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Biochemical composition and storage protein profiling of mungbean (*Vigna radiata* L. wilczek) cultivars

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Abstract
The experimental trial on 12 mungbean cultivars were conducted and the findings of the pooled data of two years revealed that biochemical composition viz. moisture, total carbohydrate, crude protein, soluble protein, methionine, crude fat, calorific value, total phenol and ash content varied highly significantly across the tested cultivars and ranged from 8.74 to 9.96%, 61.18 to 63.47%, 23.99 to 26.15%, 15.83 to 19.72%, 0.79 to 1.76 g/16gN, 1.12 to 1.62%, 347.93 to 360.19 kcal/100g, 62.35 to 89.61mg/100g and 3.42 to 4.01% with an overall mean of 9.61%, 62.55%, 25.08%, 17.97%, 1.26g/16gN, 1.37%, 353.65kcal/100g, 73.04mg/100g and 3.67%, respectively. SDS-PAGE of seed storage proteins of 12 cultivars of mungbean led to detection of 26 polypeptide bands with molecular weights of the resolved peptides ranged from 104 kDa to 18 kDa. Dendrogram based on electrophoretic data grouped the 12 cultivars into two major clusters, cluster I and cluster II. In the present study, a peptide band of 18 kDa was detected on SDS-PAGE that may be 11 S globulin subunit, peptides with molecular weights 28 kDa was also detected on SDS-PAGE that may be basic 7S subunit, while peptide with molecular weight 23kDa may be 11S globulin and peptide with molecular weight 32 kDa and 25 kDa peptide might be 8S vicilin subunit according to earlier reports.

Keywords: biochemical composition, cluster, dendrogram, SDS-PAGE, storage protein and mungbean

Introduction
Pulses are most important constituents in human diet of large number of people which help to supplement cereal diets, improving their nutritive values. Pulses are deficient in sulphur containing amino acids *viz*. methionine and cysteine while rich in lysine. Pulses also supply vitamin-B, minerals and fats (Khan and Dixit, 2001) [21]. Pulses are the main source of protein in primarily vegetarian Indian diet. Besides proteins, pulses are also good sources of vitamins, minerals, ω-3 fatty acids and dietary fibre or non-starch polysaccharides. Grain legumes are being cultivated in India since time immemorial. They have high total protein content (20-26%) and can be considered as a natural supplement to cereals.

Mungbean (*Vigna radiata* L. Wilczek), also known as green gram, belongs to family Fabaceae, subgenus *Ceratotropis* in the genus *Vigna* and is a self-pollinating diploid grain legume (2n = 22) with a genome size of 560 Mb (Arumuganathan and Earle, 1991) [8]. It is a major source of dietary protein for the predominantly vegetarian population of India. It is an excellent source of easily digestible protein, which causes low flatulence and complements the staple rice/ wheat diet in Asia.

Mungbeans are rich sources of dietary fiber, carbohydrates, energy, vitamins and minerals *viz.* iron, magnesium, phosphorus, potassium, copper and folate while riboflavin and niacin are found in trace amount (Khalil, 2006; Charmaine, 1998) [20, 11]. It is also rich in lysine, 5.24-5.85 g/100 g of protein (Adel *et al*., 1980) [2], but deficient in methionine in contrast with the high methionine value observed for rice bean (Andersen, 2007) [6].

The seed storage proteins on the other hand, are non-enzymatic and have the sole purpose of providing proteins (nitrogen and sulphur source) required during germination and establishment of a new plant. Albumins and globulins comprise the storage proteins of dicots, whereas prolamins and glutelins are major proteins in monocots. Seed protein patterns obtained by electrophoresis have been successfully used to resolve the taxonomic and evolutionary problems of several plants (Pervaiz *et al*., 2011; Shah *et al*., 2011; Emre, 2011) [33, 39, 13]. Analysis of seed protein can also provide a better understanding of genetic affinity of the germplasm (Shah *et al*., 2011) [39]. This method can also be used as a promising tool for differentiating the cultivars of a particular plant species. SDS-PAGE is also considered to be a practical and reliable method for species identification because seed storage proteins are

largely independent of environmental fluctuation. Polymorphism in seed storage proteins has been associated with geographical origin (Ghafoor *et al.,* 2002) [14]. Seed storage protein analysis represents a valid alternative and/or improved approach to varietals identification. The SDS-PAGE proved to be a powerful tool for differentiating *Vigna radiata* and *Vigna mungo*; whereas a low level inter specific genetic diversity was observed and no clear differentiation was observed both for agronomic characteristics and for geographical origin (Ghafoor *et al.,* 2002) [14].

The biochemical composition of mungbean has been studied by a number of workers. Since the composition is influenced by genetical constituents, cultural practices and environmental factors. It is, therefore, important to evaluate the local grown cultivars in order to assess their nutritional quality.

