

Changes in some nutrients of fenugreek sprouts in response to natural food grade additives

ANWAR HUSSAIN, IMTIYAZ MURATAZA AND SHABIR HUSSAIN KHAN

SUMMARY : The paper focuses on the changes in some of the nutritive components of fenugreek varieties (Methi Local and Methi Shalimar Improved) due to sprouting and the effect of natural elicitors in enhancing these nutrients. The total sugar content increased from 6.3 per cent to 12.88 per cent, where as the reducing sugar increased from 0.72 per cent to 6.5 per cent in case of Methi Shalimar Improved after 5 days of germination. Chitosan (1500 ppm) was the most effective elicitor in increasing these parameters. Little change was observed in non reducing sugar content during sprouting. Calcium increased from 141 to 399.57 mg100g⁻¹ in case of Methi Shalimar Improved after 8 days of germination. Sprouting for 8 days also resulted in gain in iron content *i.e.*, from 12.5 to 15.56 mg100g⁻¹ in the cultivar Methi Local with a simultaneous increase in magnesium content from 73.14 to 158.86 mg100g⁻¹ in case of Methi Shalimar Improved. Folic acid (100 μ M) resulted in maximum increase in the calcium and iron contents and chitosan (1500 ppm) resulted in maximum increase in magnesium content of the fenugreek sprouts as compared to the other treatments used. It is thus concluded that, the cultivar Methi Shalimar Improved is better as compared to the another variety regarding the change in most of the essential nutrients and chitosan 1500 ppm and folic acid 100 μ M are the promising candidates among the elicitors used.

KEY WORDS : Fenugreek, Sprouts, Elicitors, Total sugar, Iron, Chitosan

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Renugreek (*Trigonella foenum-graecum* L.) is a leguminous herb cultivated in India and North African countries. It belongs to the family *Fabaceae* and is variously called in different languages, *viz.*, *Greek hay* (English), Fenugrec (French), *Methi* (Hindi), *Bockshorklee* (German), *Fienogreco* (Italian), *Pazhitnik* (Russian), *Alholva* (Spanish), *Koroha* (Japanese), *Hulba* (Arabian), *Halba* (Malaya), and *K'u-Tou* (Chinese). The seeds are used as spices

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SHABIR HUSSAIN KHAN, Division of Olericulture, Sher-e-Kashmir University of Agricultural Sciences and Technology (K), SRINAGAR (J & K) INDIA worldwide, whereas the leaves are used as green leafy vegetables in the diet. Fenugreek seeds are bitter to taste and are known for a long time for their medicinal qualities. Fenugreek seeds have been in use for over 2500 years. India is the major producer of fenugreek and its main consumer for culinary and medicinal uses. The seeds of fenugreek are used as a spice for seasoning, a flavouring agent and in comparatively larger quantities in making soups and pan cakes. Fenugreek emerges from food ranking system as an excellent source of essential nutrients, including vitamin A, vitamin C, and vitamin B_{12} . It is also a very good source of dietary fiber, calcium, iron and magnesium. This combination of vitamins, minerals, and phytonutrients makes fenugreek a health superstar. Fenugreek being rich in phytochemical has traditionally been used as a food, forage and medicinal plant. Fenugreek seeds contain lysine and L-tryptophan rich proteins, mucilaginous fibre and other rare chemical constituents such as saponins, comumarin, fenugreekine, nicotinic acid, sapogenins, phytic acid, scopoletin and trigonelline, which are thought to account for many of its presumed therapeutic effects.

Although a dried seed is characterized by remarkably low metabolic rate, just sprouting of seed can triggers tremendous and complex changes which consist of three main types: the breakdown of complex fats, starch conversion into simple sugars and breakdown of protein into amino acids and also transport of materials from the breakdown products formed to the targeted areas. Vitamins, including A, B-complex (B-12), C, E and K are increased, essential minerals such as calcium, magnesium, iron and zinc are supplied in organic form "chelated" for better assimilation and nutrient density is enhanced at the expense of calories. Sprouting also removes some anti-nutrients such as enzyme inhibitors in the seed that make sprout safe for the diet.

The nutritional value of the sprouts was discovered by the Chinese thousands of years ago. Recently, in USA, numerous scientific studies suggest the importance of sprouts in a healthy diet. Although the use of sprouts as a food source for man is old as man's use of seeds, it is only in recent times that science has begun to unravel the chemistry of sprouting seed and its potential significance in both human and animal nutrition. Each sprout may contain as many phytochemicals as an entire plant, *i.e.* when we eat a sprout, we are eating the entire plant at a very young age, *i.e.*, we eat the root, stem, and head. Sprouting in fenugreek is known to improve its essential nutrient contents and also reduce the phytic, tannic and trypsin inhibitors (El-Shimi *et al.*, 1984 and Mansour *et al.*, 1994).

