Atomic Absorption Spectrophotometer Analysis For Determination Of Variation In Mineral Content In Fenugreek Genotypes Cultivated At Three Different Locations.

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ABSTRACT: Fenugreek is an annual herbaceous legume, well known for its medicinal importance. It is rich in bioactive compounds besides being a renowned nutraceutical. It is consumed as a vegetable during the growing season and is available as a spice round the year. The following study aims at the estimation of some micronutrients namely calcium, iron, zinc, magnesium and copper from seeds of fenugreek genotypes using atomic absorption spectrophotometry. The genotypes in the present study have been cultivated at three different places in India. The present study aims to observe the variation in the micronutrient content, if any, in the 13 genotypes under study. The results show the mineral content and the variation in all the genotypes analyzed here.

KEY WORDS: Fenugreek; legume; micronutrients.

I. INTRODUCTION:

Fenugreek (commonly known as methi in Hindi) is an annual, self pollinated leguminous crop cultivated in Indian and Eastern Mediterranean region. The tender green leaves of fenugreek are consumed as a vegetable while the seeds are used as spice (Petropolus, 2002). The seeds of fenugreek have a peculiar odor and bitter taste. Fenugreek is a well known medicinally important legume beneficial in various ailments. It has been recognized as an antioxidant, blood sugar and cholesterol lowering agent (Al- Habori and Raman 2002; Suboh et al., 2004). It has been associated with gastro protective, immune modulatory, laxative and anti mutagenic effects as well. Fenugreek is rich in carbohydrates, proteins, fiber and various bioactive compounds such as diosgenin, trigonelline, galactomannan etc. (Jayaweera , 1981; Yoshikawa *et al.*, 1997). There are various clinical trials that confirm the therapeutic benefits of fenugreek (Sameer *et al.*,2004; Amr Amin *et al.*,2005; Tadigoppula *et al.*,2006). Minerals are also an important part of a balanced diet. The minerals or micronutrients perform various essential roles in the body. It is, therefore, essential to ensure that the diet must include the recommended dietary allowance of the micronutrients. Incorporation of natural ingredients in the diet is beneficial as it will provide various nutrients besides providing the longevity promoting phytochemicals and antioxidants. The present study aims at estimating quantitatively the micronutrient content (calcium, zinc, iron, copper, magnesium) in the seeds of fenugreek genotypes that have been cultivated at three different locations.

II. MATERIALS AND METHODS:

The seeds of the fenugreek genotypes were obtained from Vegetable Research Centre, Pantnagar (Uttarakhand) 2) Hisar Agriculture University, Hisar (Haryana) 3) S.K.N.College of Horticulture, Jobner (Rajasthan). All the standard solutions used were obtained from Merck. The nitric acid and hydrogen peroxide used were of analytical grade. All the glassware's used were washed with reverse osmosis water, soaked in 10 % HNO₃ overnight and finally washed using ultra pure water. For analysis of micronutrients one gram of powdered seed was weighed accurately. HNO₃ (10 mL) and H_2O_2 (2 mL) were added to the weighed sample and digested on a hot plate. The digested samples were diluted to 50 mL using 0.1 % HNO₃ using a volumetric flask. The diluted samples were filtered and stored for further analysis. The quantitative estimation of micronutrients was done using atomic absorption spectrophotometer. The statistical analysis has been using ANOVA. The mean value of the micronutrients of each genotype from all the three locations were compared using ANOVA to find out the presence or absence of significant difference in the micronutrient.

III. RESULTS AND DISCUSSION:

The micronutrient content estimation on the fenugreek genotypes was performed for five minerals namely calcium, iron, zinc, copper and magnesium. The results given in table (1,2,3,4,5) show the difference observed in the genotypes with respect to the micronutrient content.

