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Protein landmarks for diversity assessment in wheat genotypes

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Grain proteins from 20 Indian wheat genotypes were evaluated for diversity assessment based seed storage protein profiling on sodium dodecylsulphate polyacrylamide gel electrophoresis (SDS-PAGE). Genetic diversity was evaluated using Nei's index, Shannon index and Unweighted pair group method with arithmetic mean (UPGMA) cluster analysis by constructing dendrogram of fractions of proteins, which were used for the calculation of similarity coefficients between these varieties. Diversity analysis attributes exhibited the importance of seed storage as a marker system. The similarity ranged from 32.14% to as high as 100% between genotypes. Adoption of this technology would be useful to plant protection regulatory systems, especially for plant variety identification and registration of new plant varieties, breeding programs and protection purposes.

Key words: Sodium dodecylsulphate polyacrylamide gel electrophoresis (SDS-PAGE), genetic diversity, population diversity index, coefficient of similarity.

INTRODUCTION

Wheat (*Triticum* sp.) is a staple food crop for more than 35% of the world population and also one of the widely cultivated crops in India. In 2007, world production of wheat was 653 million tons, making it the third most-produced cereal after maize (840 million tons) and rice (696 million tons). India is the second largest producer of wheat after China in 2010 (FAO, 2010) with an annual output of over 85.93 mt. (Economic Survey, GOI. 2010) where drought is the main abiotic stress limiting its grain yields. Varieties have been a landmark in the genetic improvement of wheat, as it resulted in increase in its potential for grain yield. Information about genetic diversity and genetic relatedness among elite material is a fundamental element in plant breeding (Zhu et al., 2000). Cultivar identification is useful for describing a new cultivar, testing genotype purity and speeding up distinctness uniformity- stability (DUS) test for candidate cultivar (Chan and Sun, 1997). For acquiring plant breeder's rights (PBR), varieties of agricultural importance have to be tested for distinctness (D), uniformity (U) and stability (S) (DUS testing) (Ardley and Hoptroff, 1996). Evaluation of genetic diversity in wheat has been on differences in morphological and agronomic traits or pedigree information (Bernard et al., 1998). A number of methods are currently available for analysis of genetic diversity in germplasms accessions, breeding lines and segregating populations. These methods have relied on pedigree, morphological, agronomic performance, biochemical and molecular (DNA-based) data (Mohammadi and Prasanna, 2003). Morphological traits can be used for assessing genetic diversity but are often influenced by the environment. The use of biochemical/molecular markers for the evaluation of genetic diversity has received much attention in recent years. A large number of germplasm lines can be characterized for biochemical markers in a short period of time. In addition, the data reflects

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more truly the genetic variability as biochemical markers are direct product of genes expression (Perry and McIntosh, 1991; Masood et al., 2000). Among biochemical techniques, SDS-PAGE is widely used due to its simplicity and effectiveness for describing the genetic structure of crop germplasm (Murphy et al., 1990; Javaid et al., 2004; Anwar et al., 2003.). The analysis of storage protein variation in wheat has proved to be a useful tool not only for diversity studies but also to optimize variation in germplasm collections (Ciaffi et al., 1993; Masood et al., 2000).

Wheat storage proteins (WSP), namely gliadins and glutenins, are the main components of gluten, which is the main contributor to the rheological and breadmaking properties of wheat flour. Gluten proteins give dough its unique viscoelastic properties. Glutenin proteins are polymeric, with disulphide bonds linking the individual glutenin polypeptides, which are known as subunits and two distinct glutenin groups: high and low molecular weight glutenin subunit (HMW-GS and LMW-GS respectively). The subunits of these two groups differ in terms of amino acid composition, molecular weight, that is from 23 to 68 kDa for LMW-GS and from 77 to 160 kDa for HMW-GS and in their structure (Kasarda, 1999). The gliadins are monomeric proteins, with a molecular weight of around 30 to 60 kDa. Studies on the genetic determination of wheat storage proteins have revealed that both gliadins and glutenins are heritated at several loci on each genome A, B and D. HMW-GS genes are located on the long arm of chromosomes 1A, 1B and 1D at loci Glu-A1, Glu-B1 and Glu-D1, respectively (Payne, 1987). LMW-GS genes are located at Glu-A3, GluB3 and Glu-D3 loci on the short arms 1AS, 1BS and 1DS, respectively (Singh and Shepherd, 1988). Genes coding for ω- and many γ-gliadins are tightly clustered at three homologous loci named Gli-A1, Gli-B1 and Gli-D1, at the distal end of 1AS, 1BS and 1DS respectively. The Gli-1 loci are close to the LMW-GS coding Glu-3 loci (Singh and Shepherd, 1988; Pogna et al., 1990). Some ω-gliadins are also encoded by genes proximal to Gli-1 loci and named Gli-A4, Gli-A5 and Gli-B3 (Metakovsky et al., 1997a). The α-, β- and some γ- gliadins are encoded by tightly clustered genes at a single locus on each of the short arms of the chromosomes of group 6, named Gli-A2, Gli-B2 and Gli-D2 respectively. For each HMW-GS and LMW-GS coding loci a high degree of polymorphism was revealed by SDS-PAGE for bread and durum wheat (Payne and Lawrence, 1983; Gupta et al., 1990; Branlard et al., 1989).