Keeping in the view of above facts, the present investigation was undertaken to determine the biochemical composition as well as storage protein profiling of twelve mungbean cultivars grown under rainfed condition in Chitrakoot Region.

Materials and Methods

The mungbean seeds of 12 released varieties viz. Samrat, BPMR-145, HUM-2, MUM-2, Pairy Mung, PKV AKM-4, Pant M-2, Pant M-4, Pusa-0672, Pusa-9072, RMG-62 and RMG-268 were obtained from Indian Institute of Pulses Research (ICAR), Kalyanpur, Kanpur (U. P.). The experiment was laid out in Randomized Block Design (RBD) with three replications for each cultivar during Kharif season, 2013-14 and 2014-15 at Agricultural Farm, Rajoula, M.G.C.G.V.V, Chitrakoot, Satna (M.P.). The biochemical analysis of undehulled seed materials were done in triplicates for both the year. The data presented in Table-1, represent pooled mean of two years.

The moisture content was determined by the method of AOAC (1970) [1]. Total carbohydrate was determined by Anthrone method as described by Hedge and Hofreiter, (1962) [17]. The nitrogen content of mungbean seed samples were estimated by Micro-Kjeldhal Method (AOAC, 1970) [1] and crude protein content was determined by multiplying the total nitrogen per cent by the factor 6.25. The soluble protein content in mungbean was determined by following the procedure given by Lowry *et al.* (1951) [24]. Methionine content was determined by the procedure of Horn *et al*. (1946) [18]. The crude fat was determined from oven dried finely ground sample by the Soxhlet extraction apparatus using petroleum ether of B.P. 60-80ºC (AOAC. 1970) [1]. Calorific values of mungbean seed samples were determined with the help of the Bomb Calorimeter. Total phenol content in mungbean seed was determined as procedure laid down by Malik and Singh (1980) [25]. The ash content was determined as described in AOAC method (1970) [1].

The experimental data were analyzed following standard statistical procedure of CRD (Completely Randomized Design) experimental design as per procedure laid down by Panse and Sukhatme (1978) [31].

Seed protein extraction: For extraction of soluble proteins, 30 mg seeds were grounded in 50 mM phosphate buffer (pH 7.8) and centrifuged in micro-centrifuge machine for 10 min at 14,000rpm. The supernatant was separated and used for protein profiling. Protein concentration of extracts was measured by Lowry method (1951) [24]. 60 µl protein samples were mixed with 40 µl of loading dye (10 ml containing 0.2g SDS, 0.001g bromophenol blue, 0.5 ml β-mercaptoethanol, 1.25ml 0.5M Tris, pH 6.8, 2.5 ml glycerol and 5.75 ml) on Vortex and heated at 95°C for 5-7 minutes to denature the proteins.

Seed protein profiling: Proteins profiling of samples was performed using SDS polyacrylamide gels on a Bio-Rad mini gel electrophoresis apparatus using discontinuous system (4 % stacking gel and 10% resolving gel) according to the procedure of by Laemmli (1970) [23]. Equal quantities (25µg) of each samples along with protein molecular marker were loaded into well with the help of micropipette, after proper loading the electrophoresis unit was connected with power supply and run at constant voltage (100 V). When the tracking dye reached of the end of running gel after complete separation of molecules, power supply was turned off.

Staining & Destaining of gel: The gel was gently removed from the space between the plates, immersed in the staining solution (CBB, R-250) contained in a tray and kept it for overnight. The gel was destained by putting it into the destaining solution (45 ml methanol and 10 ml acetic acid and adjusts volume up to 100 ml with double distilled water). The process was continued until the back ground was colourless.

Gel documentation and analysis: Gel was photographed using Gel documentation system (Bio-Rad). Electrophoregrams for each variety was scored and the presence (1) or absence (0) of each band noted. Presence and absence of bands were entered in a binary data matrix. The dendrogram was constructed using Unweighted Paired Group Method with Arithmetic means (Sneath and Sokal, 1973) [41] employing Sequential Agglomerative Hierarchic and Non-overlapping clustering (SAHN). All analysis was carried out using a statistical package NTSYS-pc, version 2.02e (Rohlf, 1997) [35].The coefficient scale of dendrogram indicated the dissimilarity coefficient among cultivars. Subunit molecular weight was estimated by using the protein molecular weight marker ranged from 10.5 to 250 kDa.

