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CHARACTERISATION OF A CORE SET OF COMMON BEAN (*Phaseolus vulgaris* L.) GERMPLASM FOR SEED QUALITY TRAITS

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SUMMARY

This study reports one of the first large-scale characterisation of the bean germplasm in the Jammu and Kashmir state of India for seed quality traits. The seed quality traits such as seed dry weight, seed wet weight and water absorption percentage as well as characteristics such as hydration and swelling varied greatly as about 80-90% of tested genotypes were spread across a broad range. The genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) values were higher for seed wet weight, water absorption, swelling and hydration traits. PCV values were higher than vales of GCV indicating significant influence of environment for these traits. The heritability parameters were invariably high for all traits except bulk density (0.45) and as high as 97% for traits like seed length, hydration coefficient, seed wet weight and swelling coefficient were recorded. All the 203 accessions were grouped into 7 clusters regardless of geographic origin and each cluster were composed of varied number of accessions. In this study, positive and significant correlations were found in case of seed water absorption and cooking traits. The suitable genotypes identified can be used as parents for hybridisation programme for improving quality traits in common bean.

Key words: Common bean, seed quality traits, cooking time score, sensory evaluation, principal component analysis

Key findings: The study characterised a core set of genotypes representing diverse market classes for seed quality traits and identified substantial variation that could be harnessed to improve common bean for quality traits pertaining to seed physical, biochemical, and cooking as well as sensory traits.

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INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is the most important legume grown worldwide for direct human consumption (Gepts, 2001). Also known as Rajmash, string bean, field bean, flageolet bean, French bean, garden bean, haricot bean, pop bean, or snap bean; it is a herbaceous annual plant grown worldwide for dry (mature) beans, shell beans (seeds at physiological maturity), and green pods. Its leaf is also occasionally used as a vegetable and the straw as fodder. When consumed as seed, beans constitute an important source of dietary protein

(22% of seed weight) that complements cereals for over half a billion people. Common bean has the potential to help mitigate malnutrition and hunger by virtue of being rich in quality globulin protein (20-28%) and micronutrients especially iron (70 mg/kg) and zinc (33 mg/kg) along with vitamin A. Menotti *et al.* (1999) reported that bean consumption is correlated with low incidence of coronary heart disease. Lately, domestic bean consumption has increased because of the rising importance of ethnic foods and the perceived health benefits related to the blood-cholesterol-lowering effects of beans (Anderson *et al.*, 1989) and high levels of certain minerals and vitamins.

In India, common bean is confined to the hilly tracts of Himachal Pradesh, Uttar Pradesh, Jammu and Kashmir and North Eastern states as a niche crop, as it requires moderate temperature for growth. Market preferences for seed physical traits like size, shape and colour have largely been the targets of breeding programmes in India. In Jammu and Kashmir (J&K), common bean is an important crop especially for the rain fed highlands where it is grown under maize-bean intercropping system. In J&K, pulses are cultivated over an area of about 30000 hectares with a production of 17000 tonnes and productivity of about 0.56 t/ha. In Kashmir valley, common bean is cultivated over an area of about 2000 hectares with production of about 1600 tonnes and yield of about 0.8 t/ha (Anonymous, 2012). The quality of common bean especially protein content, the cooking time, taste and freedom from flatulence are key parameters for farmer acceptability as well as marketability of common bean varieties. Hard to cook trait is the major bottleneck that is caused by hard seed coat, low seed coat permeability, less water absorption capacity and prolonged storage (> 5 years). Fast cooking beans have been found to imbibe more amount of water to justify its use as an indirect selection criterion (Kigel, 1999). Phytic acid and raffinose are considered to be the main antinutrional factors in common bean. Phytic acid chelates iron and reduces its bioavailability while as raffinose causes flatulence. There has been no serious effort in comparative evaluation of different market classes of Rajmash for quality traits to establish the relative superiority of local Rajmash for post-harvest seed culinary and quality traits.

Common bean is regarded as a nearly perfect food by virtue of its being a chief source of protein as well as providing substantial amounts of minerals and anti-oxidants. A diet including beans provides substantial health benefits, decreasing the risk of heart and renal diseases, protecting against several cancer types. The proteins of common bean have been reported to prevent the activity of HIV reverse transcriptase, thereby slowing the progress of virus. In fact, the niche value of the crop in marginal farming system is more for its quality than the yield *per se*. Therefore, quality improvement has been most important breeding objective of common bean breeders rather to the yield potential *per se*. The protein content of common bean is largely composed of phaseolin and legumin.

Common bean is considered a good source of protein. The major seed storage protein of common bean is phaseolin and it contributes to the nutritional value of common bean seed proteins in a significant way. Many studies have been carried out to assess the variability for protein content. The great majority of samples reported in the literature have given crude protein values ranging from 16-30% dry base matter. Some variability has been found for protein content as related to seed position in the pod and pod position in the plant, but this is minor variability when compared with inter-varietal differences or with interplant variability for the same material (Tulman, 1978). The same range of 16-30% has been found at the Bean Nutrition and Quality Laboratory, CIAT for the 200 samples studied between 1983 to 1988. This value is strongly affected by environmental conditions.