S.No.	Calcium content in mg/gram				
	Genotype	Pantnagar (mg/gm)	Rajasthan (mg/gm)	Hisar (mg/gm)	
1	PRM 45	5.521 ± .74	8.271 ± 2.23	9.935 ± 3.90	
2	AFG 3	9.942 ± 2.6	6.757 ± 2.60	6.277±2.04	
3	AFG 4	8.014 ± 4.27	8.007 ± 2.60	8.205 ± 2.23	
4	LFC 105	9.442 ± 1.86	7.042 ± 2.23	6.317 ± 2.04	
5	LFC 103	5.534. ± 1.48	7.672 ± 2.41	7.060 ± 2.23	
6	HM 348	10.192 ± 2.97	6.284 ± 1.86	8.310 ± 2.60	
7	HM 355	10.001 ± 3.16	7.909 ± 4.27	4.843 ± 3.90	
8	UM 330	6.382 ± 1.30	2.988 ± 2.04	4.738 ± 1.67	
9	UM 364	10.350 ± 2.23	9.323 ± 1.48	8.540 ± 1.30	
10	UM 365	8.310 ± 1.48	5.922 ± .93	7.902 ± 1.86	
11	UM 366	9.126 ± 1.86	8.818 ± 2.04	7.244 ± .74	
12	NDM 19	9.277 ± 2.41	8.073 ± 2.23	8.178 ± 2.60	
13	NDM 20	6.514 ± 3.90	6.534 ± 4.83	6.461 ± 2.79	

Table 1: Calcium estimation in fenugreek genotypes collected from three different locations

No significant difference was found (p< .05)

Table 2: Magnesium estimation in fenugreek genotypes collected from three different locations

S.No.	Magnesium content in mg/gram				
	Genotype	Pantnagar (µg/gm)	Rajasthan (µg/gm)	Hisar (µg/gm)	
1	PRM 45	269.176 ± .76	258.13 ± .46	260.32 ± .39	
2	AFG 3	276.1156 ± .53	255.27 ± .43	247.37 ±.36	
3	AFG 4	259.175 ± .29	283.64 ± .46	260.32 ± .39	
4	LFC 105	267.187 ± .34	212.36 ± .07	255.87 ± .44	
5	LFC 103	263.2023 ± .46	265.99 ± .39	247.04 ± .35	
6	HM 348	273.81 ± .49	251.36 ± .39	247.55 ± .36	
7	HM 355	289.64 ± .29	275.34 ± .66	214.49 ± .09	
8	UM 330	265.81 ± .58	270.17 ± .41	249.88 ± .38	

9	UM 364	262.29 ± .51	279.66 ± .42	253.03 ± .41
10	UM 365	258.73 ± .36	264.00 ± .48	271.54 ± .56
11	UM 366	271.78 ± .61	210.35 ± .06	240.01 ± .28
12	NDM 19	275.79 ± .58	260.04 ± .41	260.10 ± .40
13	NDM 20	249.43 ± .38	277.82 ± .62	258.08 ± .46

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No significant difference was found (p< .05)

Table 3: Iron estimation in fenugreek genotypes collected from three different lo	ocations
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S.No.	Genotype	Pantnagar (µg/gm)	Rajasthan (µg/gm)	Hisar (µg/gm)
1	PRM 45	89.04 ± 0.07	23.64 ± .006	19.15 ± .10
2	AFG 3	30.80 ± .06	14.20 ± .11	24.72 ± .09
3	AFG 4	31.61 ± .05	10.69 ± .12	10.77 ± .12
4	LFC 105	76.75 ± .05	5.89 ± .01	15.52 ± .11
5	LFC 103	28.36 ± .07	9.72 ± .016	16.84 ± .11
6	HM 348	23.55 ± .08	17.36 ± .05	14.15 ± .10
7	HM 355	49.38 ± .01	14.96 ± .02	33.93 ± .05
8	UM 330	22.28 ± .08	44.80 ± .03	13.43 ± .02
9	UM 364	41.42 ± .04	28.13 ± .06	20.22 ± .09
10	UM 365	22.56 ± .09	24.80 ± .08	37.86 ± .07
11	UM 366	21.12 ± .08	19.10 ± .12	30.42 ± .06
12	NDM 19	20.22 ± .09	15.34 ± .10	16.54 ± .10
13	NDM 20	$45.82 \pm .02$	15.52 ± 11	24.57 ± .07

Significant difference was found between the genotypes (p<.05)