SDS-PAGE can be used as a promising tool for distinguishing cultivars of particular crop species (Jha and Ohri, 1996). The main objective of our research was to evaluate the potential of SDS-PAGE technique to assess the genetic diversity and relatedness among 20 Indian wheat genotypes based on seed storage protein profiles and to develop an optimized and efficient operational system for their use.

MATERIALS AND METHODS

Plant sample

In the present study, 20 Indian wheat varieties used extensively in the breeding programs were collected from different ecological regions of India (Table 1).

SDS-PAGE analysis

The variability of seed storage-proteins was analyzed by using SDS-PAGE. For extraction of protein, each variety had three replications and 10 seed for every replication was randomly selected and ground to fine powder with mortar and pestle, and a total of 400 ul sample buffer was added to a 0.01 g (10 mg) seed powder and mixed thoroughly by vortex in an Eppendorf tube (1.5 ml) with a small glass rod. The extraction buffer contained the following final concentration: 1.5 M Tris-HCl (pH 8.8), 2.5% SDS, 10% glycerol and 5% 2-mercaptoethanol, kept overnight at 40°C and centrifuged at 10000 rpm for 10 min. To monitor the movement of the protein in the gel, bromophenol blue (BPB) was used as a tracking dye. Seed protein was analyzed through slab-type SDS-PAGE using 7% polyacrylamide gel. The molecular weights of the dissociated polypeptides were determined using protein standards (MW-SDS-70) of Merck, India. SDS-PAGE of total seed protein was carried out in a discontinuous buffer system following the method of Laemmli (1970). The gels were stained with Coomassie brilliant blue (CBB) and destained till the background became transparent.

Data analysis

For each variety, electrophore gram was scored and the presence (1) or absence (0) of each band was noted and bi-variate 1 - 0 data matrix was generated (Table 2). In the present study, the population diversity based on SDS PAGE banding patterns was calculated using POPGENE 1.31 (Yeh et al., 1999) software. The similarity coefficient matrix generated from primers data for 20 *Triticum* sp. genotypes was subjected to algorithm unweighted pair group method with arithmetic mean (UPGMA) and clusters were generated using NTSYS 2.02pc program (Rohlf, 2000).

RESULTS AND DISCUSSION

In this study, high and low molecular weight glutenin subunits of different wheat varieties were separated by SDS-PAGE electrophoresis for characterization and evaluation of genetic diversity among the given set of varieties with a protein weight marker of 3.5 to 205 kDa was used for this purpose. Electropherogram showing proteins banding pattern of different wheat varieties are presented in Figure 1.

For analysis of banding pattern on the gels, they were recorded as present or absent and assigned each band a value of 1 for presence and 0 for absence. Perusal of Table 2 revealed that a total of 32 bands were observed on gel out of which 24 were polymorphic with 75% of polymorphism and also show that the total number of bands varied from 17 (Raj3765 and Raj3077) to 25 (HI8498) in different varieties. The size of polypeptides resolved ranged from 6.5 to 129.0 KDa. Eight bands

Table 1. Pedigree of 20 genotypes *Triticum* sp. used for study.