Result and Discussion

The result of variance analysis of data revealed that all the biochemical parameters varied highly significantly amongst cultivars. The results of all the biochemical parameters are presented in Table1. Moisture content is important factor in determining the total dry matter content. The perusal of data revealed that moisture per cent showed genetic variability and ranged from 8.74 to 9.96 per cent with grand mean of 9.61 %. The maximum and minimum value for moisture per cent were found in Pusa-0672 (9.96%) and Samrat (8.74%), respectively. This finding is in conformity with Mubarak (2005) [30] and Blessing and Gregory (2010) [9]. Carbohydrates provide the majority of dietary calories for animals including human being. The total carbohydrate content ranged from 61.18% to 63.47% with overall mean of 62.55%. The maximum carbohydrate content was found in RMG-62 (63.47%), while the minimum value was recorded in HUM-2 (61.18%). The results of this study corroborate with those of Mubarak (2005) [30], Agugo and Onimawo (2008) [3], Paul *et al*., (2011) [32] who have reported the carbohydrate content to be 62.9%, 61.47% and 60.35%, respectively. The proteins are the most abundant macromolecules in cells and constitute over half the dry weight of most organism. The crude protein content varied from 23.99% to 26.15% with overall mean of 25.08%. The maximum crude protein content was recorded in HUM-2 (26.15%) which was recorded at par with Pairy Mung (25.74%), while the minimum value was recorded in RMG-

268 (23.99%). The result showed that crude protein fell in the range of 20.97-31.32%, reported by Anwar *et al*. (2007) [7]. Saleem *et al*., (1998) [36] reported the crude protein content in mungbean seed from 22.88 to 24.65 per cent. Mungbean seed flour has been reported with protein contents of 25.09% (Agugo and Onimawo, 2008) [3]; 24.08% (Blessing and Gregory, 2010) [9]; 25.90% (Butt and Batool, 2010) [10]. The perusal of data revealed that soluble protein content in mungbean cultivars ranged from 15.83% to 19.72% with an overall mean of 17.97%. The data revealed the maximum soluble protein content in HUM-2 (19.72%) which was at par with Samrat (19.64), while the minimum value was obtained in PKV AKM-4 (15.83%). The result showed that soluble protein content fell in the range of 12.2-24.0%, reported by Anandhi and Vanniarajan (2014) [5]. The methionine content of mungbean cultivars ranged from 0.79 g/16gN to 1.76 g/16gN with an overall mean of 1.26 g/16gN. The data revealed that the cultivar HUM-2 (1.76 g/16gN) contained the highest amount of methionine which was found at par with Pairy Mung (1.72 g/16gN), while the minimum amount was obtained in MUM-2 (0.79g/16gN). Sattar *et al*., (1989) [37] reported methionine content as 1.5 g/16 g N, while Mubarak (2005) [30] reported methionine content to be 1.92 g/16g N in raw mungbean flour. The crude fat content in mungbean cultivars ranged from 1.12% to 1.62% with an overall mean of 1.37%. The maximum crude fat content was found in Pusa-0672 (1.62%) which was at par with MUM-2 (1.58%) and PKV AKM-4 (1.54%), while the minimum value was found in RMG-62 (1.12%) and HUM-2 (1.12%). The result showed that crude fat fell in the range of 0.93-3.93%, reported by Raturi *et al*. (2014) [34]. The present findings are fairly comparable to the findings of Anandhi and Vanniarajan, 2014 [5] (1.07-1.98%), Anwar *et al*., 2007 [7] (1.20-1.56%), Savage and Deo, 2000 [38] (1-1.5%), Agugo and Onimawo, 2009 [4] (1.43%), Butt and Batool, 2010 [10] (1.24 ± 0.08) and Chen, 1990 [12] (1.2-1.3%) in different mungbean germplasm. Food energy is contained in molecules of carbohydrate, fat, protein and alcohol. The perusal of data revealed that calorific value ranged from 347.93kcal/100g to 360.19 kcal/100g with an overall mean of 353.65 kcal/100g. The maximum calorific value was recorded in PKV AKM-4 (360.19kcal/100g) which was at par with MUM-2 (358.56 kcal/100g), while the minimum value was obtained in HUM-2 (347.93 kcal/100g)). Kavitha and Parimalavalli (2014) [19] reported that mungbean contains (342.12 ± 7.26) kcal energy /100g flour. Habibullah *et al.*, (2007) [15] reported that calorific value of two mungbean varieties ranged from 340 - 347 kcal /100g flour. Masood *et al*., (2014) [27] was found calorific value of mungbean flour to be 333.0 ± 0.34kcal/100g and Blessing and Gregory (2010) [9] reported calorific value of raw undehulled mungbean floor to be 336.65 kcal/100g. The variation observed in calorific value with the previous researchers may be due to variation in analytical methods as they calculated it using the Atwater factor as well as due to varietal differences.Phenols plays important role in disease resistance. The phenols of pulses are positively associated with antioxidant activity, which help in scavenging of free radicals.The total phenol content in mungbean cultivars ranged from 62.35 mg/100g to 89.61mg/100g with an overall mean of 73.04 mg/100g.The data on total phenol content evinced highest value in BPMR-145 (89.61mg/100g), while the minimum value was found in Pant M-4 (62.35 mg/100g). The observed range is fairly comparable to the results of Mondal *et al*. (2013) [29] who reported variation in phenol in the range of 60.5 to 94.7 mg/100g seeds of six mungbean varieties. Kim *et al*., (2012) [22] recorded the range of polyphenol to be 97.8±1.3 to101.1 ±1.0 mg/100g seeds. Ashing is the first step in preparing a food sample for determination of specific elemental analysis. The ash content of mungbean cultivars ranged from 3.42% to 4.01% with an overall mean of 3.67%. The maximum ash content was recorded in Pairy Mung (4.01%) which was at par with Pusa-0672 (3.97%), while the minimum value was found in Pant M-2 (3.42%). The findings are significant and comparable to those of Adel *et al*. (1980) [2], 3.31- 4.05%; Agugo and Onimawo (2008) [3], 3.43%; Habibullah *et al*. (2007) [15], 3.0-3.9%; Paul *et al*., (2011) [32], 3.85±0.05%, for mungbean flour. The cultivar HUM-2 was found to contain maximum amount of soluble protein, crude protein and methionine which was at par with Pairy Mung. Protein quality point of view HUM-2 is the best among the tested cultivar along with Pairy Mung followed by Pusa-0672. Apart from best protein quality Pairy Mung has the maximum concentration of ash. RMG-62 has maximum amount of total carbohydrate along with appreciable amount of protein. Samrat has appreciable amount of protein, carbohydrate and ash. Pusa -0672 contain highest amount of fat, ash, and good amount of protein and methoinine. On the basis of Biochemical and Nutritional point of view it can be concluded that HUM-2, Pairy Mung, RMG-62, Samrat and Pusa-0672 are the five suitable cultivars for Chitrakoot region among 12 tested cultivars.

