

Genetic Diversity Analyses of Bread Wheat (*Triticum aestivum* L.) Genotypes using Microsatellite Markers

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Abstract

The diversity studies of germplasm forms the basis of genetic improvement in crop plants. Around 900 wheat germplasm lines collected from different national and international germplasm repositories were subjected to repeated cycles of selection for earliness, yellow rust resistance and other agronomic traits over five years under research programme of department and under a University Grant Commission funded research project to ultimately constitute a set of 26 most promising candidate genotypes. These twenty six wheat germplasm lines were studied for genetic diversity using forty-two previously developed microsatellite markers. The study revealed the overall size of PCR products amplified ranged from 100-330 bp, highest polymorphic information content (PIC) value of 0.67 for *Xgwm120* while lowest value of 0.19 was observed for *Xgwm155* with an overall average of 0.48. For gene diversity, lowest value of 0.22 was observed for *Xgwm155* while highest value of 0.72 was observed for *Xgwm120* with an overall average of 0.56. The cluster analysis based on molecular analysis of twenty-six selected genotypes revealed that genotypes with 0.209 dissimilarity index value. WS-25 (WHEAR/SOKOLL*2//QUAIU #1) and WS-6 (FW-921) were found most diverse genotypes with dissimilarity index of 0.724 and the genotypes WS-15 (KACHU/6/YAR/AE.SQUARROSA (783)/4/GOV/AZ//MUS/3/SARA/5/MYNA/VUL//JUN) and WS-17 (PRL/2*PASTOR//KACHU) were reported to be similar. The dendrogram generated using molecular data of polymorphic markers grouped 26 wheat genotypes into 3 distinct main clusters and 10 sub clusters. The overall results have revealed that wheat genotypes WS-25 (WHEAR/SOKOLL*2//QUAIU #1) and WS-6 (FW-921) although being exotic and Indian in origin record maximum diversity for different traits and as such could be recommended for trait improvement studies in wheat. Further, such understanding of diverse genotypes can be helpful for selection of parental lines to breed for economically important traits in wheat.

Keywords: Bread wheat, Candidate lines, Earliness, Genetic diversity, SSR markers

1. Introduction

Wheat (*Triticum aestivum* L.) is an important cereal crop of the world both in terms of area and production, it constitutes an important staple food of majority of world

population (Grote *et al.*, 2021). Bread wheat contributes 95% to total wheat production (Sharma *et al.*, 2013). In India, wheat is the second most important cereal crop after



rice and is grown on an area of 31.88 million hectares with a production of 113.29 million tones (Anonymous, 2025). About 90% of world wheat area is occupied by bread wheat because it has an extremely buffered genotype due to polyploidy (Fariset *et al.*, 2002) and three divergent alleles may be harboured at each locus (Pradhan, 2011). It has one of the largest and most complex genomes of cereals. It is allohexaploid with chromosome number 42 ($2n = 6x = 42$) with three genetically related genomes A, B, and D. The genetic diversity is the most important tool in the hands of plant breeders in choosing the right type of parents for hybridization programme. The narrow genetic diversity is more specifically a problem in breeding for adaptation to biotic and abiotic stresses. Therefore, it is necessary to investigate the genetic diversity in wheat germplasm in order to broaden the genetic base for economically important traits and as such contribute for future breeding programmes (Uddin *et al.*, 2008, Nelwadker *et al.*, 2017, Wang *et al.*, 2025).

The molecular markers offer the best estimate of genetic diversity since they are independent of the environmental effects. In recent years, several studies have been conducted, where molecular assays were applied to assess genetic diversity among wheat cultivars. Several molecular markers like random amplified polymorphic DNA (RAPD), inter simple sequence repeat (ISSR) and simple sequence repeat (SSR) markers etc. are presently available in public domain to assess the diversity at molecular level in crop plants (Palombi and Damiano, 2002, Tsonev *et al.*, 2021). Although the application of each class of molecular markers depends upon the objective of the study, the

simple sequence repeats or microsatellites are ideally used for diversity studies because of their locus specificity, ease of use, codominance nature and their ability to detect high level of polymorphism (Roder *et al.*, 1998a; 1998b; Landjeva *et al.*, 2007; Laido *et al.*, 2013). Hence, keeping in view the importance of diversity analysis the present study has been planned to estimate the magnitude of genetic divergence present in the twenty-six wheat germplasm lines and to identify the diverse genotypes that can serve as base material for devising future breeding programmes.