Variety	Ploidy	Pedigree
Raj4083	*T. aestivium*	PBW 343 / UP 2442 // WR 258 / UP 2425
Raj3765	*T. aestivium*	HD-2402/VL-639
Raj4120	*T. aestivium*	PBW373/V1
Raj4037	*T. aestivium*	DL-788-2/RAJ-3717
Raj1482	*T. aestivium*	NAPO-63/TOBARI-66//KALYANSONA/BLUEBIRD; CARTHAGE//KALYANSONA/BLUEBIRD;NAPO-63/TOBARI-66//II-8156/3/KALYANSONA/BLUEBIRD
HD2684	*T. aestivium*	Elite *aestivium* variety
Raj3077	*T. aestivium*	HD-2267/RAJ-1482//RAJ-1802
Raj1972	*T. aestivium*	HD-2195/HD-2160; SUJATA/NP-852/3/GABO/NORIN-10-BREVOR//2*LERMA-ROJO-64-A/5/SAFED-LERMA/NP-852/4/PENJAMO-62/P-14//KENTANA-54-B/3/K-65/6/HD-2160
PBW502	*T. aestivium*	W-485/PBW-343//RAJ-1482
Lok1	*T. aestivium*	S-308/S-331; SONALIKA/CHOTI-LERMA
Raj6560	*T. durum*	Elite *durum* variety
Raj1555	*T. durum*	COCORIT-71/RAJ-911
PDW291	*T. durum*	Elite *durum* variety
PDW233	*T. durum*	YAVAROS(SIB)/(SIB)TEN; YAVAROS(SIB)/(SIB)TEZONTLE
WH896	*T. durum*	STIFFTAIL(SIB)/(SIB)YAVAROS//(SIB)PENELOPE
HI8498	*T. durum*	RAJ-6070/RAJ-911
MACS1967	*T. durum*	GULAB/CPAN-1471
PDW274	*T. durum*	DWL-6018/KARPASIA
Raj6496	*T. durum*	CHAMS-3 (Selection from CIMMYT)
MACS9	*T. durum*	N-59(TR.DR)/(TR.PO)F-183; N-59/F-185

(6.5, 14.3, 16.0, 20.58, 29.0, 31.5, 35.0 and 88.0 KDa) were monomorphic for all genotypes.

Polypeptides having molecular weight of 119.85, 52.0, 45.3, 34 (absent in Raj3765), and 61.4 KDa (Raj3765 and Raj1482) were found only in *T. aestivum* whereas, polypeptide of 56.8, 54.5, 47.6, 39.8, 36.2 and 22.14 KDa weight were found only in *T. durum*. Shuaib et al. (2007) observed similar type of results in 13 Pakistani wheat varieties and total of 21 bands were obtained among which seven bands were common in all varieties and other bands show variation.

Further analysis of the result revealed that polypeptide having molecular weight of 129, 96 and 77.6 KDa were unique for four *T. durum* varieties *viz*., HI8498, MACS1967, PDW274, Raj6496 and MACS9 whereas, polypeptide having 106 KDa molecular weight were found only in two varieties of *T. aestivum* named HD2684 and Raj1972.

In Raj4037 and Raj1482, polypeptide of 97.4 KDa was absent but it was present in all other varieties as well as 43 KDa polypeptide was absent only in Raj1555. A 27 KDa polypeptide unit was absent in Raj1555, PDW291, PDW233 and WH896. Two more polypeptides with a molecular weight of 74.7 and 66.0 KDa were present in all varieties except Raj3077 which was deficit for both bands and Raj1482 deficit for 74 KDa and Raj4120 for 66KDa only.

Diversity analysis

In the present study, the population diversity based on SDS PAGE electropherogram patterns was calculated using POPGENE 1.31software.

The Shannon diversity index (H) is one common diversity index often used to characterize allele diversity in a locus. Shannon's index accounts for both abundance and evenness of the alleles present (Shannon and Weaver, 1949), and are useful for understanding allele structure at a locus and measures gene diversity. Perusal of Table 3 reveals that Shannon's information index, was found to be 0.4260 and Nei's (1973) gene diversity or expected heterozygosity (H_e) another common diversity index in population genetics was 0.2916 and total genetic diversity during the present study was found to be 0.2916 and also equivalent to Nei's gene diversity or expected heterozygosity (H_e).

Based on the fact that the diversity within variety was not observed during study, shows that varieties are well maintained to avoid any genetic contamination through a chance of cross pollination. Total genetic diversity (H_T) and genetic diversity within varieties (H_S) were used for the determination of the inter-variety genetic diversity ($D_{ST} = H_T - H_S$). However, there was no within variety diversity observed as a result inter- variety genetic diversity yielding a value of 0.2916. The G_{ST} parameter

Table 2. Seed Storage protein profiling of 20 *Triticum* sp. genotypes appeared on SDS- PAGE.

