Nutritional and antinutritional attributes of faba bean (*Vicia faba* L.) germplasms growing in Bihar, India

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Abstract Eleven germplasms of faba bean seeds from four agroclimatic regions of Bihar, India, have been investigated to estimate their nutritional (soluble protein, free amino acids, starch, reducing and non reducing sugar, total soluble sugar) and antinutritional (total extractable phenol and condensed tannin/proanthocyanidin) parameters. These parameters were found in varying concentration in all genotypes studied. The highest concentration of total extractable phenol and proanthocyanidin (condensed tannin) (2.56 and 1.59 % leucocyanidin equivalents respectively on dry matter basis) were found in Samastipur while the lowest from Patna (0.95 and 0.426 % leucocyanidin equivalent on dry matter basis). The different nutritional parameters were also found to be in variable concentration among different germplasms viz. total soluble protein $\approx$ 20–32 %, free amino acids $\approx$ 188–348 mg/100 g, starch $\approx$ 27–33 %, reducing sugars $\approx$ 85–188 mg/100 g, non reducing sugars $\approx$ 0.7–1.7 % and total soluble sugars $\approx$ 0.8–1.9 %.

Keywords Faba bean · Nutritional parameters · Antinutritional parameters

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Introduction

Faba bean (*Vicia faba* L.) is a unique legume crop because of its extremely rich nutrient content which also serves as an excellent source of proteins, complex carbohydrates, dietary fibre, choline, lecithin, minerals and secondary metabolites such as phenolics. Interestingly, in contrast to cereals, faba bean is found to contain high level of lysine and arginine, which may complement the low levels of those in cereals. Much attention is now being paid on the antioxidant capacity of the flavonoids and the phenolics (Sinha et al. 2013). The major Indian pulse producing areas often encounter adverse climatic conditions causing huge yield loss which consequently widens the gap between demand and supply and hence a huge import price. One of the possible ways to narrow down this gap between demand and supply could be the utilization of underutilized pulse crops. In addition to Faba bean's agricultural potential, it is also known for its industrial value as it is rich in levodihydroxy phenylalanine (L-dopa), the precursor of dopamine. Its consumption can increase the levels of L-dopa in the blood, with a marked improvement in the motor performance of the patients suffering from Parkinson disease, without any side effects (Liu et al. 2000; Rabey et al. 1992; Apaydin 2000). L-dopa, in the natural faba bean background, was observed to show positive clinical benefits (Mohseni Mehran and Golshani 2013). Faba bean is also being investigated as a dietary complement for other diseases such as hypertension, renal failure and liver cirrhosis (Kwok et al. 1997; Randhir and Shetty 2004; Randhir et al. 2002). Considering the high agricultural as well as industrial potential of this underutilized pulse crop and the need for diversification of a potential legume crop along the lines of cereals, it is found desirable to study the agricultural potential, performance and prospects of faba bean. Therefore, we studied the nutritional and antinutritional attributes of eleven germplasms of faba bean covering all four agroclimatic region of Bihar, India.

Materials and Methods

The investigations were carried out on different germplasms of faba bean which were collected from farmer's field of different agro-climatic regions of Bihar, India (Pradhan et al. 2014). All the germplasms were grown in University Research Farm (Rajendra Agricultural University, RAU, Bihar, India) under normal agronomic conditions and the dried seeds were used in present study. Name of germplasms used in this study are same as their site of collection.

The protein was estimated by Bicinchoninic acid (BCA) method. The protocol of protein estimation was followed as per manufacturer's instruction given in Bicinchoninic acid (BCA) protein assay kit (G-Biosciences, St Louis, MO, USA). The amount of protein content of different seed samples were measured against Bovine Serum Albumin (2 mg/ml) standard solution. The amino acids in the free pool of seeds or protein hydrolysate of different samples were determined by using ninhydrin (Lee and Takahashi 1966).

