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Variation in biochemical constituents and cooking quality of rajmash genotypes of Himachal Pradesh

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Abstract

Dry mature seeds of 16 genotypes of rajmash procured from HAREC, Kukumsari during the year 2004, were analyzed for various physical and biochemical attributes of dietary significance by following standard procedures. Among physical parameters 100-seed weight, seed husk and water absorption capacity ranged from 22.06 to 61.15g, 5.52 to 8.26 and 90.81 to 110 % in that order. The analysis of biochemical attributes of these genotypes revealed wide variations in moisture content (7.59 to 11.14%), proteins (20.80 to 26.72 %), essential limiting amino acids viz. methionine (0.48 to 0.86 g/100g of protein) and tryptophan (0.59 to 1.14 g/100g of protein), *in vitro* protein digestibility (63.40 to 72.68 %), total carbohydrates (50.83 to 64.80%), starch (38.43 to 50.49%), total sugars (7.0 to 9.0%), total tannins (127 to 1221 mg/100g of dry weight), PPO activity (6.27 to 14.13 units/min/g) and raffinose content (0.28 to 0.74 %). In all, the study indicated that among various genotypes VLF-106, HPR-43 and HPR-354 in seed protein; HURG-33, SR-71(I) and HPR-396 in carbohydrates; K-38, HPR-43 and Triloki X JJC-2 in methionine; K-52, Triloki X JJC-2 and Hans in tryptophan; SR 71(I), HPR-337 and K-52 for *in vitro* protein digestibility; Hans, HPR-43 and HPR-337 in tannins; HPR-337, Hans and Kanchan in raffinose; and Hans, HPR-191 and HPR-337 with comparatively less cooking time were adjudged as promising genotypes for individual quality attribute(s) to be useful for further crop improvement purpose. Moreover, cumulative varietal grading based on over all performance of varieties/ genotypes with regard to various nutritional characteristics showed that Hans, HPR-337 and HPR-43 emerged versatile genotypes of rajmash in order of excellence.

Key words: Raffinose, tannins, polyphenol oxidase (PPO) activity, *in vitro* protein digestibility.

Introduction

Rajmash (*Phaseolus vulgaris* L.) belongs to the family *Leguminoceae*. It is native to South Mexico and Central America, while secondary centre lies in Peru, Bolivia and Eucador region of American continent (Smartt, 1990). Rajmash plays an important role as a source of essential nutrients in the traditional diets of millions throughout the world particularly in developing countries. Its food value chiefly lies in proteins, fats, carbohydrates, fibres and trace elements like magnesium, manganese, iron, etc. In developing countries like India, the dietary consumption of rajmash has increased appreciably in the form of various culinary preparations. In India, it is grown over an area of about 1.5 lakh hectares with an annual production of 4.2 lakh metric tonnes (Anonymous, 2002). Rajmash also known as kidney or French bean is widely grown pulse crop in parts of northern India. Owing to its adaptation to high altitude areas, it is viewed as potential source of protein and other nutrients for supplementary diet for people of hills in particular. It is grown as sole crop as well as inter crop with maize in *kharif* in hills as well as its cultivation is under trial in *rabi* season in plains. The crop is adapted to wide range of climate from temperate to sub-temperate climate and hence, Himachal Pradesh contributes substantially in its production. In Himachal Pradesh, it occupies an area of about 2444 hectares with an annual production of 27202 metric tonnes (Anonymous, 2004). In Himachal Pradesh, it is extensively grown in Kinnaur, Lahual-Spiti, Sirmour, Bilaspur, Solan, Chamba, Mandi and Kangra districts. Undoubtly, rajmash seeds are rich source of protein and energy, however, their proteins have been characterized as having relatively low in nutritive value due to limiting amounts of sulphur containing amino acids, low digestibility, presence of toxic principles and other antinutritive factors including flatulence causing oligosaccharides

(raffinose) (Tezoto and Sgarbieri, 1990).

Literature revealed that research efforts are on in relation to quality improvement of food legumes, however, systematic information on variation in biochemical constituents of rajmash genotypes particularly grown in Himachal Pradesh still appears to be scanty. Therefore, the present study was undertaken to evaluate the biochemical constituents and cooking quality of dry mature seeds of rajmash genotypes grown in Himachal Pradesh.