KDa	Raj 4083	Raj 3765	Raj 4120	Raj 4037	Raj 1482	HD 2684	Raj 3077	Raj 1972	PBW 502	Lok1	Raj 6560	Raj 1555	PDW 291	PDW 233	WH8 96	HI 8498	MACS 1967	PDW 274	Raj 6496	MAC S 9
129	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	1
119.85	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0
106.5	0	0	0	0	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0
97.4	1	1	1	0	0	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
96	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	1
94.44	0	0	0	1	1	1	0	1	0	0	1	1	1	1	1	1	1	1	1	1
88	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
80	1	0	0	1	1	1	0	0	1	1	1	1	1	1	1	1	1	1	1	1
77.6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	1
74.7	1	1	1	1	0	1	0	1	1	1	1	1	1	1	1	1	1	1	1	1
71.8	1	0	0	0	1	0	0	1	1	1	1	1	1	1	1	1	1	1	1	1
66	1	1	0	1	1	1	0	1	1	1	1	1	1	1	1	1	1	1	1	1
61.4	1	0	1	1	0	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0
56.8	0	0	0	0	0	0	0	0	0	0	1	1	1	1	1	1	1	1	1	1
54.5	0	0	0	0	0	0	0	0	0	0	1	1	1	1	1	1	1	1	1	1
52	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0
50	1	0	0	1	0	0	0	0	0	0	1	1	1	1	1	1	0	0	0	0
47.6	0	0	0	0	0	0	0	0	0	0	1	1	1	1	1	1	1	1	1	1
45.3	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0
43	1	1	1	1	1	1	1	1	1	1	1	0	1	1	1	1	1	1	1	1
39.8	0	0	0	0	0	0	0	0	0	0	1	1	1	1	1	1	1	1	1	1
36.2	0	0	0	0	0	0	0	0	0	0	1	1	1	1	1	1	1	1	1	1
35	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
34	1	0	1	1	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0
31.5	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
29	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
27	1	1	1	1	1	1	1	1	1	1	1	0	0	0	0	1	1	1	1	1
22.14	1	1	1	1	1	1	1	1	1	1	0	0	0	0	0	0	0	0	0	0
20.58	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
16	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
14.3	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
6.5	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1