The amount of total soluble sugars was determined as per the method described by Francis et al. (1971). 300 mg of seed sample was extracted with 80 % ethanol and the colour of final product was read at 600 nm against glucose standard (100 µg/ml). The same extract were used for estimation of reducing sugars as described in case of total soluble sugar and the method was followed as per the procedure described by Nelson (1944). Blue colour produced in reaction mixture was measured at the absorbance level of 520 nm and amount of reducing sugars were calculated against standard of glucose. Non-reducing sugars were estimated by subtracting the total soluble sugar and reducing sugar. Starch was estimated in different samples by following the method described by Hodge and Hofreiter (1962). The intensity of colour formed was read at 620 nm against glucose standard solution (100 µg/ml). The readings were multiplied by a factor 0.9 as 0.9 g of starch yields 1 g of glucose on hydrolysis.

Total extractable phenol and condensed tannins (Proanthocyanidins) were measured in seed extracts of each sample. Extract for these purpose were prepared according to the method described by Makkar and Goodchild (1996), using 70 % aqueous acetone. Total phenolic content of seed extract was determined by spectroscopy at 725 nm, on the basis of a colorimetric reaction with Folin-Ciocalteu reagent, as described by Makkar and Goodchild (1996). The results were expressed as tannic acid equivalent on a dry matter basis. Proanthocyanidin (condensed tannin) content was determined colorimetrically at 550 nm using butanol-HCl and ferric reagent (Makkar and Goodchild 1996; Porter et al. 1986) and was expressed as leucocyanidin equivalent on a % dry matter basis. After cooling the tubes, absorbance was read at 550 nm (Porter et al. 1986).

All experiments and/or measurements reported in this study were replicated three times. In each case a mean value of standard deviation was calculated. Analysis of variance (ANOVA) was performed and differences in mean values determined using Tukey's b studentized test at $p < 0.05$. The statistical analysis was performed using Statistical Package for the Social Sciences (SPSS, 2003).

Results and Discussions

The different nutritional and antinutritional parameters were found in varying concentrations in all samples taken in present studies. Proteins represent the major storage compound which is a common trait of most legume seeds. It is also evident in our studies that protein content was very high in almost all faba bean germplasms which ranges from 19.5 to 33.3 % on dry matter basis. As compared to different seed samples, protein content was highest in Motihari seeds whereas lowest in Katihar seeds (Fig. 1). With respect to free amino acid composition Gaya and Siwan have significantly higher

Fig. 1 Protein and free amino acids content of mature seeds of 11 germplasms: Results are mean values of triplicate determinations, $\pm$ standard deviation and are on dry weight basis. Error bars are standard error of means

Table 1 Total soluble sugars, reducing and non-reducing sugars, and starch content of mature seeds of eleven germplasms of *Vicia faba*

Name of germplasms	Total soluble sugar (%)	Reducing sugar (%)	Non reducing sugar (%)	Starch (%)
Samastipur	1.879±0.04 ^f	0.101±0.007 ^b	1.777±0.03 ^f	27.83±0.23 ^c
Vaishali	0.879±0.13 ^b	0.141±0.00 ^d	0.738±0.13 ^b	32.83±0.23 ^c
Motihari	1.564±0.18 ^c	0.155±0.001 ^d	1.408±0.18 ^c	30.50±1.17 ^d
Madhubani	1.166±0.12 ^d	0.101±0.005 ^b	1.065±0.12 ^d	32.16±0.23 ^c
Siwan	0.805±0.02 ^a	0.110±0.00 ^b	0.695±0.03 ^a	29.00±0.00 ^c
Katihar	1.286±0.15 ^d	0.145±0.001 ^d	1.142±0.15 ^d	27.83±0.70 ^b
Purnia	0.907±0.06 ^c	0.085±0.005 ^a	0.822±0.07 ^c	27.16±0.23 ^a
Patna	1.157±0.13 ^d	0.188±0.001 ^c	0.969±0.14 ^d	29.83±0.23 ^d
Jahanabad	1.094±0.05 ^d	0.153±0.00 ^d	0.957±0.06 ^d	28.50±0.24 ^c
Gaya	1.546±0.08 ^c	0.138±0.001 ^c	1.407±0.08 ^c	33.00±0.00 ^c
Bhagalpur	1.083±0.01 ^d	0.081±0.001 ^a	1.001±0.01 ^d	32.17±0.70 ^c