Current investigation suggests that exogenously applied natural elicitors which would be food grade in quality can stimulate endogenous essential nutrients of the fenugreek sprouts. These elicitors used were vitamin C, folic acid and chitosan. Both vitamin C and folic acid are water soluble vitamins and chitosan is a natural polymer, appear to be promising candidates regarding the change in nutrient content in fenugreek sprouts. The parameters measured to characterize the effect of these elicitors were total sugar, reducing and non reducing sugars, calcium, iron and magnesium contents.

EXPERIMENTAL METHODS

The present investigation was carried out in the Biochemistry laboratory of the Division of Post Harvest Technology, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, J & K, India during the year 2010-11.

Seed treatment and sprouting :

Seeds of the fenugreek cultivars *i.e.*, 'Methi Local' and 'Methi Shalimar Improved' were provided by the Division of Olericulture, SKUAST-K. The elicitors used in the study were folic acid (Titan Biotech Ltd.), vitamin C (S.D. fine-chem Ltd.) and chitosan (HiMedia Laboratories Pvt. Ltd.) as pretreatments. Dry seeds of the selected cultivars were soaked in conical flasks containing different pretreatments *viz.*, distilled water (control), vitamin C (100 μ M), vitamin C (500 μ M), folic acid (50 μ M), folic acid (100 μ M), chitosan (1000 ppm) and chitosan (1500 ppm). The flasks were then placed on a shaker at a speed of 120 rpm at room temperature for 18 hours. The pre-soaked seeds were washed in distilled water and germinated in glass jars. The germinating seeds were kept moist with distilled water and sprouts were raised for 10 days. Sprouts were collected on 0, 2nd, 5th, 8th and 10th day of sprouting for analysis.

Analysis of total sugars :

The amount of total soluble sugars was estimated by phenol sulphuric acid reagent method given by Dubois *et al.* (1951). 500 mg each of dried sample was homogenized in 10 ml of 80 per cent ethanol. Each sample was centrifuged at 2000 rpm for 20 min. The supernatant was collected separately. To 1 ml of alcoholic extract, 1 ml of 5 per cent phenol solution was added and mixed. Then 5.0 ml of 96 per cent sulphuric acid was added rapidly. Each tube was gently agitated during the addition of the acid and then allowed to stand in a water bath at 26-30°C for 20 minutes. The OD of the characteristic yellow orange colour thus, developed was measured at 490 nm in a spectrophotometer after setting for 100 per cent transmission against the blank standard curve prepared by using known concentration of glucose. The quantity of sugar was expressed as per cent of fresh weight of tissue.

Analysis of reducing sugars :

The estimation of reducing sugar was done according to the protocol given by Nelson and Somogy (1952).

Reagents:

Alkaline copper tartarate :

Solution (i): 2.5 g anhydrous sodium carbonate, 2 g sodium bicarbonate, 2.5 g potassium sodium tartarate and 20 g anhydrous sodium sulphate was dissolved in 80 ml water and volume was made up to 100 ml.

Solution (ii): 15 g copper sulphate was dissolved in a small volume of distilled water. To this one drop of sulphuric acid was added and total volume was made up to 100 ml.

 $4\,ml$ of solution (ii) and 96 ml solution (i) was mixed before use.

Arsenomolybdate reagent :

This reagent was prepared by dissolving 2.5 g ammonium molybdate in 46 ml water. 2.5 ml sulphuric acid was added and mixed well and then 0.3 g disodium hydrogen arsenate dissolved in 25 ml water was added followed by incubation for 24 to 48 hours at 37 $^{\circ}$ C.

Standard stock glucose solution :

100 mg of glucose was dissolved in 100 ml distilled water.

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Working standard :

Working standard was made by diluting 10 ml of standard stock glucose solution in 100 ml distilled water (100 μ gml⁻¹).