	Zinc content in mg/gram				
S.No.	Genotype	Pantnagar (µg/gm)	Rajasthan (µg/gm)	Hisar (µg/gm)	
1	PRM 45	60.45 ± .03	53.82 ± .003	65.65 ± .02	
2	AFG 3	61.5 ±.008	37.92 ± .02	68.37 ± .03	
3	AFG 4	47.05 ± .01	54.97 ± .006	63.72 ± .02	
4	LFC 105	38.22 ± .02	25.4 ± .02	53.07 ± .01	
5	LFC 103	43.85 ± .02	48.03 ± .01	44.8 ± .02	
6	HM 348	37.15 ± .016	35.4 ± .02	61.5 ± .01	
7	HM 355	32.55 ± .015	32.05 ± .01	35.17 ± .02	
8	UM 330	42.95 ± .02	45.82 ± .02	62.95 ± .01	
9	UM 364	68.05 ± .02	50.92 ± .01	70.72 ± .03	
10	UM 365	57.9 ±.005	36.07 ± .02	52.75 ± .01	
11	UM 366	41.47 ± .02	75.5 ± .02	70.5 ± .02	
12	NDM 19	46.02 ± .02	53.37 ± .004	49.67 ± .01	
13	NDM 20	55.47 ± .007	37.97 ± .02	43.02 ± .02	

 Table 4: Zinc estimation in fenugreek genotypes collected from three different locations

Significant difference was found between the genotypes (p<.05

Table 4: Copper estimation in fenugreek genotypes collected from three different locations

	Copper content in mg/gram				
S.No.	Genotype	Pantnagar (µg/gm)	Rajasthan (µg/gm)	Hisar (µg/gm)	
1	PRM 45	7.37 ± .94	5.75 ± 1.02	9.14 ± .99	
2	AFG 3	9.17 ± 1.02	5.09 ± 1.41	10.87 ± .96	
3	AFG 4	8.11 ± .75	5.85 ± 1.26	9.86 ± .76	
4	LFC 105	12.51 ± .82	4.32 ± .53	9.07 ± .88	
5	LFC 103	6.76 ± 1.31	5.88 ± 1.29	6.71 ± 1.24	
6	HM 348	10.84 ± .92	4.08 ± 1.22	8.99 ± .77	
7	HM 355	9.09 ± .91	7.27 ± .80	10.01 ± .97	
8	UM 330	5.73 ± 1.08	5.63 ± .94	9.07 ± .88	
9	UM 364	9.12 ± .95	5.80 ± 1.19	9.91 ± .83	
10	UM 365	9.04 ± .84	2.49 ± 1.42	7.50 ± 1.12	

11	UM 366	7.57 ± 1.23	9.89 ± 2.41	10.77 ± .81
12	NDM 19	8.11 ± .75	5.65 ± .97	9.07 ± .88
13	NDM 20	9.34 ± 1.27	7.52 ± 1.16	7.40 ± .98

Significant difference was found between the genotypes (p<.05)

The estimation of micronutrients in fenugreek genotypes shows the variation in the content of micronutrients. Significant differences were observed in the content of iron, zinc and copper whereas the content of magnesium and calcium was not significantly different between the genotypes (p < .05). Calcium is an important micronutrient. It is required for the proper functioning of heart, muscles and nerves besides being involved in blood clotting (Pravina et al., 2013). Its deficiency may lead to osteoporosis (Soetan et al., 2010). The results of calcium estimation show the presence of calcium ranging from 2.988 ± 2.04 mg/gram to 10.350 ± 2.23 mg/gram. The lowest calcium content among the genotypes analysed was found in UM 330 cultivated at Jobner, Rajasthan while the highest calcium content was found in UM 364 cultivated at Pantnagar, Uttarakhand . The recommended dietary allowance of calcium is 1000 mg. There are several factors involved in determining the calcium content. The absorption of calcium by roots is a genetically determined factor that decides the rate of root growth as well as the apoplastic and symplastic calcium absorption by the roots and subsequent transfer to the other plant parts (Mclaughlin and Wimmer 1999). High level of root growth can increase the rate of calcium absorption by the plants. The magnesium content ranged from $212.36 \pm .07 \ \mu g \ /gm$ to $289.64 \pm .2907 \mu g/gm$. The dietary allowance from magnesium is 400 mg. The magnesium content was almost consistent in all the genotypes analyzed. Magnesium is important for energy production and transport in the body as it is involved in glycolysis, oxidative phosphorylation. It is also required for maintaining normal heart rhythm (Soetan et al., 2010).. Magnesium is also involved in muscular activity and it is also required by more than 300 enzymes in the body that catalyze various important functions such as protein synthesis, muscle and nerve function.