Phytic acid, myo-inositol 1,2,3,4,5,6-hexakisphosphate (Ins P6), is the major (65-85%) storage form of phosphorus in the bean seed (Reddy *et al.*,1989). Phytate is an effective chelator of positively charged molecules and forms stable insoluble complexes with minerals and proteins. Cations bound to phytate are not absorbed in the intestine and are largely excreted, thus reducing the bioavailability of these minerals in the food. This confers phytate its notorious anti-nutritional property having health implications for humans and non-ruminants such as poultry, swine and fish that lack hydrolytic phytase enzyme in their digestive tract (Cheriyan, 1980). Moreover, a major implication to cooking quality of beans is that the breakdown of phytic acid during storage causes HTC (hard-to-cook) phenomenon (Coelho *et al*., 2007). In humans, seed based diets can lead to deficiency of micronutrients, particularly iron and zinc, due to the binding of mineral cations by phytic acid. The effect of phytate in reducing mineral bioavailability in plant foods is an important consideration, it has also been postulated that phytic acid may play a role in reducing cancer risk, possibly because of its antioxidant effects (Graff *et al*., 1990). Specifically, it has been suggested that phytic acid may lower the risk of colon cancer (Harland, 1995) and perhaps breast cancer (Vucenik, 1997). Therefore, complete elimination of phytic acid and other inositol phosphates might not be a good strategy, given their central roles in plant cellular and seed function and their positive health benefits for humans. Maintenance of moderate (~1%) phytic acid and other inositol phosphate levels through appropriate food processing, cooking, and plant breeding might instead be aimed at, thus achieving a balance between the positive and negative aspects of phytic acid (Thavarajah, 2014)

Cooking gives the beans their characteristic taste and tenderness to eat as diet. As in most crops, consumer acceptability in beans is significantly affected by its response to cooking. The cooking of beans may cause several changes in physical, biochemical and nutritional qualities of beans while prolonged cooking may even reduce the nutritive quality of beans. Cooking time and the quality of cooked product are some of the important selection criteria. Cooking time is important in view of the energy requirements associated with cooking and energy being a major issue in developing nations where beans are largely consumed. Reducing carbon footprints through reduced cooking time is a strong ecological rationale for using this trait while developing varieties. In fact the most energy- demanding process in the whole market chain is probably cooking. Even in countries like USA, where agriculture is almost entirely mechanized and production consumes large amounts of energy, 48% of energy in the food chain is spent in industrial processing and home cooking. Cooking common bean has a particularly high energy requirement because of its relatively long cooking time. This has a bearing both on the fuel costs and convenience Poor cooking quality has been related to two textural defects associated with storage of legumes, as typified by the common bean i.e. hard-to-cook defect and hard shell. Hard-to-cook (previously termed sclerema) beans as those that will not soften sufficiently because the soaked seeds do not become tender during a reasonable cooking time, while hard-shell refers to beans that fail to imbibe a sufficient quantity of water during the soaking step (Stanley and Aguilera, 1985). Structurally hard shell is associated with seed coat and failure to absorb water while as hard to cook affects the hydration of cotyledons rendering the cells unable to separate during cooking (Shehata, 1992)

Organolaptic traits also determine the niche value of crops like rajmash. In many parts of India, the rajmash enjoys its niche status mainly on account of its characteristic organolaptic qualities that have shaped its persistence as an important component of farming systems despite the crop having become less competitive due to relegation to low input farming systems. Superior organoleptic characteristics of some traditional common bean landraces (*Phaseolus vulgaris*) have been pointed to as the reason for their persistence in cultivation, despite the advance of new commercial cultivars. Obtaining reliable information on sensory traits is not easy due to the need for panels of tasters and the weak relationship between sensory and chemical traits found thus far.

With regards to the diversity of common bean genetic resources of Jammu and Kashmir for yield components and seed quality traits, no comprehensive study has been done so far. Since common bean is grown in most parts of the state with a wide range of variation in altitude, rainfall, temperature, farming system and socio-economic factors, it is essential to assess the pattern of variations for various traits among and between accessions to resolve the problems in different regions and adaptation zones.

Assessing diversity in these germplasm introductions can help to identify elite genotypes with the greatest novelty in crop improvement programmes. The present study was first of its kind in J&K state in which a core set of 200 lines were evaluated for a large number of morphological and quality traits to identify putative parental lines for developing high yielding varieties with better quality attributes.

MATERIALS AND METHODS

Location

The present study was undertaken during 2013-15 in the research farm of Faculty of Agriculture, SKUAST-K at Wadura (34^o 17' North and 74^o 33 East at altitude of 1594 metres above sea level).

Plant material

The material comprised of 203 lines of common bean belonging to diverse growth habits, maturity and market classes (comprising local landraces a well as accessions procured from national and international gene banks) including 3 checks namely Shalimar Rajmash-1, Shalimar French Bean-1 and Arka Anoop. Shalimar Rajmash-1 and Shalimar French Bean-1 are varieties released by SKUAST-Kashmir and Arka Anoop is a variety released by IIHR, Bangalore. The material was collected from diverse areas of J&K state and represented a diverse market classes defined by seed colour, shape and coat pattern (Figure 1).

Figure 1. Representative diversity of seed shape, colour and shape.

Experimental set up

The material was evaluated in augmented block design (Federer, 1956). The field design consisted of 10 blocks containing 23 genotypes in each with 20 test entries and 3 check entries. Each genotype was represented by a plot size of 2 x 2 meter dimensions with 5 lines. The plants were space planted for optimal expression of traits. Upon harvest the seeds were properly dried and stored for 2 weeks in plastic boxes to equilibrate the moisture content. The samples for various seed quality traits were drawn from these working samples for further processing.

Seed physical parameters

Seed length and breadth

Seed length and breadth was measured using vernier callipers and averaged across 5 representative seeds for each genotype.

Seed dry weight

Seed dry weight was calculated on a randomly drawn sample of 100 sun dried seeds and averaged across 3 samples.