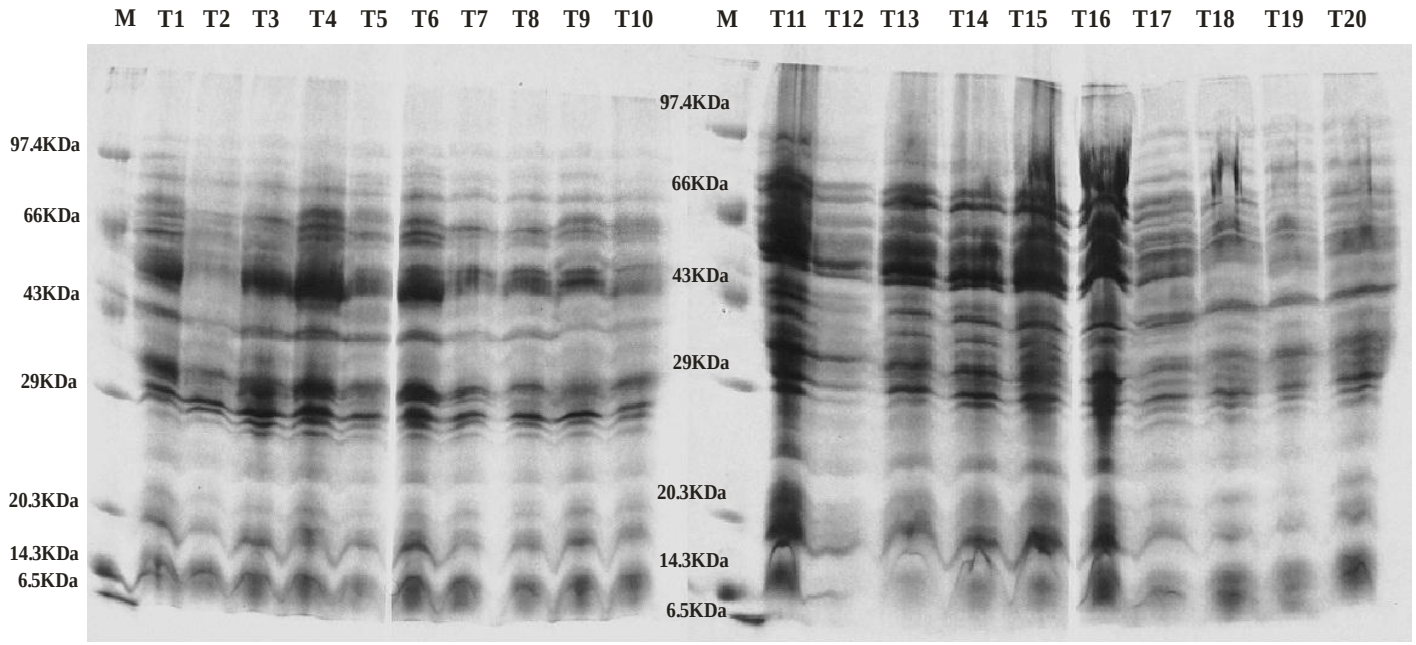

Figure 1 . SDS-PAGE profile of storage seed protein extracted from 20 genotype of Triticum sp. M, Represent medium range protein weight marker (65 Kda to 97.4Kda0; T_1-T_{20} represent the following Triticum sp. Genotype. T1, Raj 4083; T2, Raj 3765; T3, Raj 4120; T4, Raj 4037; T5, Raj 1482; T6, HD 2684; T7, Raj 3077; T8, Raj 1972; T9, PBW 502; T10, Lok1; T11, Raj 6560; T12, Raj 1555; T13, PDW 291; T14, PDW 233; T15, WH 896; T 16, HI 8498; T17, MACS 1967; T 18, PDW 274; T 19, Raj 6496; T 20, MACS 9.

Table 3. Number of bands observed, number of polymorphic bands, percent polymorphism Nei's gene diversity (h), Shannon's Information index (I), total genetic diversity (H_T), genetic diversity within varieties (H_S), inter variety genetic diversity ($D_{ST} = H_T - H_S$), relative magnitude of differentiation among varieties (Gst), and estimate of gene flow from Gst (Nm) value of each pattern for 20 varieties of *Triticum* sp. using SDS-PAGE marker system.

S/N	Number of bands observed	Number of polymorphic bands	Polymorphism (%)	Nei's gene diversity (h)	Shannon's Information index (I)	H_T	H_S	D_{ST}	G*st*	Nm
1	32	24	75	0.2916	0.4260	0.2916	0.00	0.2916	1.00	0.00

(relative magnitude of differentia-tion among varieties) was reasonable 1.000, explaining the low value of estimate of gene flow (Nm) which was 0.000.

Genetic relationship and cluster analysis

The data obtained using SDS-PAGE electrophe-rogram were further used to construct similarity matrices using the method of Jaccard's coefficient analysis of *Triticum* sp. genotypes for estimation of genetic similarity. SDS PAGE electropherogram similarity matrices of 20 *Triticum* sp. genotypes

Table 4. Jaccard's Average similarity coefficient among *Triticum* genotypes based on SDS PAGE profiling.

