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Extraction Efficacy and Composition of Biochemical Metabolites in Recent Fenugreek Varieties Grown in North India

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Abstract: Field experiments involving fenugreek (*Trigonella foenum-graecum*) crop varieties 'RMP-664' and 'AFG₃' raised for different stage cuttings revealed that fenugreek leaves have potential extractability of nitrogen. The highest total nitrogen extractability, from the leaves of three stage cuts of fenugreek varieties, were given by first cutting harvest, the values being 55.50% (RMP-664) and 57.51% (AFG₃). AFG₃ variety was found to be superior over RMP-664 variety of nitrogen-extractability. AFG₃ variety showed higher value of dry Leaf Protein Concentrate (LPC) recovery (2.64g per 100g fresh leaves) than RMP-664 variety (2.35g per 100g fresh leaves). Furthermore, dry LPC yield per hectare was higher in AFG₃ variety than RMP-664, the values being 195.03 kg/ha and 98.73 kg/ha, respectively. Extracted LPC and fibrous residue were found rich in biochemical metabolites like protein, ether extracts, ash and total carbohydrates. The total extracted protein yield from RMP-664 and AFG₃ varieties were obtained to be 42.44 kg/ha and 90.75 kg/ha, respectively.

Keywords: Leaf Protein Concentrate, Extraction efficacy, Biochemical metabolites, Fenugreek.

I. INTRODUCTION

Fenugreek (*Trigonella foenum-graecum*) or locally known as *methi*, one of the oldest known spice crop, is a nutritious green leafy vegetable. Fenugreek is raised in winter season but is consumed all along the year due to favourable environmental condition of Tarai region area, during Kharif and Rabi seasons in India. It has been reported that fresh leaves Fenugreek are rich in particularly protein, iron and carotene content of around 4.9g, 16.9mg and 234µg, per 100g respectively (1, 2 and 12). Green leaves when available in excess could be preserved by preparing nutritionally rich leaf protein concentrate (LPC). However, leaves from Fenugreek varieties have been reported to vary in composition (2,10).

Fenugreek also occupies an important medicinal value among its peer green roughages. Every part of fenugreek is beneficial for human health but seeds are particularly used as spices and condiments. Apart from India, usage of Fenugreek in developed countries such as US, Canada etc in preparation of chutneys /sauces, spice blends, maple syrups, bean soups as well as beef stew are well known today. It is also added to sweets given to ladies after their post natal period.

India today, one of the significant exporter of spices to the world, can overtake others and can comment its leadership particularly in case of Fenugreek. There seems to be no systematic and serious efforts being made to develop and standardization of its production technology for its seeds and the relevant aromatic products.

Fenugreek crop to being harvested a couple of times in order to achieve optimization of plant nutrients which is of utmost importance to enhance the productivity at all stages of cutting (11).

Therefore, it was thought worthwhile to study the variability in the extractability and composition of LPC from some recent and widely used Fenugreek varieties in North Indian farmlands particularly in the state of Uttar Pradesh.

II. MATERIALS AND METHODS

A field experiment was conducted, with two recent fenugreek varieties (RMP-664 and AFG₃) chosen, in factorial randomised complete block design plot size (8mx5m) in replicate at the Vegetable Farm centre of the village Nizampur, District Budaun situated 8km from Gangetic plains.

Nitrogen (as urea), phosphorus (as single superphosphate) and potassium (as muriate of potash) were applied as full basal dose of 90kg/ha, 60kg/ha and 50kg/ha respectively at the time of sowing. The pots were kept free from weeds by hand weeding and

irrigation was given soon after sowing, twice a week for first four weeks and subsequently at weekly intervals to maintain optimum moisture. Green leaves were harvested at every 15 days' interval. The first cutting was taken after two months of plant growth.

A. Extractability Procedures

For extractability studies, the green leaves from different replications as well as all three cuttings were collected separately. The green leaves collected in the morning were brought to the laboratory in polythene bags, numbered properly, and were then processed immediately. The vegetation was thoroughly washed with water to remove the adhering dust etc. prior to processing for extraction. The extraction procedure as given by Pirie (3) and AOAC (1985) was followed (4). 500g of green leaves were minced in green vegetation mincer and the resultant was squeezed by using a long cloth. The extractant was heated to 80°C to coagulate protein which was centrifuged, washed with distilled water and finally with dilute HCl (pH 4.0). The coagulum was dried in an oven at 70°C for 24 hours and weighed. The per hectare yield of dried LPC was calculated on the basis of total yield of green leaves and the recovery of dry LPC.

B. Biochemical Composition of LPC and Fibrous Residue

LPCs prepared by coagulating and drying the coagulum as well as residue from green leaves of fenugreek were analysed for various biochemical metabolites. Nitrogen in the LPC and residue was determined by conventional micro-Kjeldahl method. Protein was determined by multiplying nitrogen values with factor 6.25. Ether extractable lipid content in LPC and residue was determined by the Soxhlet extraction procedure using petroleum ether (4). Ash content was determined by incineration of LPC and residue in the muffle furnace at 550°C till constant weight was obtained (4). Total carbohydrate content was calculated by difference method.