Procedure :

100 mg of sample was taken and the sugars were extracted with hot 80 per cent alcohol twice (5 ml each). The supernatant was collected and evaporated on water bath followed by the addition of 10 ml water to dissolve the sugars. Aliquot of 0.2 ml of alcohol free extracts was pipetted out to separate test tubes. 0.2 ml, 0.4 ml, 0.6 ml, 0.8 ml and 1 ml of working standard solution were taken into a series of test tubes. The volume in both sample and standard tubes was made up to 2 ml with distilled water. A blank was also made in a separate tube with 2 ml of distilled water. 1 ml of alkaline copper tartarate reagent was added to each tube. The tubes were then placed in boiling water for 10 minutes after which these were cooled and 1 ml of arsenomolybdic acid reagent was added. The volume in each tube was made up to 10 ml with water and absorbance of blue colour was read at 620 nm after 10 minutes. From the graph drawn, the amount of reducing sugar present in the sample was calculated.

Reducing sugar (%) =
$$\frac{x}{0.2}$$
 x 10 mg of glucose

where, x is the graph value corresponds to 0.2 ml of aliquot taken.

Analysis of non-reducing sugars :

Non reducing sugar was obtained by subtracting reducing sugar from the total sugar obtained.

Determination of mineral contents (Ca, Fe and Mg):

The digestion of the above minerals was done by dry ashing. The procedure for mineral analysis followed was that of Chapman and Pratt (1961) with slight modifications. 0.5 g of the ground sample was taken in 50 ml porcelain crucibles. The crucibles were then placed into a cool muffle furnace and temperature was increased gradually to 550°C. Continuous ashing was done for 5 hours after attaining 550°C. After that the muffle furnace was shut off and the door was opened cautiously for rapid cooling. The crucibles were taken out carefully after cooling. The cooled ash was dissolved in 5 ml 2N HCL and mixed with a glass rod. 2N HCL was made by diluting 165.6 ml conc. HCL (37 %, sp.gr.1.19) in distilled water and final volume was brought up to 1 litre volume with distilled water. After 15-20 minutes, the volume was made up to 50 ml using distilled water. Thorough mixing of the solution was followed by allowing the solution to stand for about 30 minutes. A blank containing the same contents was also maintained. The supernatant was used for analyzing the minerals with the help of atomic absorption spectrophotometer.

Statistical analysis of data :

Data obtained were subjected to statistical analysis following the CRD model and the variation among the treatment means was tested for significance of analysis of variance technique described by Gomez and Gomez (1984). Levels of significance used for T-test was p=0.05 from the table given by Fisher (1970). The critical difference has also been worked out.

EXPERIMENTAL FINDINGS AND ANALYSIS

The results of the present study as well as relevant discussions have been presented under following heads:

Changes in total sugar content :

As per the data given in Table 1, it is depicted that an increase in the total sugar content was found up to 5th day followed by decrease on the subsequent days *i.e.*, 8th and 10th days of sprouting. The two selected cultivars of fenugreek viz., Methi Local and Methi Shalimar Improved contain total sugar of 12.83 per cent and 12.88 per cent, respectively on the peak day. The data also revealed that all the food grade elicitors used in the current study resulted in significant change in the total sugar content in fenugreek sprouts with highest effect on 5th day. Chitosan (1500 ppm) showed maximum effect on all the days as compared to other pretreatments. Its value was 13.18 per cent on this day. Whereas the minimum influence was shown by water treatment (control) on the same day and its value thus observed was 12.6 per cent. Increase in total sugar content on sprouting was also reported by Rafik El-Mahdy and Laila El-Sebaiy (1983) in fenugreek and Ghazali and Cheng (1990) in black gram. A marked increase in total sugar during early stages may be attributed to the conversion of starch into total sugar and decrease in the later stages may be due to consumption of this sugar during sprouting (Doblado et al., 2007).

Table 1 : Effect of chemical treatments on total sugar (%) of fenugreek sprouts

	Total sugar (%)				
Treatment	0	2 nd	5 th	8 th	10 th
	day	day	ay	day	day
Variety					
V ₁ (Methi Local)	6.22	8.47	12.83	10.82	8.30
V2 (Methi Shalimar Improved)	6.30	8.54	12.88	10.90	8.38
CD (p≤0.05)	NS	NS	NS	NS	NS
Chemical					
T ₀ (Control)	6.05	8.25	12.60	10.60	8.05
T_1 (Vitamin C (100 μ M)	6.15	8.45	12.75	10.80	8.20
T_2 (Vitamin C (500 μ M)	6.25	8.60	12.90	10.95	8.30
T_3 (Folic acid (50 μ M)	6.25	8.40	12.80	10.65	8.40
T4 (Folic acid (100 µM)	6.25	8.40	12.80	10.65	8.40
T ₅ (Chitosan (1000 ppm)	6.40	8.65	13.00	11.10	8.45
T ₆ (Chitosan (1500 ppm)	6.50	8.80	13.18	11.30	8.60
C.D. (p≤0.05)	0.17	0.18	0.19	0.16	0.20