Variety	Raj 4083	Raj 3765	Raj 4120	Raj 4037	Raj 1482	HD 2684	Raj 3077	Raj 1972	PBW 502	Lok1	Raj 6560	Raj 1555	PDW 291	PDW 233	WH 896	HI 8498	MACS 1967	PDW 274	Raj 6496	MACS 9
Raj 4083	1.0000																			
Raj 3765	0.7727	1.0000																		
Raj 4120	0.8182	0.8421	1.0000																	
Raj 4037	0.8696	0.7273	0.7727	1.0000																
Raj 1482	0.7826	0.7143	0.6818	0.8182	1.0000															
HD 2684	0.8333	0.7727	0.8182	0.8696	0.7826	1.0000														
Raj 3077	0.7727	0.7895	0.9444	0.7273	0.7143	0.7727	1.0000													
Raj 1972	0.8333	0.7727	0.8182	0.7917	0.7826	0.9130	0.7727	1.0000												
PBW 502	0.9545	0.8095	0.8571	0.8261	0.8182	0.8696	0.8095	0.8696	1.0000											
Lok1	0.9545	0.8095	0.8571	0.8261	0.8182	0.8696	0.8095	0.8696	1.0000	1.0000										
Raj 6560	0.5714	0.5000	0.4286	0.5357	0.5185	0.5172	0.3929	0.5172	0.5357	0.5357	1.0000									
Raj 1555	0.5000	0.4231	0.3571	0.4643	0.4444	0.4483	0.3214	0.4483	0.4643	0.4643	0.9091	1.0000								
PDW 291	0.5357	0.4615	0.3929	0.5000	0.4815	0.4828	0.3571	0.4828	0.5000	0.5000	0.9545	0.9524	1.0000							
PDW 233	0.5357	0.4615	0.3929	0.5000	0.4815	0.4828	0.3571	0.4828	0.5000	0.5000	0.9545	0.9524	1.0000	1.0000						
WH 896	0.5357	0.4615	0.3929	0.5000	0.4815	0.4828	0.3571	0.4828	0.5000	0.5000	0.9545	0.9524	1.0000	1.0000	1.0000					
HI 8498	0.5161	0.4483	0.3871	0.4839	0.4667	0.4688	0.3548	0.4688	0.4839	0.4839	0.8800	0.8000	0.8400	0.8400	0.8400	1.0000				
MACS 1967	0.4839	0.4643	0.4000	0.4516	0.4828	0.4839	0.3667	0.4839	0.5000	0.5000	0.8400	0.7600	0.8000	0.8000	0.8000	0.9600	1.0000			
PDW 274	0.4839	0.4643	0.4000	0.4516	0.4828	0.4839	0.3667	0.4839	0.5000	0.5000	0.8400	0.7600	0.8000	0.8000	0.8000	0.9600	1.0000	1.0000		
Raj 6496	0.4839	0.4643	0.4000	0.4516	0.4828	0.4839	0.3667	0.4839	0.5000	0.5000	0.8400	0.7600	0.8000	0.8000	0.8000	0.9600	1.0000	1.0000	1.0000	
MACS 9	0.4839	0.4643	0.4000	0.4516	0.4828	0.4839	0.3667	0.4839	0.5000	0.5000	0.8400	0.7600	0.8000	0.8000	0.8000	0.9600	1.0000	1.0000	1.0000	1.0000

revealed the relationship among them (Table 4). The Jaccard's similarity coefficient values between different genotypes ranged from 0.3214 (Raj3077 and Raj1555) to 1.000 (PBW502 and Lok1, PDW291 and PDW233, PDW291 and WHH896, PDW233 and WHH896, MACS1967 and PDW274, MACS1967 and Raj 6496, MACS1967 and MACS9, PDW274 and Raj6496, PDW274 and MACS9, Raj6496 and MACS9) with an average of 0.6478 (Table 4). However, average diversity estimated was 35.22%, with a range from 00 to 67.86% diversity. Perusal of the Table 4 reveal that genotypes of *T. durum* were comparatively less diverse than *T. aestivum* with an average diversity of 11.85%(ranged from 00 to 24%) and 17.98% (ranged from 00 to 31.82%) respectively.

The PAGE cluster tree analysis of 20 *Triticum* sp. genotypes showed that they were clearly divided into two major clusters namely group I and group II at a similarity coefficient of 0.4700 (Figure 2). First cluster named group I that represents *T. aestivum* were further divided into two subgroups that is subgroup Ia which consisted of seven genotypes viz., Raj4083, PBW502, Lok1, HD2684, Raj1972, Raj4037 and Raj1482 and subgroup Ib which consisted of three genotypes viz., Raj3765, Raj4120 and Raj3077; both subgroups were joined at a similarity coefficient of 0.7800 whereas, *T. durum* genotypes commonly represent group II were further subdivided only into two group named subgroup IIa which consisted of five genotypes viz., Raj1555, Raj6560, PDW291, PDW233 and WH896 and subgroup IIb which consisted of five genotypes *viz*., HI8498, MACS1967, PDW274, Raj6496 and MACS9; both of these subgroups were joined at a similarity coefficient of 0.8100.