2. Materials and Methods

2.1. Plant materials

Around 900 wheat germplasm lines were collected from different national and international germplasm repositories viz., CIMMYT, Mexico; CIMMYT Centre, Ankara, Turkey; IARI sub-station, Wellington, Tamil Nadu; IIWBR, Karnal, IIWBR Flowerdale, Shimla; PAU Ludhiana; SKUAST-K, Shalimar, Srinagar. The collections were subjected to repeated cycles of selection for earliness, yellow rust resistance and other important agronomic characters over five years under research programme of department on wheat to ultimately constitute a set of twenty-six wheat germplasm lines. The experimental materials for the study constituted those pre-selected twenty six (26) wheat (*Triticum aestivum* L.) germplasm lines that include two local check varieties Shalimar Wheat-1 (WS-9) and Shalimar Wheat-2 (WS-12) (Table 1) that were grown in the experimental fields of Division of GPB, FoA, Wadura, SKUAST-K and whose morphological characterization has been reported by Nelwadker *et al.*, (2017).

Table 1: List of wheat germplasm lines studied for molecular characterization

Sl. No.	Genotype/ Germplasm lines	Code	Source	Unique traits
1	FW-226	WS-1	IARI, Wellington	Earliness and Yield attributing traits
2	FW-920	WS-2	IARI, Wellington	Earliness
3	FLW-16	WS-3	IIW&BR, Karnal	Earliness and yield attributing traits
4	FW-638	WS-4	IARI, Wellington	Yield attributing traits
5	FLW-1306	WS-5	IIW&BR, Karnal	Yellow rust resistance
6	FLW-921	WS-6	IIW&BR, Karnal	Yellow rust resistance, Yield attributing traits
7	<i>Yr10</i> reference line	WS-7	CIMMYT, Mexico	Yellow rust resistance
8	<i>Y18</i> reference line	WS-8	CIMMYT, Mexico	Yellow rust resistance
9	Shalimar Wheat-1	WS-9	SKUAST- Kashmir	Yield attributing traits
10	Selection-42	WS-10	IIW&BR, Karnal	Yield attributing traits



11	FLW-9	WS-11	IIW&BR, Karnal	Yield attributing traits
12	Shalimar Wheat-2	WS-12	SKUAST- Kashmir	Earliness, Yield attributing traits
13	FW-1142	WS-13	IARI, Wellington	Yellow rust resistance
14	BISA-3049	WS-14	BISA, Ludhiana	Yellow rust resistance Yield attributing traits
15	BISA-6012	WS-15	BISA, Ludhiana	Yellow rust resistance
16	BISA-4041	WS-16	BISA, Ludhiana	Yellow rust resistance
17	BISA-5031	WS-17	BISA, Ludhiana	Yield attributing traits, Yield attributing traits
18	BISA-7049	WS-18	BISA, Ludhiana	Yield attributing traits
19	BISA-11058	WS-19	BISA, Ludhiana	Yield attributing traits
20	BISA-11057	WS-20	BISA, Ludhiana	Yield attributing traits, Yield attributing traits
21	BISA-11045	WS-21	BISA, Ludhiana	Yield attributing traits
22	BISA-10041	WS-22	BISA, Ludhiana	Yield attributing traits
23	BISA-10044	WS-23	BISA, Ludhiana	Yield attributing traits
24	BISA-10046	WS-24	BISA, Ludhiana	Yellow rust resistance Yield attributing traits
25	BISA-3029	WS-25	BISA, Ludhiana	Yellow rust resistance, Yield attributing traits
26	BISA-11047	WS-26	BISA, Ludhiana	Yield attributing traits

2.2 Diversity analysis

The genetic diversity analysis of wheat germplasm lines was carried out in Molecular Biology Laboratory, Division of Genetics and Plant Breeding, Faculty of Agriculture and Regional Research Station, Wadura, Sopore, SKUAST-Kashmir using forty-two (42) microsatellite markers (Table 2).