Coat proportion

Seed coat proportion was determined on 20 seeds per accession, as the ratio in weight between coat and cotyledon expressed in percentage, after removing the seed coat from the cotyledons, both after soaking and keeping them for 24h at 105^{o}C.

Bulk density

The bulk density of the bean seeds was calculated using the standard method of Shimelis and Rakshit (2005). 100 g of the sample seeds were transferred to a measuring cylinder, which had 100 ml distilled water at 20ºC. Seed volume (ml/100 g seeds) was obtained after subtracting 100 ml from the total volume (ml). The bulk density was then calculated and recorded in g/ml.

Water absorption traits

Seed water absorption parameters were calculated as per the procedure of Bishnoi and Khetarpaul (1993). The moisture contents of the dry bean samples were equilibrated to each other before analysis of water absorption by storing them for 2 weeks in sealed plastic containers at ambient temperatures and relative humidity.

Water absorption percentage

The percent water absorption was determined by first soaking 30 seeds for 24 h in deionised water at room temperature and dividing the difference in weight before and after soaking by the dry weight of the 30-seed sample. Seed coat proportion was determined on 20 seeds per plot, as the ratio in weight between coat and cotyledon expressed in percentage, after removing the seed coat from the cotyledons, both after soaking and keeping them for 24h at 105^{o}C.

Swelling coefficient

Swelling coefficient was determined using the Youssuf's method (1978). The swelling coefficient was calculated as the percentage increase in volume of beans after soaking:

Swelling coefficient (%) = $(V\,a/\,V\,b) \times 100$

Where *Vb*, volume of bean seeds before soaking (ml); *Va*, volume of bean seeds after soaking (ml).

Hydration coefficient

Hydration coefficient was determined using the Youssuf's method (1978). The raw bean seeds were soaked in distilled water for 24 hours and the volume of the bean seeds was estimated before and after soaking by determination of displaced water. The hydration coefficient was calculated as the percentage increase in weight of beans.

Hydration coefficient (%) = $(Ma/\,Mb) \times 100$

Where *Mb*, weight of seeds before soaking (g); *Ma*, weight of bean seeds after soaking (g);

Swelling capacity

Seeds, weighting 100 g, were counted, their volume noted and soaked overnight. The volume of soaked seeds were noted in a graduated cylinder (Bishnoi and Khetarpaul, 1983). Swelling capacity (Sc) was calculated as change in volume per number of seeds.

Swelling capacity (ml/seed) = $(Va - Vb)/N$

Hydration capacity

Seeds, weighting 100 g, were counted and soaked overnight. After the water was drained, the soaked seeds were blotted dry and weighted. Hydration capacity (Hc) was calculated as change in weight per number of seeds.

Hydration capacity (g/seed) = $(Ma - Mb)/N$

Hard shell percentage

Hard shell percentage was done as per the method of Correa *et al*. (2010). The samples were washed and immersed for 8 hours in distilled water and the seeds that did not absorb water were counted. These grains were visually verified for shell wrinkling and those without wrinkle were treated as hard shell. The result was expressed as hard-shell percentage (without water-holding capacity).

Biochemical traits

Protein content

Protein content was estimated using Near Infrared Reflectance Spectroscopy (CROP SCAN 2000G) facility at Seed Technology Laboratory of SKUAST-K. It is a quick and non-destructive method for estimation of protein content.

Phytic acid

Phytic acid content was estimated according to modified Haug and Lantzsch (1983) method. The determination was based on indirect spectrophotometric determination of phytic phosphorus in dry bean extracts. Phytic acid was precipitated by addition of ferric ammonium sulphate. Part of iron forms insoluble ferric phytate, and the remaining iron was determined spectrophotometrically. Calibration curve was prepared by series of standard solutions of sodium salt of phytic acid. All reagents were of analytical grade procured from HIMEDIA. 0.5 g of powdered sample was extracted with 100 ml of 2.4% HCl during 3 h with constant stirring. The extract was filtered through Whatman No 41 filter paper and 0.5 ml of extract was transferred into a glass tube with stopper, ammonium iron (III) - sulphate solution (0.2 g of $NH_4Fe(SO_4)_2.12H_2O$ dissolved in 100 ml of 2 mol/l HCl and filled to mark with distilled water) was added. Closed glass tube was held in boiling water bath for 30 min., cooled in refrigerator for 15 min. and left to attain room temperature. Tube was centrifuged at 3000 r/min. One ml of supernatant was transferred to another glass tube and 1.5 ml of 2,2'-bipyridine solution (10 g 2,2'-bipyridine dissolved in 10 ml thioglycolic acid and filled to mark with distilled water) was added. After exactly defined time, absorbance was measured at 519 nm.

Cooking and Sensory evaluation

Cooking time score

The procedure of cooking in an autoclave followed the method described byRevilla and Vivar-Quintana (2008), with modifications. Fifty soaked grains were placed in glass beaker, filled with 200 ml of distilled water, covered with watch glass, and cooked under the conditions 110°C for 5 min. Following scale was used for scoring cooking properties of bean genotypes. The softness/hardness (cookability) of the beans was determined subjectively by pressing the cooked beans between the thumb and forefinger (Vindiola *et al*., 1986).