Material and Methods

Seed samples of 16 rajmash varieties/ genotypes grown in the randomized block design with three replications by following the recommended cultural practices at Hill Agriculture Research and Extension Centre, Kukumseri (H.P.) during 2004, were procured and analysed for various physical and biochemical attributes. Dry mature seeds of these rajmash genotypes were cleaned and freed from extraneous substances and dried in an electric oven at $60^0 \pm 5^oC$. The oven dried samples were ground in a Willey mill to pass through 40-mesh sieve and stored in polythene bags. The samples were labelled and kept at room temperature for further analysis. Moisture and crude protein were analysed as mentioned in AOAC (1970), carbohydrate by the method of Yemm and Wills (1954), total sugars were extracted in alcohol (80%) by following the procedure of AOAC (1970) and then estimated in the test extract by following the phenol sulphuric acid method of Dubois *et al.* (1956). The total sugar free residue was dried and the starch was extracted with 52 per cent perchloric acid at room temperature as describe by Clegg (1956) and the starch content was then estimated by employing the procedure given by Dubois *et al.* (1956). Methionine and tryptophan were estimated by the procedures described by Horn *et al.* (1946) and Spies and Chambers (1949), respectively. Among anti-nutritional factors total tannins were determined by the method Makkar *et al.* (1993), polyphenol oxidase (PPO) activity by following Bastin and Unluer (1972). Estimation of raffinose was done by the method of Somiari and Balogh (1993). *In vitro* digestibility of protein was evaluated by the method described by Calsamiglia and Stern (1995). 100-seed weight (in grams) was recorded by weighing randomly selected one hundred seeds. Water absorption by the seeds was worked out by soaking 25 seeds (pre-weighed) in water for 24 hours at ambient temperature. The husk percentage was found out by soaking 100 g of seeds in water for 48 hrs, removing and drying wet husk in hot air oven at 65°C and then weighing it till constant weight was obtained. In order to record the cooking time, fifty whole rajmash seeds were cooked in a beaker with water (1: 10 w/v). Promising rajmash genotypes superior in individual quality parameter(s) were identified that might be useful for further crop improvement through breeding/biotechnological approaches. Besides, an attempt was also made to carry out over all genotypic ranking in respect of various quality attrtibutes *viz*; protein, carbohydrates, limiting amino acids- methionine and tryptophan, *in vitro* protein digestibility and cooking time, tannins and raffinose content in order to identify nutritionally superior multipurpose genotypes suited for their direct inclusion in human diet.

The data pertaining to various biochemical and other attributes of selected genotypes of rajmash were analysed statistically by using Analysis of Variance (Panse and Sukhatme, 1984) and by Duncan's Multiple Range Test (Gomez and Gomez, 1984).

Results and Discussion

The results on physical, biochemical and cooking quality characteristics of dry mature seeds of rajmash genotypes are presented in Table 1. It was observed that 100-seed weight ranged from 22.06 (HPR-191) to 61.15 g (HPR-354) thereby indicating significant variation among various genotypes with respect to this yield attributing trait. An overview of cooking quality of food legumes revealed an association between cooking quality, husk content and water absorption capacity. The husk content of dry mature seeds of various genotypes varied from 5.52 (VLF-106) to 8.26 (HPR-191 and HPR-354), while, the water absorption capacity ranged from 90.81 (VLF-106) to 110.0 per cent (HPR-372). Seed moisture content which roughly indicates degree of maturity was observed to vary from 7.59 (Triloki X JJC-2) to 11.14 (HURG-33) per cent in dry mature seeds of rajmash. Wide range of variation among genotypes with respect to moisture content might be attributed to differences in accumulation of photosynthates and other dietary constituents in dry matter of the seeds.

The highest protein content was found in VLF-106 (26.72%) followed by HPR-43 (25.00%) and HPR-354 (24.60%) genotypes. The lowest value

Table 1. Variation in physical and biochemical attributes of dry mature seeds of rajmash genotypes