Results are mean values of triplicate determinations, ± standard deviation. Results are on a dry weigh basis. Mean followed by different superscripts in each column are significantly ($p<0.05$) different from one another

amount, while Patna, Jahanabad, Bhagalpur, Vaishali and Motihari are grouped under the lowest amount of free amino acid group (Fig. 1). Although free amino acids have little effect on the nutritional value of the seeds (Kaldy and Kasting 1974), but as seeds mature, these stored free amino acids are converted to storage proteins and/or non protein constituents. High level of free amino acids in maturing seeds has been shown to contribute to a faster and greater accumulation of protein in protein bodies.

The total soluble sugars were found to be highest in Samastipur seed (1.879 %) while lowest in Siwan seeds (0.805 %). With respect to reducing sugars, Patna and Bhagalpur were found to contain highest and lowest amount respectively. Whereas, Samastipur contained highest amount and Siwan had lowest concentration of non reducing sugar. As far as starch content is concerned, Gaya and Purnea occupied highest and lowest position respectively among all germplasms. The data related with above mentioned parameters are given in Table 1. The quantitative changes in free amino acids, carbohydrates and total proteins in maturing

seeds may be closely related to one another but may vary among cultivars. It is hypothesized that carbon from starch is used either for lipid synthesis or for protein synthesis especially in pulses seed where proteins are by far the major storage compounds. Amino acids needed for reserve protein synthesis, especially those that are not provided by maternal seed parts are also synthesized by the hydrolysis of starch. Thus this type of information can become valuable in future studies to regulate the quantity of these constituents in maturing seeds by possibly altering certain genetic and agronomic factors. Although different soluble sugars were not determined separately in present study but major soluble sugars present in seeds are sucrose and raffinose family oligosaccharides (RFOs).

The antinutritional activity of phenolics and tannins may be due to complexation with digestive protein in general and with digestive enzymes, such as trypsin and chymotrypsin, in particular (Liener 1989). This function of tannin is responsible for the astringency of tannin-rich foods which actually reduces its wider acceptability by end users. However, the polyphenols and tannins

Fig. 2 Total extractable phenol and condensed tannin content of mature seeds of 11 germplasms: Results are mean values of triplicate determinations, ± standard deviation and are on dry weight basis. Error bars are standard error of means

have implications for health and nutrition due to their antioxidant activities. The highest concentrations of total extractable phenol were present in Motihari seed and lowest in Patna. Proanthocyanidins (condensed tannin) were found in Samastipur at highest concentration while the lowest from Patna seed (Fig. 2). The higher content of phenolics affects the nutritional value of seeds, however, antioxidant activity of phenolic compounds (condensed tannin) in general may exert a positive influence on potential beneficial health effect of faba bean. It is widely known that significant antioxidant activity of food is related to high total phenolic contents (TPC). Condensed and hydrolysable tannins of relatively high molecular weight have been shown to be effective antioxidants with greater activities than simple phenolics (Hagerman et al. 1998).

In order to ascertain the possibilities of incorporation of faba bean (*Vicia faba* L.) in human diet and its further improvement for agricultural or industrial purposes, the evaluation of different germplasms for its different qualities are largely required. Cultivar identification has attained critical importance in the national and international seed programme as it is realized that if true to type seed of a variety is not available then the entire crop production programme may fail. In this direction earlier we reported the characterization of these germplasms on the basis of different storage proteins and isozymes (Pradhan et al. 2014). Thus it is of paramount importance to characterize varieties for their correct identification. The present study also showed valuable information for plant breeder for its further improvement.

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Conflict of interest Authors declare no conflict of interests.

Description of contribution Conceived and designed the experiments: SKS. Performed the experiments: AK, SKS. Analyzed the data: SKS, N, NP. Contributed to the writing of the manuscript: SKS

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