Scale	Designation	Description
1	Undercooked	Grain is difficult or not able to smash and cotyledon feels hard
2	Slightly undercooked	Grain is less difficult to smash and cotyledon feels slightly hard
3	Average cooked	Grain is firm but smashes easily and cotyledon feels soft
4	Slightly overcooked	There is little resistance to smash grain and cotyledon feels mushy
5	Overcooked	Grain is easily pressed into a mush

Sensory evaluation

Sensory evaluation of beans was done as per the method of Calvo and del Rey (1999) and Casanas *et al.* (2006) with slight modifications. For cooking 100 grams of beans were soaked in 500 ml low mineralised water for 12 hours at room temperature. The beans were cooked on low heat until done and one gram salt was added to each sample. The sample was served to a panel of 14 panellists including a sensory evaluation expert, faculty members as well as students. Before the sensory evaluation test was initiated, all the panellists were apprised of basic protocol for evaluation of sensory traits using different cooked samples. The parameters included visual appearance, colour, flavour, taste (sweetness), texture (seed coat roughness and whole seed creaminess) as well as overall acceptability measured on a scale of 0-4.

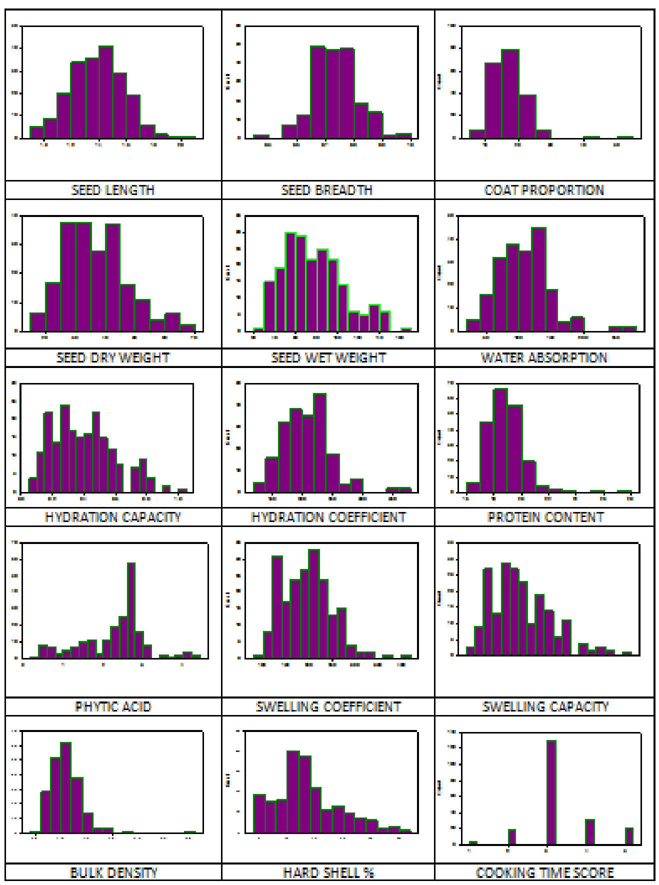

Figure 2. Frequency distribution of 15 seed quality traits in 203 common bean genotypes.

Statistical analysis

The quantitative traits were analysed for various statistical parameters viz. mean, range, variances, correlations etc. The frequency distribution (Figure 2) was done by Sigma Plot 11.0 (SYSTAT). Phenotypic and genotypic coefficients of variation (PCV and GCV) for each trait were computed as PCV = (σ^2P/ mean) x 100, GCV = (σ^2G/ mean) x 100 as per (Burton, 1952) and categorized the range as per Sivasubramanian and Madhavamenon (1978). Broad sense heritability was estimated as h^2 (bs) = (σ^2G / σ^2P) x 100 as per Lush (1940) and further classified into low, medium and high as per Robinson (1996). Expected genetic advance was computed as EGA = k x (σ^2G / σ^2G) x σ^2P as per Johnson *et al*. (1955). Here the standard value of k is taken as 2.06 at 5% selection intensity; σ^2G is genotypic variance; and σ^2P is phenotypic variance. Genetic advance was expressed as % of mean as GA (%) = (EGA/mean) x 100. The significance of variances was tested at the 5% probability level. The genetic diversity to find out genetic similarity/dissimilarity and principal component analysis (PCA) was done using the statistical software SAS 9.1 (SAS Institute USA).

RESULTS AND DISCUSSION

Frequency distribution and descriptive statistics for seed physical, water absorption and biochemical traits

The material was evaluated in the field for 22 descriptor traits (data not presented). Even though all colour classes were represented for seed coat colour, about 36.45% were red followed by brownish (27.59%) whereas seeds representing other classes were less than 10%. Most of the genotypes were having plain seed coat (44.83%) followed by mottled ones (38.42%) and specked (10.34%) ones. Seed shapes were kidney (43.35 %), followed by cuboidal (38.43%) and oval (17.73%). Figure 1 shows some of the representative diversity classes of germplasm used in present study in terms of seed colour, shape and size. In terms of seed size about half of accessions were medium (48.77%), while remaining were large (33.99%) and small (17.24%). Frequency distributions graphs for seed quality traits depicted in the Fig. 2. showed that 87.68% accessions had seed length in the range of 1.1-1.6 cm. Seed breadth was in a narrow range and about 70% had seed breadth in the range of 0.7-0.8 cm as was case with protein content, phytic acid and cooking time score (93.59% in range of 17-21%, 66.50% in the range of 2-3.2% and 63.05% with a score of 3 respectively). Seed dry weight, wet weight, water absorption percentage as well as traits related to hydration and swelling had broader range of variation with about 80-90% genotypes spread across a broad range. Similar results have been reported in the Andean and Mesoamerican gene pools of common bean (Rana*et al*., 2015). These results are consistent with findings of different scientists who have reported wide variation in seed shape and size in bean germplasm (Rai*et al.,* 2006; Cabral *et al.* 2010; Lioi *et al*. 2012). The most elaborated work done by Singh (1989) and Singh *et al*. (1991) have given high taxonomic value for seed size, colour and shape in comparison to the vegetative characteristics of plant in common bean.