Parameters*	HPR-43	HPR-191	HPR-337	HPR-354	HPR-372	HPR-396	K-38	K-52	K-190-1	VLF-106	PDR-21	HURG-33	SR-71(I)	Hans **	TrilokiX JJC-2	Kanchan **	SE (±m)	CD (5%)
100-seed wt. (gm)	26.40	22.06	25.67	61.15	47.45	57.72	30.96	35.88	34.97	58.96	51.88	54.10	57.06	22.57	51.57	56.83	0.23	0.66
Husk (%)	7.39	8.26	8.12	8.26	6.79	7.27	7.51	8.23	7.34	5.52	6.57	5.86	7.20	6.79	6.46	5.92	0.03	0.80
Water absorption (%) capacity	95.43	100.6	101.8	99.50	110.0	100.0	104.2	104.7	92.06	90.81	93.39	98.81	98.62	92.08	106.2	97.05	0.28	0.82
Moisture (%)	10.20	10.20	10.20	9.60	9.46	9.90	10.16	10.06	9.58	10.28	10.18	11.14	10.10	10.10	7.59	10.90	0.1	0.18
Crude Protein (%)	25.00	22.70	22.60	24.60	22.70	23.92	21.49	21.60	22.90	26.72	24.14	23.63	20.80	23.40	23.40	22.20	0.2	0.66
Carbohydrates (%)	53.00	51.00	55.80	51.30	54.40	55.90	53.20	52.61	54.80	53.63	50.83	64.80	57.40	51.20	55.10	53.20	0.1	0.31
a) Starch (%)	42.00	38.43	43.70	40.10	43.10	42.44	41.22	40.06	41.50	40.29	40.35	50.49	45.70	40.20	44.20	39.90	0.3	0.9
b) Total sugars (%)	7.20	8.30	7.80	7.10	7.40	9.00	7.60	8.10	8.70	8.90	8.00	8.70	7.30	7.50	7.00	9.00	0.2	0.37
Methionine (g/100g protein)	0.86	0.48	0.55	0.66	0.57	0.62	0.86	0.55	0.52	0.62	0.51	0.51	0.76	0.65	0.84	0.62	0	0.03
Tryptophan (g/100g protein)	0.59	0.68	0.59	0.84	0.66	0.62	0.61	1.14	0.76	0.65	0.83	0.96	0.83	1.08	1.14	0.9	0	0.01
Total tannins (g/100g)	358	411	381	595	917	626	1004	1221	1043	543	509	762	608	127	941	593	0.99	2.84
PPO activity (u/min/g)	13.09	14.13	13.11	12.13	9.19	9.20	6.31	7.62	6.27	8.00	6.29	6.85	7.36	13.01	6.93	7.58	0.03	0.09
Raffinose (%)	0.54	0.65	0.28	0.43	0.55	0.38	0.74	0.67	0.41	0.53	0.43	0.38	0.64	0.34	0.44	0.36	0.003	0.01
In vitro protein digestibility (%)	69.53	70.24	72.38	66.74	66.49	67.40	67.40	72.18	67.94	67.06	63.40	64.92	72.68	69.03	69.32	71.47	0.14	0.40
Cooking time(min)	45	40	40	60	50	55	45	50	45	55	50	55	60	40	55	60		

* Values on dry weight basis. ** Released varieties of the State of Himachal Pradesh

of protein content was noticed in SR-71(I) genotype. The results of the present study are very close to those reported by other investigators on this aspect earlier (Samman *et al.*, 1999) and reported that the protein content varied from 18 to 20 per cent among various genotypes of rajmash in north-western Argentina.

The status of two essential and limiting amino acids of rajmash was evaluated and the results showed wide variation among different genotypes with respect to status of methionine and tryptophan content. The methionine content in rajmash genotypes varied from 0.48 to 0.86 g/100g, with highest value exhibited by K-38 and HPR-43 and the lowest in case of HPR-191. Since, methionine is actively involved in several trans-methylation reactions in the plants and animals, the genotype K-38 and HPR-43 might have an advantage over other genotypes in this aspect. The maximum and minimum value with regards to tryptophan content was found in K-52 and Triloki X JJC-2 (1.14 g/100g protein each) and HPR-43 and HPR-337 (0.59 g/100g protein each), respectively. Thus, genotype K-52 and Triloki X JJC-2 emerged out as superior ones over other genotypes with respect to tryptophan content. Almost similar trend of variation in tryptophan content was reported by Sood *et al.* (2003) in this crop. However, the level of methionine content was found slightly lower on different genotypes of rajmash in comparison with the results of Sood *et al.* (2003). This might be attributed to the genotypic variability and environmental factors associated with crop productivity.

In vitro protein digestibility gives an idea of protein quality of food legumes. Low protein digestibility is attributed to presence of various anti-nutritional factors including tannins, protease inhibitors and similar other factors (Srivastava and Srivastava, 2003). As such, *in vitro* protein digestibility in dry mature seeds of rajmash was also evaluated, which was found to vary from 63.40 to 72.68 per cent and genotypes SR-71(I) accompanied by HPR-337 and K-52 exhibited the highest value for *in vitro* protein digestibility of dry mature seeds of rajmash. The range of variation *in vitro* protein digestibility observed in rajmash genotypes was relatively higher than that in hyacinth legumes (Osama *et al.*, 2002). However, it was lower in case of rajmash genotypes, when compared with *in vitro* protein digestibility (76 to 82 %) of raw and cooked cow pea (Oliverira *et al.*, 2004). It indicated that the quality of the proteins was relatively better in cowpea than that of rajmash.