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Changes in reducing sugar content :

Just like total sugar, a continuous increasing trend in reducing sugar content up to 5th day was observed followed by decline on the remaining stages of study (8th and 10th day) in samples of both the selected varieties of fenugreek (Table 2). Reducing sugar content was 5.22 per cent in case of Methi Local and 6.5 per cent FW in case of Methi Shalimar Improved on 5th day of sprouting. A non-significant influence of all the treatments on reducing content was observed on 0 day, however, on other days the reverse is true. Up to 5th day, there was a continuous increase in the said parameter followed by a continuous decline on the last two stages of observations. Chitosan (1500 ppm) resulted in highest content of the said nutrient of 6.75 per cent, whereas, samples treated with water (control) with a value of 4.51 per cent resulted in lowest concentration on 5th day. The change in the reducing sugar content during sprouting of the fenugreek seeds are in consonance with findings of Rafik El-Mahdy and Laila El-Sebaiy (1983) in fenugreek, Ghazali and Cheng (1990) in black gram and Ayernor and Ocloo (2007) in rice. The difference in reducing sugar between the two varieties during various stages of sprouting may be attributed to genetic frame work of cultivars. An increase in reducing sugar during early stages may be attributed to the conversion of starch into simple sugar with special reference to reducing sugar during sprouting (Doblado et al., 2007). The decreasing trend in reducing sugar content on the later stages may be due to its consumption during the sprouting process.

Table 2 : Effect of chemical fenugreek sprouts	treatme	ents on	reducing	g sugar	(%) of	
· · · · · · · · · · · · · · · · · · ·	Reducing sugar (%)					
Treatment	0	2^{nd}	5 th	8 th	10 th	
	day	day	day	day	day	
Variety						
V ₁ (Methi Local)	0.52	2.75	5.22	3.54	3.08	
V ₂ (Methi Shalimar Improved)	0.72	3.38	6.50	4.62	4.04	
C.D. (p?0.05)	0.10	0.11	0.11	0.12	0.10	
Chemical						
T ₀ (Control)	0.60	2.30	4.51	3.50	2.50	
T_1 (Vitamin C (100 μ M)	0.60	2.65	5.10	3.95	3.10	
T_2 (Vitamin C (500 μ M)	0.60	3.00	5.80	4.20	4.15	
T ₃ (Folic acid (50 µM)	0.60	3.10	6.20	4.00	3.65	
T_4 (Folic acid (100 μ M)	0.60	3.10	6.20	4.00	3.65	
T ₅ (Chitosan (1000 ppm)	0.70	3.25	6.50	4.30	3.85	
T ₆ (Chitosan (1500 ppm)	0.70	4.10	6.75	4.65	4.05	
C.D. (p <u><</u> 0.05)	NS	0.22	0.21	0.23	0.20	

Changes in non-reducing sugar content :

During sprouting, a continuous increase in the non reducing sugar content of sprouts of both the cultivars of

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fenugreek was observed and peaked on 5th day followed by decrease onwards up to 10th day (Table 3). Among the two cultivars studied, Methi Local exhibited the higher amount of this type of sugar on all the days of study as compared to the another variety Methi Shalimar Improved with highest contents on 5th day of 7.6 per cent and 6.38 per cent, respectively. Contradictory to the results obtained in reducing sugar, the control (water) gave the highest nutrient content on 5th day of 8.1 per cent and Chitosan 1500 ppm, gave the lowest content of 6.4 per cent on the very day. For all the elicitor concentrations, the non reducing sugar increased up to 8th day and declined on last day of investigation. These results regarding increase in this sugar are in accordance with those reported by Ayernor and Ocloo (2007) in rice, Rafik El-Mahdy and Laila El-Sebaiy (1983) in fenugreek and Ghazali and Cheng (1990) in black gram. The reported increase in non-reducing sugar during early stages may be attributed to the conversion of starch into this type of sugar and decrease in the later stages may be due to its consumption during sprouting (Doblado et al., 2007).