The iron content ranges from $5.89 \pm .01 \,\mu\text{g}$ /gm to $89.04 \pm 0.07 \,\mu\text{g}$ /gm. Iron is an important mineral as it performs several important functions such as being a component of enzymes involved in energy production , metabolism of proteins, nucleotides as well as in synthesis of proteins and neurotransmitters. It is a component of heamoglobin and myoglobin hence involved in oxygen transport in the body (Soetan *et al.*, 2010)..The lowest content of iron was found in Jobner,Rajasthan cultivated genotype LFC 105 while highest content was reported in PRM 45 cultivated at Pantnagar, Uttarakhand It was observed that the iron content was comparatively higher in the genotypes that were grown at Pantnagar as compared to those grown at Rajasthan. The genotypes grown at Rajasthan were lagging behind in the iron content may be the alkaline soil type of the arid area of Rajasthan which is highly permeable and loose textured that ultimately results in decreased mobilization of iron. The recommended dietary allowance of iron is 18 mg. Iron deficiency is a commonly encountered problem prevalent in the female population. To alleviate the iron deficiency the supplementation with iron capsules is not enough as the absorption of iron is also a major factor.

It has been found that vitamin C enhances the potential of iron for mucosal uptake (Cook and Reddy, 2001). Fenugreek is also rich in vitamin C (12 mg/100 gm) (Meghwal and Goswami, 2012). Therefore consumption of fenugreek can be a good remedy for iron deficiency control as it will promote better bioavailability. The zinc and copper content ranged from $25.4 \pm .02 \ \mu g \ /gm$ to $68.05 \pm .02 \ \mu g \ /gm$ and from $2.49 \pm 1.42 \ \mu g \ /gm$ to $12.51 \pm .82 \ \mu g \ /gm$, respectively.Zinc is vital for the immune system. It is also involved with the sense of taste , smell and apetite as the areas of brain that are involved in these perceptions are activated by zinc. Zinc is also important for growth, so it becomes vital during pregnancy because of the rapidly dividing cells of the foetus.Zinc also prevents night blindness and prevents development of cataract (Soetan *et al.*, 2010).. Copper is required for the development of foetal brain and maintenance of brain throughout the life . It is also involved in the formation of the cells of the immune system and it also maintains proper structure and function of circulating blood vessels (Soetan *et al.*, 2010).. The recommended dietary allowance for zinc and copper is 15 and 2 mg per day respectively.

The lowest zinc content was observed in Jobner Rajasthan cultivated genotype LFC 105 while the maximum zinc content was found in Jobner Rajasthan cultivated genotype UM 366. The lowest and highest

copper content was found in UM 365 from Jobner, Rajasthan and LFC 105 from Pantnagar, Uttarakhand respectively.

IV. CONCLUSION:

This study shows a comprehensive evaluation of five micronutrients estimated in thirteen genotypes grown at three different environmental conditions. The results indicate that fenugreek genotypes cultivated at different locations exhibited variation in the content of mineral elements. The content of micronutrients in the genotypes is not very significant as far as the recommended dietary allowance is considered but the incorporation of fenugreek in diet will provide a small part of the mineral requirement besides safeguarding the body from oxidative damage as a result of it's antioxidant properties (Anuradha and Ravikumar,2001). The incorporation of natural sources for supplementing the dietary requirements is the best possible measure as besides the desired nutrients there are a lot more health promoting phytochemicals that a natural source offers to the body. Therefore fenugreek can be considered a longevity promoter that will boost the overall health status on consumption.

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