Although variation in storage protein banding pattern was revealed by SDS-PAGE, however, its magnitude was low. Based on SDS-PAGE, 32 bands were used for analysis and genetic diversity was estimated based on the number of different protein peptides between the two compared. A low level of population genetic diversity index may be attributed to narrow genetic base of a wheat crop. SDS-PAGE electrophoresis of seven wheat varieties has been previously investi-

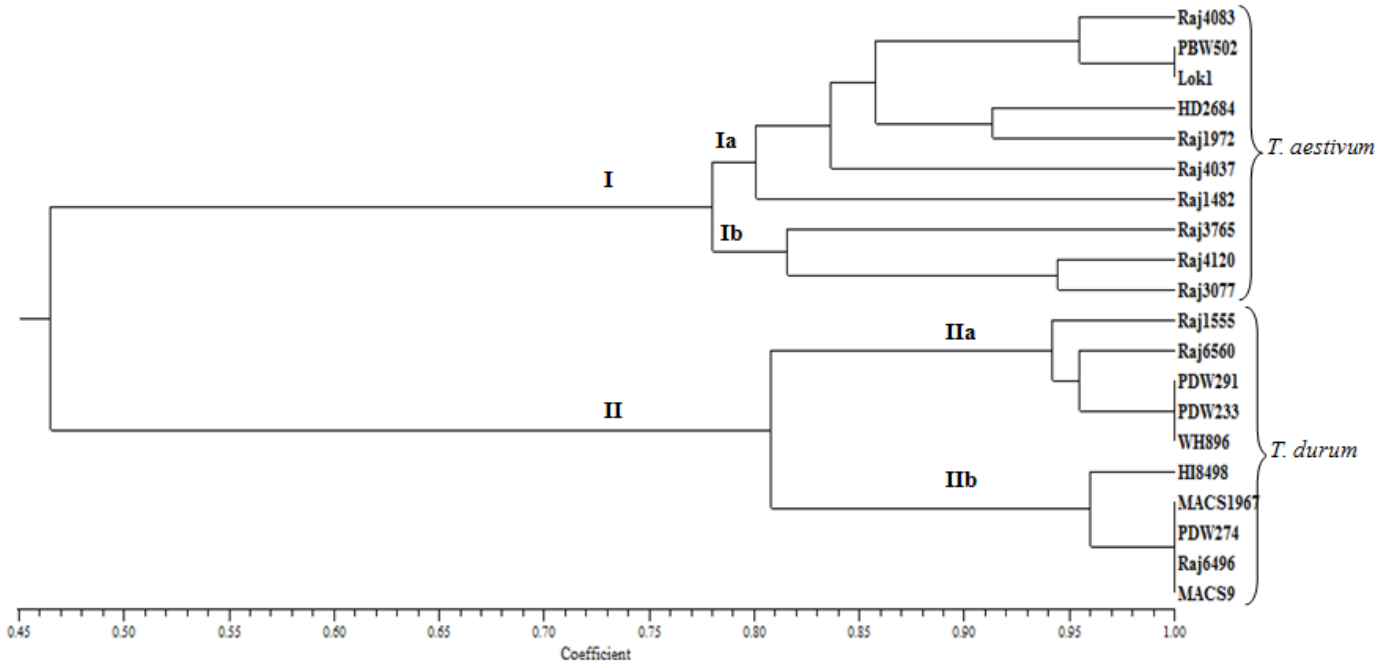

Figure 2. Dendrogram showing relationship among 20 *Triticum* genotypes generated by UPGMA analysis based on SDS PAGE profiling.

gated; however, their varieties were different but the final result was correlated (Khan et al., 2002). Together with physiochemical and molecular characteristics already reported (Zeb et al., 2006), this study presents a good tool to characterize seed storage protein. The dendrogram calculated from the Jaccard similarity coefficient and unweighted pair group method with averages constructed by HMW and LMW glutenin subunit bands cluster analysis is presented in Figure 2. Genetic diversity of European spelts wheat was evaluated by constructing the dendrogram for HMW and LMW glutenin subunit bands (Xueli et al., 2005). The dendrogram as a whole revealed low genetic diversity at protein levels because most varieties are in the same cluster. Fufa et al. (2005) reported that the genetic diversity estimates based on seed storage protein were lowest because they were the major determinants of end-use quality, which is a highly selected trait. The variety PBW502 and Lok1, PDW291 and PDW233, PDW291 and WHH896, PDW233 and WHH896, MACS1967 and PDW274, MACS1967 and Raj 6496, MACS1967 and MACS9, PDW274 and Raj6496, PDW274 and MACS9, Raj6496 and MACS9 showed 100% similarity with one another.

Conclusion

It is therefore concluded that genetic diversity estimates based on seed storage protein were low because they were the major determinants of end-use quality, which is a highly selected trait. After all seed storage, protein profiles could be useful markers in genotype identification, registration of new varieties, pedigree analysis and in the studies of genetic diversity and classification of adapted cultivars, thereby improving the efficiency of wheat breeding programs in cultivar development.

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