The results pertaining to descriptive statistics of seed physical, water absorption and biochemical traits are presented in Table 1. There was substantial variability indicated by a broad range of variation in the traits, except seed breadth where the range was comparatively narrower, that provides ample opportunities for selection towards desirable direction for these traits. The range of variation recorded for various seed quality traits was seed length (0.89-2.08 cm), seed breadth (0.47-1.00 cm), coat proportion (4.55-28.90%), bulk density (0.463-4.722 g/ml), seed dry weight (13.34-68.87 g), seed wet weight (20.30-166.93 g), water absorption percentage (21.80-294.01%), swelling coefficient (103.89-456.917), hydration coefficient (121.80-394.01), swelling capacity (0.017-1.132), hydration capacity (0.048-1.038), hard shell percentage (5.02-30.01%), cooking time score (1-5), protein content (16.08-28.02%), and phytic acid (0.385-4.95%). Sofi *et al.* (2014) has also reported wide variation in seed physical and water absorption traits in common bean genotypes. Seed traits including the size, shape, and colour and coat pattern are important parameters that determine consumer acceptability of common bean and therefore have received considerable attention in all breeding programmes. In Kashmir valley, there is a greater acceptability for red coloured beans with kidney or cuboid shapes. Therefore, material in this study was purposefully drawn from the germplasm bank in light of the varietal attributes specified by farmers during the participatory rural appraisal conducted by the University for identification of farmers' acceptability criteria for common bean (Umar Gull, 2015). In fact, seed traits determine the ultimate market value of common bean and in view of greater demand for pulses as well as the market orientation, it is imperative to identify the genotypes that could meet the aspirations of small holding farmers and fetch better market values for livelihood enhancement. In terms of consumer acceptability, physical traits of seeds are of paramount importance. While the length, breadth and weight of seeds determine the general consumer preference, coat proportion is implicated in determining the limits of water absorption thereby influencing cooking properties. Moreover, this trait also has implication in development of hard shell and hard-to-cook (HTC) traits in common bean; while as bulk density determine the weight per unit volume or the packing density of carbohydrates in seed which influences 100-seed weight. A large number of studies have been done wherein the natural variation for seed physical parameters has been studied (Lioi and Piergiovanni ,2013; Sofi *et al.*, 2014). There is also a general understanding that beans absorbing water at high rate and quantities take less time for cooking .In this study, we identified a large number of genotypes that not only absorbed substantial amounts of water but also underwent significant increase in volume Substantial natural variation for water absorption traits has also been reported in common bean (Nicri *et al.*, 2014, Wani *et al.*, 2014 and Sofi *et al.*, 2014). .Genotypes identified on the basis of desirable trait attributes such as seed length (WB-969), seed breadth (WB-969), seed dry weight (WB-441), coat proportion (WB-256),seed wet weight and hydration capacity (WB-431), water absorption and hydration coefficient (WB-262), protein content (WB-901), phytic acid (WB-491), swelling coefficient and capacity (WB-261), hard shell (WB-435) and bulk density(WB-431) can be used in breeding programmes for trait based improvement for seed physical traits.

Variability parameters of seed physical, water absorption and biochemical traits

The variability parameters for seed physical, water absorption and biochemical traits (Table 1) revealed significant differences among the genotypes studied by significant values of σ^2g. The GCV and PCV values were higher for traits like seed wet weight, water absorption, swelling capacity, and hydration capacity and hydration coefficient. The heritability parameters were invariably high for all traits except bulk density (0.45 g/ml) and as high as 97% for traits like seed length, hydration coefficient, seed wet weight and swelling coefficient. The genetic advance as percentage of mean was higher (>35%) for most of the traits except seed length, seed breadth, bulk density and hard shell where it was low (<35%). For improvement of any set of traits, high mean, high heritability, broad range and high values of GCV and genetic advance are desirable. In this study, desirable attributes of variability parameters were recorded for seed quality traits and as such these traits can be improved using appropriate selection methodologies. Response to selection and success of hybridisation in common bean primarily depends on the nature and magnitude of genetic diversity present in the germplasm used (Rana *et al.*, 2015). Mavromatis (2012) made a comprehensive study on water absorption traits in common bean and concluded that the trait can effectively be used for comparing large germplasm sets for cooking quality as the varieties having high hydration and swelling capacity cook faster. However, the lines that have low hydration capacity have longer storage life (Castillo *et al.*, 2008).

Table 1. Descriptive statistics and variability parameters for seed physical traits.