The DNA was isolated from twenty six wheat germplasm lines using CTAB method (Saghai-Maroo *et al.*, 1984). Each isolated DNA sample was quantified and master/stock samples of DNA of each entry was diluted at 25 µg/µl to form working sample. The PCR reactions were set up following standard protocols by using a set of 42 different primers on each genotype. The markers were selected from molecular linkage maps of wheat and chosen in such a way to cover all 21 chromosomes of wheat genomes so that maximum available diversity among the genotypes under study can be targeted. In total 42 primers (2 from each chromosome) were used for analyzing the banding pattern of the amplified product through Agarose gel and PAGE (poly acryl amide gel electrophoresis) system. The banding pattern obtained after using all 42 primers on each genotypes under study was scored and recorded in the form of binary scores (presence of band= 1 and absence of band=0). The binary scores (1/0) of amplified products obtained from both Agarose gel electrophoresis and PAGE were used for estimation of genetic diversity among wheat germplasm lines under study using DARwin 5.0 (Perrier and Jacquemoud-Collet, 2006) software

programme. The binary (presence/absence = 1/0) data was fed to the software for generating dendrogram, where Jaccard's coefficient with 1000 boot stripping was used to generate dissimilarity matrix and unweighted neighbor joining method was used for constructing a tree. The banding patterns of all genotypes against each primer was compared. Bands present in one genotype and absent in other genotype was graded as variable and used to score for polymorphism.

In order to check the informativeness and discriminatory power of genic SSR markers utilized in this study, certain parameters like polymorphic information content, gene diversity and number of alleles were calculated by using software Power Marker v3.25 (<http://www.powermarker.net>).

2.3. Polymorphism information content (PIC)

Average PIC value indicates the ability of markers to differentiate the genotypes. It was calculated by following (Roldan-Ruiz *et al.*, 2000).

$$PIC = 1 - \sum (f_i)^2$$

Where f_i = frequency of bands present

3. Results and Discussion

3.1. Diversity analysis using microsatellite markers

A set of twenty six (26) preselected wheat genotypes were characterized at molecular level using forty two genic SSRs. The trait phenotyping of this set of genotypes has been reported by Nelwadker *et al.*, (2017). Forty two molecular



Table 2: List of microsatellite (SSR) markers used for molecular characterization studies