The variations in biochemical parameters discussed here are mainly attributed to genetic makeup of cultivars, method of analysis, cultural practices along with some environmental factors which lead to differential synthesis of these compounds.

Table 1: Biochemical Composition of 12 Mungbean (*V.radiata* L. Wilczek) Cultivars

S. No.	Cultivar/Variety	Moisture (%)	Total Carbohydrate (%)	Crude Protein (%)	Soluble Protein (%)	Methionine (g/16g N)	Crude Fat (%)	Calorific Value (kcal/100g)	Total Phenol (mg/100g)	Ash (%)
1	Samrat	8.74	63.00	24.75	19.64	1.15	1.27	354.88	68.57	3.74
2	BPMR-145	8.94	62.44	24.81	16.21	1.45	1.44	351.20	89.61	3.70
3	HUM-2	9.95	61.18	26.15	19.72	1.76	1.12	347.93	76.75	3.66
4	MUM-2	9.90	62.89	25.16	17.43	0.79	1.58	358.56	73.83	3.52
5	Pairy Mung	9.43	61.36	25.74	17.81	1.72	1.33	351.20	72.12	4.01
6	Pant M-2	9.88	63.04	24.87	18.69	0.99	1.30	355.29	74.82	3.42
7	Pant M-4	9.88	62.96	24.58	18.98	1.20	1.40	351.61	62.35	3.62
8	PKV AKM-4	9.64	62.56	25.33	15.83	1.50	1.56	360.19	62.49	3.60
9	Pusa-0672	9.96	61.84	25.45	18.13	1.54	1.62	356.11	76.11	3.97
10	Pusa-9072	9.76	62.76	25.04	16.58	0.82	1.18	351.61	66.10	3.63
11	RMG-62	9.39	63.47	25.04	19.51	1.02	1.12	356.11	72.28	3.47
12	RMG-268	9.89	63.16	23.99	17.09	1.16	1.46	349.16	81.40	3.67
Grand Mean		9.61	62.55	25.08	17.97	1.26	1.37	353.65	73.04	3.67
SEm±		0.112	0.102	0.226	0.044	0.026	0.044	1.00	0.099	0.057
CD@5%		0.326	0.298	0.661	0.131	0.076	0.129	2.93	0.288	0.166

Note: Data represented on the basis of two years pooled mean

The results pertaining to storage protein profiles of mungbean seeds are presented in Table 2 and Figures (Fig.1and Fig.2).