The study further revealed wide variations in carbohydrates to range from 50.83 (PDR-21) to 64.80 per cent (HURG-33) thereby indicating that PDR-21 had comparatively higher readily available source of energy in comparison to other genotypes. With regards to starch content, genotypes HURG-33 and HPR-191 exhibited the higher (50.49 %) and the lowest (38.43 %) value, respectively, while, the total sugars varied from 7.0 (Triloki X JJC-2) to 9.0 (Kanchan and HPR-396) per cent. The values obtained for these parameters were found slightly lower in case of red and white kidney beans as reported by Zia-ur-Rehman *et al.* (2001).

Keeping in view the adverse effect of anti-nutritional factors on nutritional quality of food and human health, status of prominent anti-nutritional factors *viz.*, total tannins and raffinose content in dry mature seeds of rajmash was evaluated. The range of variation with regard to total tannin content was noticed from 127 to 1221 mg per 100 g dry mature seeds. The genotypes Hans, HPR-43, HPR-337 and HPR-191 exhibited lowest value for tannins in that order. The range of variation in tannin content of this study is in agreement with the observations of other investigators in different legumes including rajmash (Sood, *et al.*, 2003, Srivastava and Srivastava, 2003). Significant variation was also observed in the polyphenol oxidase activity to vary from 6.27 (K-190-1) to 14.13 units/min/g (HPR-191). The findings of this study on this aspect are in close proximity with those of Sood *et al.* (2003). The study showed that raffinose content varied significantly from 0.28 to 0.74 per cent dry mature seeds with its lowest value found in the genotypes HPR-337, Kanchan and HURG-33. The trend of variation in raffinose content is in agreement with those reported by Mc Phee *et al.* (2002) who reported raffinose content vary from 0.1 to 4.1 mg/g of dry weight and the results of the study are in consonance with these workers.

Cooking time in different genotypes ranged from 40 to 60 minutes. The minimum cooking time was noticed in HPR-191 followed by HPR-337 and Hans whereas, the maximum cooking time was observed in Kanchan, SR-71(I) and HPR-354 in that order. The findings of the study with regards to variation in cooking time are in accordance with Singh *et al.* (1997), who observed 41 to 55 minutes of cooking time in black gram gentypes.

Table 2. Genotypic grading of rajmash varieties/genotypes in search of nutritionally superior multipurpose genotypes

Parameters	VLF-106	HPR-43	HPR-354	PDR-21	HPR-396	HURG-33	Hans	Triloki X JJC-2	K-190-1	HPR-372	HPR-191	HPR-337	Kanchan	K-52	K-38	SR-71(I)
1. Proteins*	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
2. Carbohydrates*	8	11	13	16	3	1	14	5	6	7	15	4	10	12	9	2
3. Tryptophan*	10	13	5	6	11	3	2	1	7	9	8	13	4	1	12	6
4. Methionine*	6	1	4	10	6	10	5	2	9	7	11	8	6	8	1	3
5. Total tannins**	6	2	8	5	10	11	1	13	15	12	4	3	7	16	14	9
6. Raffinose**	8	9	6	6	4	4	2	7	5	10	12	1	3	13	14	11
7. *In vitro* protein digestibility*	12	6	13	16	10	15	8	7	9	14	5	2	4	3	11	1
8. Cooking time**	4	2	5	3	4	4	1	4	2	3	1	1	5	3	2	5
Total	55	46	57	66	53	54	40	47	62	72	67	44	52	70	78	53
Cumulative grading	8	3	9	11	6	7	1	4	10	14	12	2	5	13	15	6

* Genotypes graded in the descending order for nutritionally desirable characters

** Genotypes graded in ascending order for nutritionally undesirable constituents

Based on biochemical evaluation of rajmash genotypes; it was observed that genotypes VLF-106, HPR-43 and HPR-354 in seed protein ; HURG-33, SR-71(I) and HPR-396 in carbohydrates; K-38, HPR-43 and Triloki X JJC-2 in methionine; K-52, Triloki X JJC-2 and Hans in tryptophan; SR 71(I), HPR-337 and K-52 in *in vitro* protein digestibility; Hans, HPR-43 and HPR-337 in total tannins; HPR-337, Hans and Kanchan in raffinose content and Hans, HPR-191 and HPR-337 with least cooking time emerged promising genotypes for individual quality parameters to be useful for further crop improvement purpose.

Besides, cumulative varietal grading based on over all performance of genotypes in respect of seed protein, carbohydrate, essential limiting amino acids-methionine and tryptophan, tannins, raffinose, *in vitro* protein digestibility and cooking time showed that Hans, HPR-337 and HPR-43 genotypes of rajmash were adjudged as overall superior multipurpose genotypes in order of excellence. Moreover, further studies are warranted to conduct animal feeding trials (nutritional studies) before recommending these versatile genotypes for mass cultivation and direct human consumption purposes.

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