Trait	Mean	SE mean	Range	σ^2 g	GCV	PCV	h^2	GA as % of
Seed length	1.393	0.018	0.89-2.055	0.077**	13.995	14.174	0.97	28.45
Seed breadth	0.738	0.017	0.485-0.965	0.013**	10.49	11.33	0.85	20.017
Coat proportion	16.992	0.836	5.218-51.843	57.109**	26.1787	26.8271	0.95	52.624
Seed dry weight	37.020	1.994	15.590-66.75	223.932**	28.07	29.086	0.93	55.806
Bulk density	1.094	0.191	0.587-3.468	0.060**	22.483	33.398	0.45	32.463
Seed wet weight	77.389	3.390	24.190-163.010	1639.551**	36.736	37.255	0.97	74.624
Water absorption percentage	106.956	6.044	22.536-270.210	3314.217**	37.638	38.477	0.95	75.844
Hydration co-efficient	206.956	6.046	122.536-370.210	3314.218**	19.451	19.885	0.95	39.195
Swelling co-efficient	195.456	15.201	98.713-413.473	2580.212**	25.988	28.219	0.85	51.811
Hydration capacity	0.404	0.018	0.063-1.022	0.080**	49.194	49.813	0.97	100.019
Swelling capacity	0.316	0.024	0.020-0.920	0.030**	55.442	56.343	0.97	118.05
Hard shell	13.516	2.452	5.020-30.010	25.8409	37.604	45.520	0.68	4.085
Protein content (%)	18.881	0.695	16.595-27.350	3.674**	6.164	8.066	0.58	9.703
Phytic acid content (mg/g)	2.299	0.299	0.385-4.285	1.287**	32.375	37.24	0.75	57.981

Table 2. Cluster means for seed quality traits in seven clusters in 203 common bean lines.

Trait	I	II	III	IV	V	VI	VII
Seed length	1.38	1.38	1.59	1.40	1.31	1.21	1.32
Seed breadth	0.75	0.70	0.79	0.75	0.73	0.67	0.74
Coat proportion	16.34	15.92	15.41	17.01	19.32	17.64	14.85
Seed dry weight	37.14	36.03	47.14	41.15	31.63	26.03	25.74
Seed wet weight	77.55	83.93	113.65	74.45	54.01	43.58	92.37
Water absorption %	109.25	134.18	142.29	80.55	69.84	67.64	259.55
Hydration Coefficient	209.25	234.18	242.29	180.55	169.84	167.64	359.55
Hydration Capacity	0.40	0.48	0.67	0.33	0.22	0.18	0.67
Protein content	19.17	19.12	19.12	18.17	18.73	18.60	19.42
Phytic acid	2.27	2.19	2.13	2.46	2.38	2.59	2.21
Swelling coefficient	184.13	236.50	147.20	133.46	211.06	303.28	220.91
Swelling capacity	0.29	0.45	0.20	0.14	0.35	0.53	0.38
Bulk density	1.12	1.14	12.30	1.05	1.04	1.10	0.78
Hard shell %	12.74	11.63	1.15	19.13	13.89	14.35	5.75
Cooking time score	3.22	3.56	12.30	3.08	2.85	3.00	2.75

Table 3. Non-rotated component loadings (values of principal component traits of common bean).

TRAIT	PC 1	PC 2	PC 3	PC 4	PC 5
SL	0.330	-0.138	0.128	0.018	0.176
SB	0.244	-0.258	0.097	-0.018	0.011
CP	-0.169	-0.063	-0.179	0.031	0.573
SDW	0.357	-0.296	0.155	0.175	-0.007
SWW	0.439	-0.048	0.080	0.072	0.054
WA	0.328	0.393	-0.081	-0.155	0.082
HCO	0.328	0.393	-0.081	-0.155	0.082
HCA	0.441	0.087	0.033	0.010	0.082
PROTEIN	0.012	0.175	-0.345	0.485	0.196
PA	-0.022	-0.121	0.312	-0.166	0.607
SCO	-0.169	0.391	0.393	0.383	0.010
SCA	-0.043	0.311	0.618	0.196	-0.034
BD	0.099	-0.090	-0.234	0.653	0.085
HARDSHELL	0.008	-0.435	0.262	0.191	-0.191
Cooking	0.174	0.092	-0.130	0.024	-0.404

Multivariate analysis for genetic diversity assessment for seed quality traits

Based on the principal component analysis performed on 15 seed quality traits, all the 203 accessions were grouped into 7 clusters regardless of geographic origin and each cluster was found to have varied number of accessions (Table 2). The number of accessions falling in each cluster were highest (52) in cluster 5 (C5) followed by C2 (39), C3 (38), C1 (32), C4 (26), C6 (12) and C7 (4). The mean value of accessions grouped into each cluster (Table 4) showed that accessions in C3 had highest cluster means for 9 out of 15 seed quality traits namely seed length (1.59 cm), seed breadth (0.79 cm), seed dry weight (47.13), seed wet weight (113.64), water absorption (142.28), hydration coefficient (242.28), bulk density (1.15) and cooking time score (3.63). This cluster also had lower cluster mean of 2.12% for phytic acid. The cluster C7 had lowest value of coat proportion (14.85%). The cluster C6 had highest cluster mean for swelling coefficient and swelling capacity (303.28 and 0.52 respectively). Similarly the highest cluster mean for hydration capacity and protein content were recorded for C7 (0.66 and 19.41 respectively), while as the genotypes with higher mean of hard shell were clustered in C4 (19.12). Rana *et al.* (2015) characterised 4274 accessions of common beans held in NBPGR for physiological and morphological traits and found that the entire collection could be grouped into 10 genetically diverse clusters, irrespective of origin, based on multivariate analysis.