The total seed storage proteins extracted from 12 mungbean cultivars were separated using SDS-PAGE. Protein distribution pattern were studied and revealed variations in terms of band number staining and molecular weight. SDS-PAGE of seed storage proteins of 12 cultivars of mungbean led to detection of 26 polypeptide bands with molecular weights of the resolved peptides ranged from 104 kDa to 18 kDa (Fig-1and Table-2) and showed 69.23 % polymorphism among them. Hameed *et al.* (2012) [16] also reported molecular weights of the resolved peptides ranged from 103 kDa to 16 kDa. These proteins were indicated as Mungbean Seed Storage Proteins (MSSP) followed by their molecular weights. Out of these, eight polypeptides (Mwt.79, 64, 58, 54, 50, 35, 25, 23 kDa) were universally present, while rest 18 polypeptide band were varied in their expression. The number of band observed ranged from 15 to 21. The maximum (21) number of bands was observed in both PKV AKM-4 and Pusa-9072 followed by both RMG-62 (20) and RMG-268 (20), while minimum number (15) of bands was observed in both Samrat and BPMR-145. In cultivars MUM-2, Pairy Mung, Pant M-2, Pant M-4 and Pusa-0672, 19 polypeptide bands were observed in each. In HUM-2, 18 polypeptide bands were observed. The banding pattern of each cultivar was differed from each other.

Seed storage protein profiles of 12 mungbean cultivars along with the dendrogram are presented in Fig.1 and Fig.2.The dendrogram revealed the genetic dissimilarity coefficient among the twelve cultivars of mungbean ranged from 0.08 to 0.67, but mostly concentrated between 0.08 and 0.29. Dendrogram based on electrophoretic data grouped the 12 cultivars into two major clusters, cluster I and cluster II. Further the major cluster II was sub clustered into II A, II B and II C, at dissimilarity coefficient 0.08 to 0.67. Major cluster I included cultivars C-1 (Samrat) and C-2 (BPMR-145) at dissimilarity coefficient of 0.16 depicted a low genetic diversity between both cultivars. Sub-cluster II A included cultivars C-3 (HUM-2), C-4 (MUM-2), C-6 (Pant M-2) and C-7 (Pant M-4), within them C-4and C-6 were closer than other two. Sub-cluster II B comprised of 4 cultivars including C-9 (Pusa-0672), C-10 (Pusa-9072), C-11 (RMG-62) and C-12 (RMG-268). Among them cultivars C-9 and C-10 are closer while C-12 is more distinct than others. Sub-cluster II C included C-5 (Pairy Mung) and C-8 (PKVAKM-4) which is most distinct from cluster-I (Samrat and BPMR-145), while very much close to C-12 (RMG-268) of sub cluster II B.

Seed storage protein in mungbean (*Vigna radiata*) can be classified on the basis of vicilin type (8S) and basic 7S globulins and legumin type (11S) globulins. Malviya *et al.*, (2008) [26] identified 11S and 2S globulins as seed storage protein having molecular weights of 17 kDa, and 14 kDa in mungbean. In the present study, a peptide band of approx. 18 kDa was detected on SDS-PAGE that may be 11 S globulin subunit.

Mendoza *et al.*, (2001) [28] isolated protein fractions from mungbean [*Vigna radiata* (L.) Wilczek] that were globulin types with 360 kDa, legumin with 200 kDa and vicilin with 135kDa for basic 7S subunit on native gel. While SDS-PAGE revealed that 11S was composed of two bands of 40 kDa and 24 kDa, 8S was composed of 60 kDa, 48 kDa, 32 kDa, and 26 kDa bands, and basic 7S was composed of 28 kDa and 16 kDa bands. In the present study, peptides with molecular weights 28 kDa was also detected on SDS-PAGE that may be basic 7S subunit, while peptide with molecular weight 23kDa may be 11S globulin. 32 kDa and 25 kDa peptide may be 8S vicilin subunit according to earlier report (Mendoza *et al.*, 2001) [28].

SDS-PAGE profiling of seed storage proteins proved to be an economical and simple technique for analysis of genetic variation in mungbean germplasm. Variability in seed proteins was mainly in basic globulins, vicilin and legumin. Narrow genetic variability in mungbean germplasm based on seed storage proteins pointed towards the need to exploit the large germplasm collection with diverse morpho-agronomic traits. SDS-PAGE for Tris soluble proteins found suitable for testing distinctness, uniformity, stability of varieties for registration and identification (Singh *et al*., 2015) [40].

Table 2: Molecular Weights of resolved Peptides indicated as Mungbean Seed Storage Proteins (MSSP) followed by their molecular weight (kDa).

