels in rats fed a hypercholesterolemia-inducing diet (Sharma, 1984). The solvent-extracted seeds of fenugreek also showed this effect in man and experimental animals (Sharma, 1986; Valette *et al.*, 1984). The extracted seeds containing all the seed protein are obtained as an industrial byproduct after removal of steroidal sapogenins which are used for the synthesis of corticosteroid, contraceptive agents and sex hormones (Sauvaire & Baccou, 1976). Considering that these seeds may be used therapeutically for the control of hypercholesterolemia in children, the protein quality of the seeds was determined. Furthermore, data on chemical composition of fenugreek seeds are scanty and incomplete. There are only two reports (Sauvaire & Baccou, 1976; Elmadfa & Kuhl, 1976) on the evaluation of protein quality of fenugreek seeds whose results are contradictory. One of them reported negative PER but the other reported positive PER for these seeds, although experimental conditions in both the studies were identical. It is likely that the Indian variety may be different in protein quality from those grown in other parts of the world. The following investigations were therefore undertaken in rats to study (a) the protein nutritional quality of fenugreek seeds (whole and extracted), (b) the effect of supplementation of extracted seeds at different levels in a casein diet on growth and utilization of nutrients, (c) the effect of cooking on protein quality (since seeds are normally consumed after cooking).

MATERIAL AND METHODS

Fenugreek seeds were purchased locally and ground to pass a 40 mesh sieve. The powder was kept in an airtight container until used.

The seed powder was extracted for 16 h in Soxhlet apparatus with ether, followed by alcohol extraction for 24 h. The residue was air-dried for 4 days at room temperature. The defatted meal obtained was used for the experiment along with the unextracted whole seed powder. The defatted meal was not bitter (unlike the whole seeds).

Chemical composition

Whole, as well as solvent-extracted, seeds of fenugreek were analysed for moisture, protein, fat and ash according to AOAC methods (1970). Minerals such as Zn, Cu, Mg, Mn and Cr were estimated using an atomic absorption spectrophotometer. Dietary fibre and mucilage contents were

TABLE 1
Percentage Composition of Experimental Diets

Group diet	*Vit. mix*[a]	*Choline chloride*	*Salt mix*[b]	*Oil*	*Casein*	*Defatted fenugreek flour*	*Whole fenugreek flour*	*Starch*	*Protein contribution (g) to the diet by*	
									Casein	*Fenugreek*
I. Protein-free	1	1	4	10	—	—	—	84	—	—
II. Casein	1	1	4	10	13	—	—	72	10	—
III. Casein + 5% DFF[c]	1	1	4	10	10·8	5	—	68	8·55	1·45
IV. Casein + 10% DFF	1	1	4	10	9·2	10	—	65	7·1	2·9
V. Casein + 20% DFF	1	1	4	10	5·5	20	—	58	4·2	5·8
VI. 35% DFF (cooked)	1	1	4	10	—	35	—	49	—	10
VII. 35% DFF (raw)	1	1	4	10	—	35	—	49	—	10
VIII. 40% whole fenugreek (cooked)	1	1	4	8[a]	—	—	40	46	—	10
IX. 40% whole fenugreek (raw)	1	1	4	8[a]	—	—	40	46	—	10

[a] Vitamin mixture by Campbell (1963).
[b] Salt mixture according to USP XVII.
[c] DFF = defatted fenugreek flour.

determined by the techniques described earlier (Sharma, 1986). Amino acids (lysine, methionine and tryptophan) and vitamins (nicotinic acid and riboflavin) were determined microbiologically.

Biological evaluation

Fifty-four, 21 day-old, male Wistar rats of the Institute's rat colony were used. They were divided into nine groups of six animals each. Group I was kept on a protein-free diet. Group II served as control and received a casein diet which had the following composition (w/w%): casein, 13; starch, 71; groundnut oil, 10; vitamin mixture, 1; salt mixture, 4; choline chloride 1. In diets III–IX, a part of the diet was substituted at various levels with fenugreek seeds (whole or extracted) as shown in Table 1. The substitution was made at the expense of casein and starch so as to keep the protein level at 10%. But the calorie density of diets was different because of the high amounts of unavailable carbohydrates present in fenugreek seeds. Diets were mixed with twice the amount of water and cooked by steaming for 10 min.