III. RESULT AND DISCUSSION

The results of the investigation have been discussed hereunder and various data have been presented in relevant tables below: -

A. Foliage Yield

Data on foliage yield from all the three cuts are given in Table 1.

Table 1. Foliage yield (q/ha) of Fenugreek varieties at different cuttings.

Cutting stage	Varieties	
	RMP-664	AFG ₃
First	10.43	18.62
Second	15.68	25.72
Third	19.40	31.21
Total	45.50	75.55

	Variety(V)	Cutting(C)	Interaction (VxC)
SE ₊	0.041	0.50	0.22
CD at 5%	0.087	0.11	0.48

Fenugreek crop gave a total foliage yield of 45.50 q/ha from RMP-664 variety and 75.55 q/ha from AFG₃ variety including all the three stage cuts. As regards the performance of these two varieties at the three cuts, the third cut showed highest foliage yields.

B. Nitrogen Content in the Dry Matter of Pulp

Table 2 shows that the first cutting from both RMP-664 and AFG₃ varieties had highest nitrogen content in the dry matter of pulp and successive second and third cutting showed values in descending order. AFG₃ variety was found to be superior to RMP-664 at all the three cuttings in respect of nitrogen content in the dry matter of pulp.

C. Total Nitrogen Extractability

The first cutting of both RMP-664 and AFG₃ varieties gave maximum nitrogen extractability in fresh leaves with values of 55.50 and 57.51%, respectively (Table 2).

D. LPC Recovery

The best recovery of LPC 2.35g/100g of fresh leaves in RMP-664 and 2.64g/100g in AFG₃ was noticed at the first cutting. The recovery of LPC was lowest at the third harvest. (Table 2) AFG₃ variety was found superior to RMP-664 variety of fenugreek in respect of LPC recovery. Mehrotra et al. (5) found dry LPC recovery of 2.02g/100g fresh leaves from Patharchata weed. Srivastava et al. (6) studied leaf protein recovery from the haulms of eight potato varieties and found LPC recovery from 2.54 to 2.92g/100g of fresh haulms.

Table 2. Nitrogen content in the dry matter pulp (%), Nitrogen extractability (%) and LPC recovery from the leaves of Fenugreek varieties at different stage of cuttings.

Variety	No. of cutting	N content in the dry matter Pulp (%)	N-extract ability (%) fresh leaves	Dry LPC recovery (g/100g)
RMP-664	First	3.84	55.50	2.35
	Second	3.73	52.52	2.14
	Third	3.68	51.39	2.11
AFG ₃	First	3.94	57.51	2.64
	Second	3.83	54.89	2.62
	Third	3.77	53.62	2.52

E. LPC Yield

Third cutting yielded highest LPC in both varieties, the values being 40.87 kg/ha in RMP-664 and 78.85 kg/ha in AFG₃ variety followed by second and first cutting (Table 3). Total LPC yield from the three cuts of 98.73 and 195.03 kg/ha were obtained in RMP-664 and AFG₃ varieties respectively.

F. Protein Yield

Per hectare protein yield was calculated on the basis of protein content of LPC and per hectare foliage yield. Total protein yield from the foliage of all the three cuts were obtained as 42.44 and 90.95kg/ha from RMP-664 and AFG₃ varieties respectively.

Table 3. Yield of extracted LPC and extracted protein (Kg/ha) from Fenugreek varieties at different cuttings.

Variety	No. of cutting	Average yield of LPC	Protein yield
RMP-664	First	23.61	9.29
	Second	34.25	14.39
	Third	40.87	18.76
	Total	98.73	42.44
AFG ₃	First	49.21	21.95
	Second	60.98	31.06
	Third	78.84	37.94
	Total	195.03	90.95

	Variety (V)	Cutting (C)	Interaction (VxC)
CD at 5%	0.15	1.79	2.53

CD at 5%	0.43	0.53	0.75
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G. Fresh and Dry Fibrous Residue Yield

Data on fibrous residue yield showed that highest yield of both type i.e. fresh and dry was given at third cutting whereas the first cut yielded the lowest value (Table 4).

Table 4. Fresh and dry fibrous residue yield (q/ha) from Fenugreek varieties at different cuttings.