Mol. Wt.	C-1	C-2	C-3	C-4	C-5	C-6	C-7	C-8	C-9	C-10	C-11	C-12
MSSP-104	0	0	0	1	1	1	1	1	1	1	1	1
MSSP-99	1	1	0	0	1	0	0	0	1	1	1	1
MSSP-95	0	0	1	0	1	1	1	1	0	0	0	0
MSSP-92	0	0	0	1	0	1	1	1	1	1	1	1
MSSP-79	1	1	1	1	1	1	1	1	1	1	1	1
MSSP-70	1	1	1	1	1	1	1	0	1	1	1	1
MSSP-67	0	0	0	0	0	0	0	1	0	1	0	1
MSSP-64	1	1	1	1	1	1	1	1	1	1	1	1
MSSP-58	1	1	1	1	1	1	1	1	1	1	1	1
MSSP-54	1	1	1	1	1	1	1	1	1	1	1	1
MSSP-50	1	1	1	1	1	1	1	1	1	1	1	1
MSSP-39	1	1	1	0	0	0	1	0	0	0	1	0
MSSP-37	0	0	1	1	1	1	1	1	0	0	0	0
MSSP-36	0	0	0	0	1	0	0	1	0	0	0	1
MSSP-35	1	1	1	1	1	1	1	1	1	1	1	1
MSSP-33	1	1	1	1	0	1	1	0	1	1	1	1
MSSP-32	0	0	0	0	0	0	0	0	1	1	1	0
MSSP-29	1	1	1	1	0	1	1	1	1	1	0	1
MSSP-28	0	0	0	1	1	1	0	1	1	1	1	1
MSSP-27	0	0	1	1	0	1	1	1	1	1	1	0
MSSP-25	1	1	1	1	1	1	1	1	1	1	1	1
MSSP-23	1	1	1	1	1	1	1	1	1	1	1	1
MSSP-22	1	1	1	1	1	1	1	1	0	1	1	0
MSSP-21	0	1	1	1	1	0	0	1	1	1	1	1
MSSP-20	1	0	0	0	1	0	0	1	0	0	0	1
MSSP-18	0	0	1	1	1	1	1	1	1	1	1	1
No of Bands	15	15	18	19	19	19	19	21	19	21	20	20

0-absent and 1-present of peptide bands

C-1: Samrat, C-2: BPMR-145, C-3: HUM-2, C-4: MUM-2, C-5: Pairy Mung, C-6: Pant M-2, C-7: Pant M-4, C-8: PKV AKM-4, C-9: Pusa-0672, C-10: Pusa-9072, C-11: RMG-62, C-12: RMG-268.

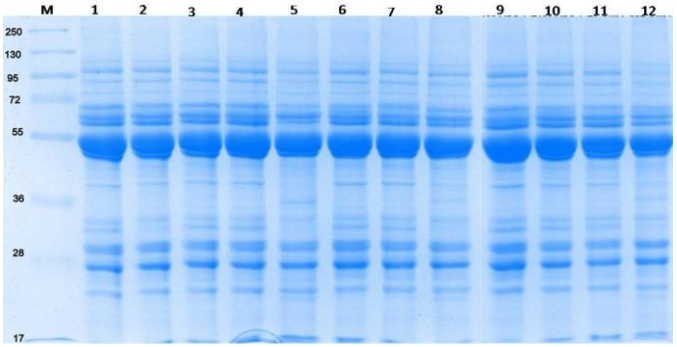

Fig 1: Total seed storage protein profile of 12 cultivars of Mungbean using Sodium Dodecyl Sulphate Polyacrylamide Gel Electrophoresis (SDS-PAGE). M: Ladder, 1: Samrat, 2: BPMR-145, 3: HUM-2, 4: MUM-2, 5: Pairy Mung, 6: Pant M-2 7: Pant M-4, 8: PKV AKM-4, 9: Pusa 0672, 10: Pusa 9072, 11: RMG 62, 12: RMG 268