Rats were housed individually in screen-bottomed cages and fed respective diets each day. Animals were weighed once a week. Records of weight gain and food intake were kept for 4 weeks. Faeces were collected during the last 4 days of the experiment. Nitrogen content was determined in faeces by the Kjeldahl method. From these data, the dry matter and protein digestibilities were calculated (Campbell, 1963). Animals were sacrificed at the termination of the experiment and eviscerated carcasses were hydrolysed in 6N HCl. Their nitrogen content was determined and net protein utilization (NPU) was calculated (Miller, 1963).

The results were statistically analysed using analysis of variance.

RESULTS AND DISCUSSION

The chemical composition of fenugreek seed is given in Table 2. The concentration of the major components, minerals and vitamins of fenugreek seeds falls within the range of values reported for other varieties grown in other parts of the world (Baccou *et al.*, 1978; Rafike & Laila, 1985). Chemical analysis indicates that the seeds are a rich source of protein, unavailable carbohydrate, mucilages and saponins. Except for fat and saponin, whole as well as extracted seeds contain almost equal amounts of amino acids, minerals and vitamins.

The contents of protein and lysine in fenugreek seeds are comparable with those in most of the commonly used legumes (Gopalan *et al.*, 1978).

TABLE 2
Analytical Data for Whole and Defatted Fenugreek Seeds

Component	*Whole fenugreek*	*Defatted fenugreek*
Moisture	2·4 g/100 g	12·1 g/100 g
Protein	25·4 g/100 g	28·9 g/100 g
Fat	7·9 g/100 g	1·1 g/100 g
Carbohydrate	1·6 g/100 g	2·1 g/100 g
Saponins	4·8 g/100 g	—
Total fibre	48·0 g/100 g	51·7 g/100 g
Gum	20·0 g/100 g	19·2 g/100 g
NDF	28·0 g/100 g	32·5 g/100 g
Ash	3·9 g/100 g	4·1 g/100 g
Fe	12·6 mg/100 g	14·3 mg/100 g
P	368 mg/100 g	381 mg/100 g
Ca	70·2 mg/100 g	69·4 mg/100 g
Zn	6·9 mg/100 g	7·0 mg/100 g
Mn	1·1 mg/100 g	1·2 mg/100 g
Cu	1·8 mg/100 g	1·7 mg/100 g
Mg	160 mg/100 g	153 mg/100 g
Cr	0·1 mg/100 g	0·1 mg/100 g
Nicotinic acid	1·5 mg/100 g	1·3 mg/100 g
Riboflavin	0·4 mg/100 g	0·3 mg/100 g
Methionine	1·2 g/16 g N	1·5 g/16 g N
Tryptophan	1·3 g/16 g N	1·5 g/16 g N
Lysine	5·7 g/16 g N	6·1 g/16 g N

Mean of three determinations carried out in duplicate.

Like other legumes, the protein is deficient in methionine. The amino acid score calculated is 68, which is reasonably good (FAO, 1968). The seed contains less starch but higher proportions of minerals (Ca, P, Fe, Zn and Mn) compared with other grain legumes (Sankara Rao & Deosthale, 1981).

It is interesting to note that all the parameters of protein quality (PER, dry matter digestibility and net protein utilization) were unaffected by inclusion of fenugreek in casein diets up to the 10% level (Table 3). Increasing the fenugreek level further, however, resulted in a significant reduction in the nutritional quality. The protein digestibility of fenugreek is low, due to gum in the seeds.

Table 4 indicates that the food intake of animals receiving fenugreek seeds alone is significantly lower ($P < 0{\cdot}05$) than that of control animals. Probably large amounts of fibre and mucilaginous material in the fenugreek seeds make these diets unpalatable. A similar difficulty was encountered while incorporating guar gum into palatable food products, because of its high viscosity. Unlike our previous study on okra seeds (Udayasekhara Rao, 1985) the rats in this study were unable to meet their

TABLE 3
Effect of Supplementation of Fenugreek Seeds on Nutritive Quality of Casein Diet