Variety	No. of cutting	Average yield	
		Fresh fibrous residue	Dry fibrous residue
RMP-664	First	3.95	1.14
	Second	6.06	1.74
	Third	7.62	2.17
	Total	17.63	5.05
AFG ₃	First	6.66	1.73
	Second	9.44	2.44
	Third	11.64	3.03
	Total	27.74	7.20

	Variety (V)	Cutting (C)	Interaction (VxC)
CD at 5% (Fresh residue)	0.04	0.05	0.07
CD at 5% (Dry residue)	0.03	0.03	0.05

H. X- Composition of Biochemical metabolites of LPC

- 1) (X₁) **Protein Content:** Protein content in LPC varied from 39.37 (first cutting) to 45.94% (third cutting) in RMP-664 variety and 49.19 (first cutting) to 48.13% (third cutting) in AFG₃ variety of fenugreek. AFG₃ appeared to be superior to RMP-664 in respect of protein content of LPC (Table 5).
- 2) (X₂) **Ether Extracts:** The ether extracts in LPC varied from 9.65 (third cut) to 11.25% (first cut) in RMP-664 variety and 8.13 (third cut) to 9.38% (first cut) in AFG₃ variety. Pirie (3), Maliwal (7) and Byers (8) have reported variability of ether extracts percentage in the LPCs. Carson et al. (9) also extracted LPCs from the tropical Napier fodder crop and reported an escalation of ether extract from 2.8 to 16.3%.
- 3) (X₃) **Ash Content:** As regards ash content in LPC, it varied from 5.72 to 6.31% in RMP-664 and 5.16 to 5.61% in AFG₃ variety of fenugreek and the lowest and highest values were given by third and first cuttings, respectively. Wide variability in ash content of LPCs has been observed by several workers (5, 7).
- 4) (X₄) **Carbohydrates:** The carbohydrate content in the LPCs was found to vary from 38.7 (third cutting) to 43.06% (first cutting) in RMP-664 variety and 38.58 to 40.82% of the same cutting in AFG₃ variety of fenugreek. Mehrotra et al. (5) prepared LPC from 'Patharchata' (*Trianthema honogyne*) which contained total carbohydrate content of 37.05%.

I. Y- Composition of Biochemical metabolites of dry Residue

- 1) (Y₁) **Protein content:** Fibre protein content in RMP-664 variety of fenugreek varied from 10.67 to 11.20% and in AFG₃ variety from 10.47 to 10.89%, the lowest and highest values in the two varieties were given by first and third cut, (13) respectively (Table 6).
- 2) (Y₂) **Ether extracts:** The ether extracts in residue ranged from 6.23 (third cut) to 7.55% (first cut) and 5.83 (third cut) to 7.20% (first cut) in RMP-664 and AFG₃ varieties respectively.
- 3) (Y₃) **Ash content:** Data in respect of ash content of dried residue varied from 8.23 to 10.46% in RMP-664 and 7.29 to 9.25% in AFG₃ variety, the lowest and highest values were given by first and third cut, respectively (14).

- 4) (Y_4) *Carbohydrate Content*: The percentage of carbohydrate ranged from 72.11 and 74.03 (lowest) to 73.55 and 75.04 % (highest) in RMP-664 and AFG₃ varieties respectively which were drawn from by third and first cut respectively (14).

IV. CONCLUSION

The significance of the Fenugreek among Indians leads to a direct health co-relation. So hunting is continuously on, among agricultural practices, with involvement of latest procedures and for best as well as most economic varieties of Fenugreek with a nutritional perspective. Out of the two chosen varieties AFG₃ and RMP-664 the former stands more superior to latter, with respect to physiological aspects such as: Foliage yield, nitrogen content in the dry matter of pulp, extractable total nitrogen, recovered Leaf Protein Concentrate, yield of LPC and protein as well as fresh and dry fibrous residual yield. In case of biochemical metabolites composition in both LPC and dry residues, the variety RMP-664 showed both ether extracts and total ash contents higher than AFG₃ whereas in regards to total carbohydrate content AFG₃ stood better than RMP-664. The protein content was more in AFG₃ for LPC studies whereas it was found less in dry residue studies in comparison to RMP-664.

Table 5 : Biochemical composition of LPC from Fenugreek varieties at different cuttings.

Variety	Cutting stage	Protein (%)	Ether Extract (%)	Ash (%)	Carbohydrate (%)
RMP-664	First	39.37	11.25	6.31	43.06
	Second	42.00	10.13	6.03	41.85
	Third	45.94	9.65	5.72	38.70
	Average	42.47	10.34	6.02	41.23
AFG ₃	First	44.19	9.38	5.61	40.82
	Second	46.38	8.50	5.43	39.70
	Third	48.13	8.13	5.16	38.58
	Average	46.23	8.67	5.33	39.70

Table 6 : Biochemical composition of dry Residue from Fenugreek varieties at different cutting.

Variety	Cutting stage	Protein (%)	Ether Extract (%)	Ash (%)	Carbohydrate (%)
RMP-664	First	10.67	7.55	8.23	73.55
	Second	11.06	6.80	9.33	72.81
	Third	11.20	6.23	10.46	72.11
	Average	10.97	6.86	9.34	72.82
AFG ₃	First	10.47	7.20	7.29	75.04
	Second	10.78	6.33	8.40	74.49
	Third	10.89	5.83	9.25	74.03
	Average	10.71	6.45	8.31	74.52

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