Table 3: Effect of chemical treatments on non reducing sugar (%) of fenugreek sprouts						
Treatment	Non reducing sugar (%)					
	0	2 nd	5 th	8 th	10 th	
	day	day	day	day	day	
Variety						
V ₁ (Methi Local)	5.70	5.71	7.60	7.28	5.21	
V2 (Methi Shalimar Improved)	5.57	5.15	6.38	6.27	4.34	
C.D. (p≤0.05)	0.09	0.11	0.10	0.11	0.11	
Chemical						
T ₀ (Control)	5.45	5.95	8.10	7.10	5.55	
T_1 (Vitamin C (100 μ M)	5.55	5.80	7.65	6.85	5.10	
T_2 (Vitamin C (500 μ M)	5.65	5.60	7.10	6.75	4.55	
T_3 (Folic acid (50 μ M)	5.65	5.30	6.60	6.65	4.75	
T_4 (Folic acid (100 μ M)	5.65	5.30	6.60	6.65	4.75	
T ₅ (Chitosan (1000 ppm)	5.70	5.40	6.50	6.80	4.60	
T ₆ (Chitosan (1500 ppm)	5.80	4.70	6.40	6.65	4.55	
C.D. (p≤0.05)	0.18	0.21	0.18	0.21	0.22	

Changes in calcium content :

An overview of the data included in Table 4 revealed that significant differences were observed regarding the calcium content between the two varieties on all the stages of sprouting. The cultivar Methi Shalimar Improved was superior on all days as compared to Methi Local. Continuous increase in the amount of this mineral in both cultivars was observed with the passage of time up to 8th day (corresponding to 397.57 mg100g⁻¹ and 399.57 mg100g⁻¹ in Methi Local and Methi Shalimar Improved, respectively), however, on 10th day there was slight decline in the said property. Analysis of data further revealed that non-significant influence was shown by food grade additives on 0

day on calcium content whereas the various pre-treatments had varied effects on other days. Also, there was increasing effect of the treatments on the parameter which go highest on 8th day, after that there was a decline on 10th day. On the peak day, folic acid (100 µM) showed superiority regarding the calcium content corresponding to 403 mg100g⁻¹ and control resulted in minimum content corresponding to 396 mg100g⁻¹. This suggests that the calcium enriched sprouts can be used as a good source of calcium as functional food. The increase in calcium content during sprouting were also reported by Srinivasan (2006) in fenugreek, Mbithi-Mwikya et al. (2000) in finger millet and Sattar et al. (1985) in corn. The difference in hereditary construction between the two varieties might have resulted in higher calcium in Methi Shalimar Improved as compared to Methi Local. The increase in calcium content may be because the minerals chelate or merge with protein, in a way that increases their function (Shipard, 2005).

Table 4: Effect of chemical ta fenugreek sprouts	reatment	ts on cal	cium (m	g/100g I	FW) of		
	Calcium (mg/100g)						
Treatment	0	2 nd	5 th	8 th	10 th		
	day	day	day	day	day		
Variety							
V ₁ (Methi Local)	140.00	142.43	177.57	397.57	395.71		
V ₂ (Methi Shalimar Improved)	141.00	143.71	179.33	399.57	397.86		
C.D. (p≤0.05)	0.57	1.03	0.91	1.03	1.12		
Chemical							
T ₀ (Control)	140.50	141.50	176.50	396.00	394.00		
T_1 (Vitamin C (100 μ M)	140.50	141.50	177.00	396.50	395.00		
T_2 (Vitamin C (500 μ M)	140.50	143.00	178.50	398.50	397.00		
T ₃ (Folic acid (50 µM)	140.50	145.00	180.50	401.00	398.50		
T_4 (Folic acid (100 μ M)	140.50	146.00	182.00	403.00	402.00		
T ₅ (Chitosan (1000 ppm)	140.50	141.50	176.67	396.50	394.50		
T ₆ (Chitosan (1500 ppm)	140.50	143.00	178.00	398.50	396.50		
C.D. (p≤0.05)	NS	1.93	1.70	1.93	2.09		

Changes in iron content :

The iron content of the sprouts varied between the two varieties *viz*. Methi Local and Methi Shalimar Improved on all the selected days of sprouting, which is shown Table 5. There was a continuous increase in the said nutrient content while proceeding through the different stages of sprouting up to 8th day followed by a slight decline on last selected day. The cultivar Methi Local resulted in higher values on all the stages with 15.56 mg100g⁻¹ as compared to variety Methi Shalimar Improved which demonstrated 15.21 mg100g⁻¹ on 8th day. The food grade additives indicated non-significant differences with respect to their effect on iron content on 0 day whereas on all other days the reverse was true. In this case, the treatment folic acid (100 μ M) was found to be most effective pretreatment

with a value of 16.55 mg100g⁻¹ while control (14.75 mg100g⁻¹) was found to be least effective in changing the iron level in fenugreek sprouts on the peak day *i.e.*, 8th day. Similar findings have been reported by Srinivasan (2006) in fenugreek, Mbithi-Mwikya *et al.* (2000) in finger millet and Sattar *et al.* (1985) in corn. The difference in hereditary construction between the two varieties might have resulted in lesser iron concentration in Methi Shalimar Improved as compared to Methi Local. When seeds are sprouted, the minerals chelate or merge with protein, in a way that increases their function (Shipard, 2005).