	Dietary groups			
	II *Casein*	*III* *Casein + 5% FG*	*IV* *Casein + 10% FG*	*V* *Casein + 20% FG*
Food intake (g/4 weeks)	238* ± 15·1[a]	228 ± 7·2[a]	236 ± 8·3[a]	219 ± 6·7[a]
Gain in body weight (g/4 weeks)	89 ± 7·3[a]	79 ± 4·6[b]	79 ± 3·1[b]	60 ± 2·1[c]
PER	3·48 ± 0·098[a]	3·37 ± 0·127[a]	3·25 ± 0·054[a]	2·58 ± 0·044[b]
Corrected PER	2·5	2·4	2·3	1·9
Dry matter digestibility (%)	92 ± 1·0[a]	93 ± 0·5[a]	90 ± 0·7[ab]	88 ± 1·0[b]
Protein digestibility (%)	89 ± 0·6[a]	84 ± 1·5[b]	82 ± 0·7[b]	73 ± 1·0[c]
NPU	69 ± 1·2[a]	66 ± 2·2[a]	64 ± 1·0[a]	52 ± 1·0[b]

* Mean ± SE.
Values with same superscript are not statistically different.

energy requirement by increasing the intake of fenugreek diet—a diet with high fibre and low calorie value (the calorific value of diets VI–IX was 30–33% less than that of the casein diet). Although okra seeds, like fenugreek seeds, are rich in fibre, they probably differ in the make-up of the fibre. The extracted seeds of fenugreek promoted better growth and thus had better PER and NPU values than the whole seeds. This may be due to the reduction in bitterness of the seeds by ether and alcohol extraction, which removes much of the lipids and saponins. Cooking the diets resulted in a slight variation in PER and NPU values of raw (whole) seeds. These seeds contain about 120 units of trypsin inhibitor activity which seems to be heat-labile. Extracted seed showed very little trypsin inhibitory activity. Contrary to our results, Sauvaire & Baccou (1976) reported a negative PER for raw fenugreek seeds. The composition of their diets did not differ much from those used in this study. The NPU values, however, agree well with those reported by Elmadfa & Kuhl (1976).

The results obtained *in vivo* on rats also allow a comparison with other legumes. Table 5 gives the biological value, protein digestibility and corrected PER values of other commonly used legumes. The results show that the nutritional quality of extracted seeds is comparable to that of other legumes (FAO, 1968). In India, fenugreek seeds are used as a condiment. The extraction of seeds allows complete elimination of the characteristic flavour of fenugreek. It is preferable to eliminate the odour by solvent

TABLE 4
Nutritive Quality of Fenugreek Seeds

	Groups				
	II *Casein*	*VI* *Defatted fenugreek cooked* (35%)	*VII* *Defatted fenugreek raw* (35%)	*VIII* *Whole fenugreek cooked* (40%)	*IX* *Whole fenugreek raw* (40%)
Food intake (g/4 weeks)	238 * ± 15·1[a]	174 ± 6·3[b]	184 ± 1·9[b]	146 ± 11·8[b]	137 ± 7·7[b]
Gain in body weight (g/4 weeks)	89 ± 7·3[a]	33 ± 2·0[b]	37 ± 1·5[b]	22 ± 3·7[c]	16 ± 3·0[d]
PER	3·48 ± 0·098[a]	1·86 ± 0·054[b]	2·0 ± 0·072[b]	1·39 ± 0·182[c]	1·09 ± 0·167[c]
Corrected PER	2·5	1·3	1·4	1·0	0·8
Dry matter digestibility %	92 ± 1·0[a]	80 ± 1·1[b]	83 ± 1·0[b]	80 ± 1·2[b]	83 ± 1·1[b]
Protein digestibility (%)	89 ± 0·6[a]	57 ± 2·0[b]	60 ± 2·3[b]	65 ± 3·0[c]	65 ± 0·8[c]
NPU	69 ± 1·2[a]	41 ± 1·8[b]	40 ± 1·7[b]	30 ± 2·6[c]	34 ± 2·4[c]

* Mean ± SE.
Values with same superscript are not statistically different.

TABLE 5
Protein Qualities of Various Legumes

	Biological value	*Digestibility*	*PER*
Chick pea (2)	68	85	1·7
Red gram (2)	57	77·7	1·54
Green gram (2)	58	73·5	1·48
Black gram (2)	62	85·0	1·45
Lathyrus (2)	46	90·0	—ve
Fenugreek (extd)	75	60·0	1·44

Legumes were tested at 10% protein level in 22-day-old animals (6–12) in a 4-week experiment.

extraction if it is proposed to use fenugreek seeds in large amounts in the human diet for therapeutic purposes.

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