The criteria followed for selecting the principal components to be included in further analysis was based on Eigen values of principal components (Kovacic, 1994). The fact that Eigen values are above unity indicates that the evaluated principal component weight is reliable (Mohammadi and Prassanna, 2003), The PCA was used to eliminate the redundancy in data set and revealed that all the 15 seed quality traits loaded on first five components accounting for about 70.40% variance in bean germplasm under study. The latent roots ranged from 4.824 for first PC to 1.013 for the fifth PC. The first component (Figure 3) explained 32.10% of variation through seed length, seed dry weight, seed wet weight, water absorption, hydration capacity and hydration coefficient followed by PC2 (15.60%) through water absorption, hydration coefficient, swelling coefficient and swelling capacity, PC3 (11.20%) through swelling coefficient and swelling capacity, PC4 (8.40) through protein content, swelling coefficient and bulk density and PC5 (6.70%) through coat proportion and phytic acid (Table 3 and 4). In this study hierarchical cluster analysis showed that some of accessions collected from various regions were grouped into the same cluster, while many others fell into different clusters. Grouping was not associated with the geographic distribution; instead accessions were mainly grouped due to their morphological differences. Thus, it cannot be generalized that all the accessions having same origin would always have low diversity among them. Results of the present investigation agreed with previous studies using morphological data to characterize germplasm of Ethiopian mustard (*B. carinata*) in which geographic origin of the collected material had no effect on grouping of the accessions/varieties (Dhillon *et al*., 1999). Sofi *et al.*(2014) studied the variability in common bean using multivariate analysis and found that more than 70% of variation was accounted for by only 4 principal components with days to flowering, days to maturity, 100-seed weight, pod length and seeds per pod as most important traits identified by principal components. They concluded that the combined use of these PCs can be successful for identification of genotypes for economically important traits. Similarly, Rana *et al.* (2015) characterised 4274 accessions of common beans and found that first 3 principal components explained 80% of the total variation which was contributed mainly by pod length, seed length, seed width, pods per plant and 100-seed weight. A large number of accessions were also found to be resistant to different diseases under field conditions, especially anthracnose. Abdolahhi *et al.* (2016) evaluated 64 Common bean (*Phaseolus vulgaris* L.) genotypes for phenological and morphological traits and reported 4 important factors which accounted for 74% of the total variation.

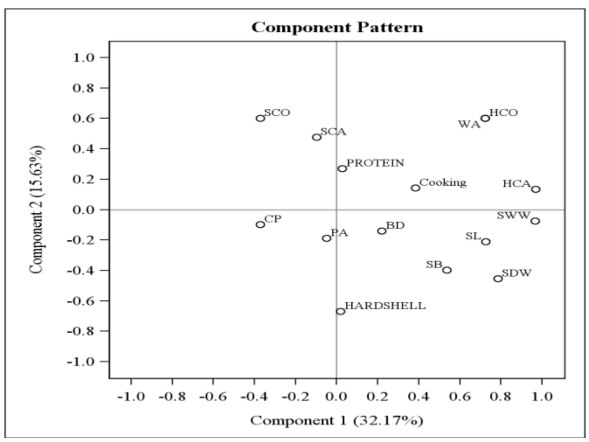

Figure 3. Biplot of different variables loaded on PC1 and PC2.

Correlation matrix for seed quality traits

In this study, positive and significant correlations (Table 5) were found in case of seed length with seed breadth, seed dry weight, seed wet weight, water absorption percentage, hydration coefficient, hydration capacity and cooking time score; seed breadth with seed dry weight, seed wet weight, water absorption percentage, hydration coefficient, hydration capacity and hard shell percentage; seed dry weight with seed wet weight, water absorption percentage, hydration coefficient, hydration capacity and cooking time score; water absorption percentage with hydration coefficient, hydration capacity and cooking time score; hydration coefficient with hydration capacity and cooking time score; protein content with bulk density; swelling coefficient with swelling capacity ;and bulk density with cooking time score. Negative and non-significant correlations were seen in case of seed length with coat proportion, swelling coefficient and swelling capacity; seed breadth with coat proportion and swelling coefficient; coat proportion with seed dry weight, seed wet weight, water absorption percentage, hydration coefficient, hydration capacity and cooking time score; seed dry weight with swelling coefficient; seed wet weight with swelling coefficient; water absorption percentage with hard shell percentage; hydration coefficient with hard shell percentage; hydration capacity with swelling coefficient; protein content with phytic acid content and hard shell percentage; swelling coefficient with hard shell percentage; and in case of swelling capacity with bulk density.

Significantly positive correlations seen between water absorption parameters with cooking time score reinforce our existing evidence that these traits can be effectively used for screening Rajmash for cooking quality time (Correa *et al*., 2010; Balcha *et al*., 2010; Mavromatis *et al*., 2012; Nciri *et al*., 2014). Similarly, most of the water absorption parameters were negatively correlated with coat proportion which is quite understandable on account of the fact seed coat has significant bearing on the amount and time of water absorption upon soaking. Similarly, water absorption traits had negative correlation with hard shell percentage which is again in confirmation of the existing evidence that the relative amount of seed coat does not solely influence water absorption, but the permeability of the coat and its adhesion with cotyledons are also implicated.

In terms of cooking time score, six genotypes were easy to cook (WB-6, WB-160, WB-216, WB-222, WB-257 and WB-112) whereas only one genotype WB-242 was hard to cook. Overall acceptability score (Table 6) based on traits such as colour, texture, taste, flavour etc was highest in case of WB-401 (3.42) followed by WB-222 (3.37), WB-1318 (3.36) and WB-185 (3.34) whereas low values were recorded for WB-956 (2.22) and WB-160 and WB-249 (2.26). The overall acceptability score for white genotypes was low despite high water absorption values owing to the fact that white seeded beans invariably lost their integrity on account of rupture of seed coat. Variability in seed physical traits and culinary traits has also been reported by various workers (Santalla *et al*., 1999 and Vakali *et al*., 2009) in common bean using coat proportion and water absorption as indicative traits.

Table 4. Latent squares (Eigen values) of principal components for seed quality traits.