Table 5: Effect of chemical fenugreek sprouts	treatme	nts on	iron (m	g/100g	FW) of
			n (mg/10	0g)	
Treatment	0	2^{nd}	5 th	8 th	10 th
	day	day	day	day	day
Variety					
V1 (Methi Local)	12.50	12.78	14.75	15.56	15.31
V2 (Methi Shalimar Improved)	12.00	12.54	14.48	15.21	15.04
C.D. (p <u><</u> 0.05)	0.05	0.08	0.18	0.35	0.17
Chemical					
T ₀ (Control)	12.25	12.50	14.20	14.75	14.55
T_1 (Vitamin C (100 μ M)	12.25	12.60	14.35	15.00	14.73
T_2 (Vitamin C (500 μ M)	12.25	12.75	14.50	15.26	15.06
T ₃ (Folic acid (50 µM)	12.25	12.75	15.15	16.40	16.15
T_4 (Folic acid (100 μ M)	12.25	12.90	15.45	16.55	16.40
T ₅ (Chitosan (1000 ppm)	12.20	12.55	14.25	14.76	14.55
T ₆ (Chitosan (1500 ppm)	12.20	12.70	14.45	15.00	14.80
C.D. (p≤0.05)	NS	0.15	0.34	0.67	0.32

Changes in magnesium content :

Examination of the data given in Table 6 revealed that, higher amount of magnesium content was observed in sprouts of Methi Shalimar Improved as compared to the Methi Local variety except on 10th day. A continuous increase in the magnesium concentration was observed with the passage of time in both the varieties up to 8th day. The values on this day were observed to be 157.86 mg100g-1 and 158.86 mg100g-1 in Methi Local and Methi Shalimar Improved, respectively. Samples treated with chitosan (1500 ppm) resulted in maximum and water treated (control) samples recorded the minimum magnesium content in fenugreek sprouts on all the selected days of study with highest values on 8th day. It was 160 mg100g-¹ in case of the former additive and 157 mg100g⁻¹ in case of the latter one. These results are in agreement with that of Srinivasan (2006) in fenugreek and Sattar et al. (1985) in corn. The difference in hereditary construction between the two varieties might have resulted in higher magnesium concentration in Methi Shalimar Improved as compared to Methi Local. Change in magnesium content is due to the chelation of the mineral

		Magnesium (mg/100g)					
Freatment	0	2 nd day	5 th	8 th	10 th day		
	day		day	day			
Variety							
V ₁ (Methi Local)	70.57	81.28	130.43	157.86	151.71		
V ₂ (Methi Shalimar Improved)	73.14	84.51	133.95	158.86	153.86		
C.D. (p≤0.05)	0.85	0.57	0.88	0.61	NS		
Chemical							
Γ_0 (Control)	66.00	77.50	127.50	157.00	151.00		
Γ_1 (Vitamin C (100 μ M)	68.50	79.00	128.50	158.00	151.50		
Γ_2 (Vitamin C (500 μ M)	71.00	81.50	131.50	158.50	154.00		
Γ ₃ (Folic acid (50 μM)	71.00	80.50	130.50	157.10	151.50		
Γ_4 (Folic acid (100 μ M)	74.00	85.00	133.00	159.00	154.00		
Γ ₅ (Chitosan (1000 ppm)	75.00	87.50	136.33	159.00	152.50		
Γ_6 (Chitosan (1500 ppm)	77.50	89.50	138.00	160.00	155.00		
C.D. (p <u><</u> 0.05)	1.59	1.07	1.65	1.15	NS		

with protein, in a way that increases its function (Shipard, 2005).

From these in vitro results, it is clear that sprouting as well as pretreatment of fenugreek sprouts with the used food grade additives possess strong capacity to increase the quantity of nutrients. The current results clearly suggest that these sprouts would be high quality food with enhanced nutrient contents.

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