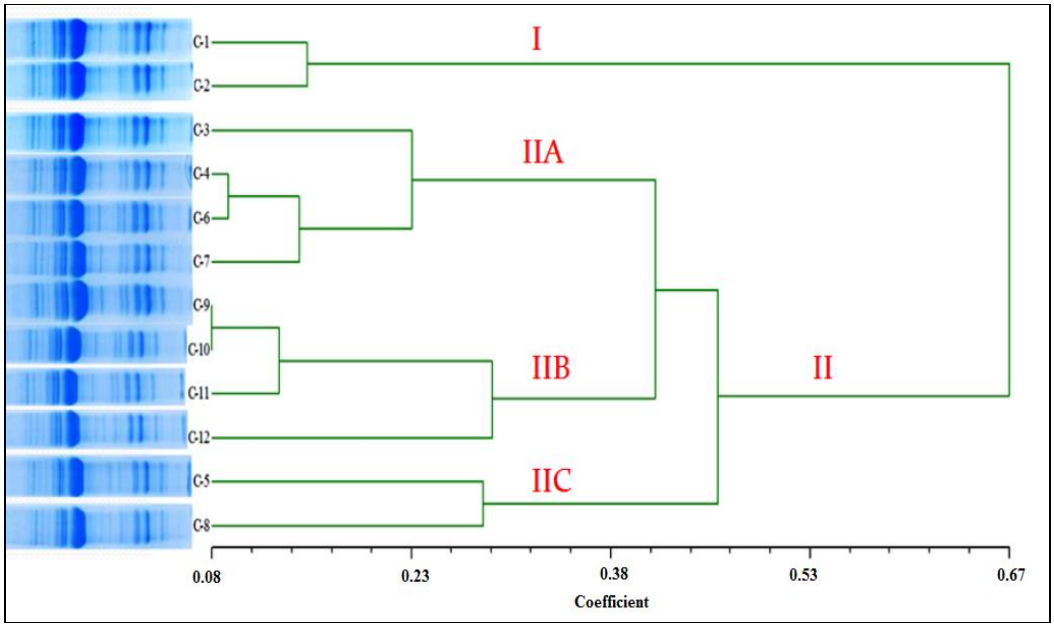

Fig 2: Dendrogram showing genetic diversity of 12 mungbean cultivars based on seed storage protein profiles. C1: Samrat, C2: BPMR-145, C3: HUM-2, C4: MUM-2, C5: Pairy Mung, C6: Pant M-2, C7: Pant M-4, C8: PKV AKM-4, C9: Pusa 0672, C10: Pusa 9072, C11: RMG 62, C12: RMG 268

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References

1. A.O.A.C. Official Methods of Analysis of the Association of Official Analytical Chemists. 1970, Washington D.C. 11th edition.
2. Adel A, Shehata Y, Thannoun M. Chemical and amino acid composition of Iraqi mungbeans. Zeitschrift fur Lebensmitteluntersuchung undforschung A. 1980; 171:360-362.
3. Agugo UA, Onimawo IA. Effect of heat treatment on the nutritional value of mungbean. M.Sc. project, Department of Nutrition and Dietetic, Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria. 2008, 32-35.
4. Agugo UA, Onimawo IA. Heat treatment on nutritional value of green gram. Electronic J Environ. Agri. Food Chemistry. 2009; 8(10):924-930.
5. Anandhi Lavanya S, Vanniarajan, C. Biochemical characterisation of elite green gram (*Vigna radiata* (L.) Wilczek) genotypes. International Journal of Scientific Research. 2014; 3(3):1-2.
6. Andersen P. Report on the Nutritive value of rice bean. European Commission 6th Framework Programme, Food Security through rice bean research in India and Nepal (FOSRIN). Report 3. Nutritional qualities of rice bean, Bergen, Norway. 2007, 1-10.
7. Anwar F, Latif S, Przybylski R, Sultana B, Ashraf M. Chemical composition and antioxidant activity of seeds of different cultivars of Mungbean. Journal of Food Science. 2007; 72(7):503-510.
8. Arumuganathan K, Earle ED. Nuclear DNA content of some important plant species. Plant Molecular Biology Report. 1991; 9:208-218.
9. Blessing IA, Gregory IO. Effect of processing on the proximate composition of the dehulled and undehulled mungbean (*Vigna radiata* (L.) Wilczek) flours. Pakistan Journal of Nutrition. 2010; 9(10):1006-1016.
10. Butt MS, Batool R. Nutritional and functional properties of some promising legumes protein isolates. Pak. J Nutrition. 2010; 9:373-379.
11. Charmaine S. Asia food. Encyclopedia of Asian food (Periplus editions) Australia*:* New Holland Publishers Pty Ltd, 1998.
12. Chen X. The 17th Intl. mungbean Nursery trial. Training Report 19th Regional Course in mungbean production and research. Kasefart University. Bangkok, Thailand. 1990, 1-6.
13. Emre I. Determination of genetic diversity in the *Vicia L.* (section *vicia*) by using SDS-PAGE. Pakistan. Journal of Botany. 2011; 43(3):1429-1432.
14. Ghafoor A, Ahmad Z, Qureshi AS, Bashir M. Genetic relationship in *Vigna mungo* (L.) Hepper and *V. radiata* (L.) R. Wilczek based on morphological traits and SDS-PAGE. Euphytica. 2002; 123(3):367-378.
15. Habibullah MA, Abbas M, Shah HU. Proximate and mineral composition of mungbean. Sarhad J Agri. 2007; 23(2):463-466.
16. Hameed A, Qureshi M, Nawaz M, Iqbal N. Comparative seed storage protein profiling of mungbean genotypes. Pakistan Journal of Botany. 2012; 44(6):1993-1999.
17. Hedge JE, Hofreiter BT. In: Carbohydrate chemistry 17 (Eds. Whistler R L., and Be Miller J N.,) Academic press New York, 1962.
18. Horn MJ, Jones DB, Blum AE. Colorimetric determination of methionine in proteins and foods. Journal of Biological Chemistry. 1946; 166:313-320.
19. Kavitha S, Parimalavalli R. Effect of processing methods on proximate composition of cereal and legume flours. Journal of Human Nutrition & Food Science. 2014; 2(4):1051.
20. Khalil AA. Nutritional improvement of Egyptian breed of mungbean by probiotic Lactobacilli. African Journal of Biotechnology. 2006; 5:206-212.
21. Khan TN, Dixit GP. Breeding pigeon pea in India and Australia. Indian Journal of Pulses Research. 2001; 14:1-9.