Principle Component	Eigen Value	% Variance	Cumulative Variance
PC 1	4.824	32.10	32.10
PC 2	2.345	15.60	47.70
PC 3	1.688	11.20	58.90
PC 4	1.267	8.40	67.30
PC 5	1.013	6.70	74.00

CONCLUSION

Common bean is an important niche crop of Kashmir valley of India and an important component of subsistence farming comprising low input marginal farmers. There is growing evidence about the livelihood and health benefits of this crop and is sometimes designated as nearly complete food. This two study reports one of the first large-scale characterisation of the bean germplasm in the Jammu and Kashmir for seed quality traits. The germplasm presented wide range of genetic variability for 16 traits among 203 accessions. Among different traits, seed traits have been found as the most predominant one which is most important in common bean and major determinants of commercial acceptability of varieties. Seed traits have also been considered highly heritable traits, therefore important in breeding programmes (Blair *et al*., 2010). There was marked variation in seed coat colour, coat pattern, shape and size and cooking quality that are in favour of need of common bean growers of Kashmir valley of India.

Table 5. Correlation matrix for 15 seed quality traits in common bean.

TRAIT	SL	SB	CP	SDW	SWW	WA %	H CO	H CA	PROT	PA	S CO	S CA	BD	HS	CT
SL	-	0.359*	-0.188	0.661**	0.691**	0.344*	0.344*	0.641**	-0.049	0.108	-0.293	-0.035	0.126	0.137	0.184
SB		-	-0.168	0.570**	0.518**	0.140	0.140	0.441*	-0.100	0.009	-0.314 *	-0.116	0.081	0.183	0.02
CP			-	-0.228*	-0.288*	-0.274*	-0.274*	-0.292 *	0.048	0.059	0.034	-0.112	0.004	-0.086	-0.145*
SDW				-	0.881**	0.192	0.192*	0.735**	-0.079	0.021	-0.396*	-0.040	0.260*	0.361*	0.197*
SWW					-	0.611**	0.611**	0.968**	0.007	-0.016	-0.325 *	-0.003	0.220 *	0.095	0.307*
WA %						-	1.000**	0.775**	0.134	-0.102	-0.031	0.060	0.017	-0.354*	0.280*
H CO							-	0.775**	0.134	-0.102	-0.103	0.060	0.017	-0.354*	0.280 *
H CA								-	0.052	-0.034	-0.256*	0.016	0.178	-0.055	0.337*
PROT									-	-0.200*	0.083	-0.088	0.255*	-0.165*	-0.052
PA										-	0.025	0.092	-0.049	0.118	-0.11
S CO											-	0.804**	0.040	-0.162	-0.094
S CA												-	-0.249*	-0.025	-0.073
BD													-	0.067	0.168
HS														-	-0.092
CT															-

SL=seed length, SB=seed breadth, CP=coat proportion, SDW= seed dry weight, SWW=seed wet weight, WA%=water absorption percentage, HCO=hydration coefficient, HCA=hydration capacity, PROT=protein content, PA=phytic acid, SCO=swelling coefficient, SCA=swelling capacity, BD=bulk density, HS= hard shell, CT=cooking time score

Table 6. Mean performance of sensory traits in 30 genotypes of common bean.

Genotype	Visual appearance	Colour	Flavour	Taste	Texture	Overall acceptability
WB- 6	3.10	2.73	3.95	3.55	3.12	3.29
WB -22	2.48	2.57	3.37	2.33	3.55	2.86
WB -83	2.95	3.20	2.28	2.50	2.37	2.66
WB -112	2.31	2.35	2.75	2.71	2.74	2.57
WB -115	3.27	3.55	3.05	2.85	3.35	3.21
WB -160	2.46	2.40	2.21	2.18	2.06	2.26
WB -185	3.15	2.95	3.47	3.55	3.57	3.34
WB -195	2.55	2.67	2.57	2.62	2.87	2.66
WB -216	2.77	2.58	2.56	2.77	2.55	2.65
WB -222	3.05	3.30	2.75	2.80	3.05	2.99
WB -242	3.35	3.52	3.37	3.47	3.16	3.37
WB -249	2.45	2.50	2.28	2.00	2.56	2.36
WB -257	2.75	2.70	2.90	3.10	3.05	2.90
WB -341	3.29	3.19	2.64	2.75	2.18	2.81
WB -401	3.50	3.55	3.38	3.45	3.20	3.42
WB -467	2.83	2.73	2.60	2.82	2.92	2.78
WB -651	3.20	3.30	3.13	3.05	2.71	3.08
WB -956	1.95	1.80	2.20	2.85	2.30	2.22
WB -1131	2.20	2.53	2.35	2.60	2.45	2.43
WB -1282	3.24	3.12	2.96	3.28	3.25	3.17
WB -1314	3.10	3.31	2.82	2.75	2.87	2.97
WB -1318	3.48	3.48	3.37	3.27	3.22	3.36
WB -1446	2.75	2.71	2.94	2.85	2.87	2.82
WB -1492	2.82	2.55	2.61	2.35	2.50	2.57
WB -1587	2.50	2.53	2.50	2.35	2.74	2.52
WB-1634	2.84	3.05	3.30	3.60	3.05	3.17
WB- 1643	2.60	2.27	3.00	2.53	3.00	2.68
Sh. Rajmash-1	3.00	2.96	2.61	2.53	2.60	2.74
Sh. French Bean-1	2.82	2.60	2.46	2.31	2.50	2.54
ArkaAnoop	2.37	1.95	2.33	2.61	3.15	2.48

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