22. Kim DK, Jeong SC, Gorinstein S, Chon SU. Total polyphenols, antioxidant and antiproliferative activities of different extracts in mungbean seeds and sprouts. Plant Foods Hum Nutr. 2012; 67:71-75.
23. Laemmli UK. Cleavage of structural proteins during assembly of the head bacteriophage T4. *Nature*, 1970; 227:680-685.
24. Lowry OH, Rosebrough NJ, Farr AL, Randall RJ. Protein measurement with folin phenol reagent. Journal of Biological Chemistry. 1951; 193:265-275.
25. Malik CP, Singh MB. Analysis of total phenols. Plant enzymology and histo enzymology, Kalyani Publishers, New Delhi, 286, 1980.
26. Malviya N, Nayak S, Yadav D. Characterization of total salt soluble seed storage proteins of grain legumes using SDS-PAGE.PGR News Letter. 2008; 156:50-56.
27. Masood T, Shah HU, Zeb A. Effect of sprouting time on proximate composition and ascorbic acid level of mung bean (*vigna radiata* l.) and chickpea (*cicer arietinum* l.) seeds. The Journal of animal & plant Sciences. 2014; 24(3):850-859.
28. Mendoza EM, Adachi M, Bernardo AE, Utsumi S. Mungbean [*Vigna radiata* (L.) Wilczek] globulins: purification and characterization. Journal of agricultural and food chemistry. 2001; 49(3):1552-1558.
29. Mondal NK, Datta JK, Banerjee A. Biochemical response of mungbean (*Vigna radiata* L.) under the influence of reduced dose of chemical fertilizer and different time and method of application of biofertilizer. Journal of Agricultural Technology. 2013; 9(3):643-658.
30. Mubarak AE. Nutritional composition and antinutritional factors of mungbean seeds (*Phaseolus aureus*) as affected by some home traditional processes. Food Chem. 2005; 89:489-495.
31. Panse VG, Sukhatme PV. Statistical Methods for Agricultural Workers, I.C.A.R., New Delhi. 1978.
32. Paul T, Mozumder NHM, Sayed MA, Akhtaruzzaman A. Proximate composition, mineral content and determination of protease activity from greengram (*Vigna radiata* L.wilczek). Bangladesh Research Publication Journal. 2011; 5(3):207-213.
33. Pervaiz ZH, Tehrim S, Rabbani MA, Masood MS, Malik SA. Diversity in major seed storage proteins of rice landraces of Pakistan. Pakistan Journal of Botany. 2011; 43(3):1607-1612.
34. Raturi A, Singh SK, Sharma V, Pathak R. Genetic variability and interrelationship among qualitative and quantitaitive traits in mungbean. Legume Research. 2014; 37(1):1-10
35. Rohlf FJ. NTSYS-PC. Numerical taxonomy and multivariate analysis system, version 2.02e. Exeter Software, New York. 1997.
36. Saleem B, Ilyas F, Ali S, Qureshi MJ, Malik IA. Studies on chemical analysis of mungbean (*Vigna radiata* (L.) Wilczek). Pakistan J Bio. Sci. 1998; 1(2):120-123.
37. Sattar A, Durrani SK, Mahmood F, Ahmad A, Khan A. Effect of soaking and germination temperatures on selected nutrients and antinutrients of Mungbean. Food Chemistry. 1989; 34:111-120.
38. Savage GP, Deo S. The nutritional value of mungbean and urd bean. Nutrition Abstract Reil C (Series A). 2000; 59:66-83.
39. Shah SMA, Rahman H, Abbasi FM, Rabbani MA, Khan IA, Shinwari ZK *et al*. Interspecific variation of total seed protein in wild rice germplasm using SDS-PAGE. Pakistan Journal of Botany. 2011; 43(4):2147-2152.
40. Singh S, Kumar P, Trivedi A, Koshal AK. Seed protein profiling through electrophoresis in mungbean [*V.radiata* (L.) Wilczek]. Journal of Plant Development Sciences. 2015; 7(4):337-340.
41. Sneath PHA, Sokal RR. Numerical taxonomy: The principle and practice of numerical classification. W.F. FREEMAN & CO; San Francisco. 1973, 573.