Primer	Chromosome number	Annealing temperature (°C)	Amplicon size (bp)*	Forward primer sequence 5' to 3'	Reverse primer sequence 3' to 5'
<i>Xcfa2056</i>	1A	52.35°	268	TGGAAGTTACCCCTGTCGGTC	GAAGAGAGGAGGAGTGCCCT
<i>Xcfa2135</i>	1A	48.55°	175	TGCCTAAAATCTAAATGCCCCG	GGATAATGTGCATGTTCACCG
<i>Xgwm33</i>	1B	51.80°	-	GGA GTC ACA CTT GTT TGT GCA	CAC TGC ACA CCT AAC TAC CTG C
<i>Xgwm124</i>	1B	49.70°	190	GCC ATG GCT ATC ACC CAG	ACT GTT CGG TGC AAT TTG AG
<i>Xgwm458</i>	1D	47.85°	115	AAT GGC AAT TGG AAG ACA TAG C	TTT GCA ATG TTG ATT TGG C
<i>Xcfd15</i>	1D	51.15°	179	CTCCCGTATTGAGCAGGAAG	GGCAGGTGGTGTGATGATCT
<i>Xgwm472</i>	2A	50.05°	150	TTG CTA CCA TGC ATG ACC AT	TTT ACC TCG ATT GAG GTC CT
<i>Xgwm71.1</i>	2A	51.95°	126	GGC AGA GCA GCG AGA CTC	CAA GTG GAG CAT TAG GTA CAC G
<i>Xgwm55.1</i>	2B	51.05°	122	GCA TGT GGT ACA CTA GGT GCC	TCA TGG ATG CAT CAC ATC CT
<i>Xgwm120</i>	2B	47.35°	162	GAT CCA CCT TCC TCT CTC TC	GAT TAT ACT GGT GCC GAA AC
<i>Xgwm52</i>	3D	50.00°	142	CTA TGA GGC GGA GGT TGA AG	TGC GGT GCT CTT CCA TTT
<i>Xgwm161</i>	3D	49.00°	154	GAT CGA GTG ATG GCA GAT GG	TGT GAA TTA CTT GGA CGT GG
<i>Xgwm160</i>	4A	48.90°	184	TTT AAT TCA GTC TTG GCT TGG	CTG CAG GAA AAA AAG TAC ACC C
<i>Xgwm601</i>	4A	49.80°	152	ATC GAG GAC GAC ATG AAG GT	TTA AGT TGC TGC CAA TGT TCC
<i>Xgwm107</i>	4B	49.75°	188	ATT AAT ACC TGA GGG AGG TGC	GGT CTC AGG AGC AAG AAC AC
<i>Xgwm165</i>	4B	48.85°	257	TGC AGT GGT CAG ATG TTT CC	CTT TTC TTT CAG ATT GCG CC
<i>Xgwm608</i>	4D	51.80°	151	ACA TTG TGT GTG CCG CC	GAT CCC TCT CCG CTA GAA GC
<i>Xcfd39</i>	4D	49.50°	175	CCACAGTACATCACTTTCCCTT	CAAAAGTTGAAACAGCAGCCA
<i>Xgwm126</i>	5A	51.20°	196	CAC ACG CTC CAC CAT GAC	GTT GAG TTG ATG CCG GAG G
<i>Xgwm156</i>	5A	49.80°	300	CCA ACC GTG CTA TTA GTC ATT C	CAA TGC AGG CCC TCC TAA C
<i>Xgwm159</i>	5B	51.65°	189	GGG CCA ACA CTG GAA CAC	GCA GAA GCT TGT TGG TAG GC
<i>Xgwm213</i>	5B	52.20°	162	TGC CTG GCT CGT TCT ATC TC	CTA GCT TAG CAC TGT CGC CC
<i>Xgwm292</i>	5D	51.35°	214	TCA CCG TGG TCA CCG AC	CCA CCG AGC CGA TAA TGT AC
<i>Xgwm565</i>	5D	52.40°	142	GCG TCA GAT ATG CCT ACC TAG G	AGT GAG TTA GCC CTG AGC CA
<i>Xgwm617</i>	6A	50.90°	133	GAT CTT GGC GCT GAG AGA GA	CTC CGA TGG ATT ACT CGC AC
<i>XWMC1EB</i>	6A	51.05°	197	TCATTCGTTGCAGATACACCAC	TCAATGCCCTGTTCTGACCT
<i>Xcfd13</i>	6B	48.70°	254	CCACTAACCAAGCTGCCAAT	TTTTTGGCATTGATCTGCTG
<i>Xiomc95</i>	6B	47.65°	196	GTTTTTGTGATCCCGGGTTT	CATGCCGTCAAGTCAAGTTTT
<i>Xgwm55</i>	6D	50.85°	128	GCA TCT GGT ACA CTA GCT GCC	TCA TGG ATG CAT CAC ATC CT
<i>Xgwm325</i>	6D	49.15°	133	TTT CTT CTG TCG TTC TCT TCC C	TTT TTA CGC GTC AAC GAC G
<i>Xgwm130</i>	7A	50.85°	126	AGC TCT GCT TCA CGA GGA AG	CTC CTC TTT ATA TCG CGT CCC
<i>Xgwm332</i>	7A	51.60°	290	AGC CAG CAA GTC ACC AAA AC	AGT GCT GGA AAG AGT AGT GAA GC
<i>Xgwm46</i>	7B	50.00°	186	GCA CGT GAA TGG ATT GGA C	TGA CCC AAT AGT GGT GGT CA
<i>Xgwm302</i>	7B	50.05°	277	GCA AGA AGC AAC AGC AGT AAC	CAG ATG CTC TTC TCT GCT GG
<i>Xgwm295</i>	7D	52.30°	254	GTG AAG CAG ACC CAC AAC AC	GAC GGC TGC GAC GTA GAG
<i>Xgwm635</i>	7D	51.40°	99	TTC CTC ACT GTA AGG GCG TT	CAG CCT TAG CCT TGG CG
<i>Xgwm539</i>	2D	50.70°	143	CTG CTC TAA GAT TCA TGC AAC C	GAG GCT TGT GCC CTC TGT AG
<i>Xcfd267</i>	2D	53.60°	280	GTGCGTGTGTAGCAGCTC	CTCTGTGTCAGGTCGT
<i>Xgwm30</i>	3A	51.15°	196	ATC TTA GCA TAG AAG GGA GTG GG	TTT TGC ACC CTG GGT GAT
<i>Xgwm155</i>	3A	47.10°	143	CAA TCA TTT CCC CCT CCC	AAT CAT TGG AAA TCC ATA TGCC C
<i>Xgwm77</i>	3B	51.80°	-	ACA AAG GTA AGC AGC ACC TG	ACC CTC TTG CCC GTG TTG
<i>Xgwm264</i>	3B	48.75°	-	GAG AAA CAT GCC GAA CAA CA	GCA TGC ATG AGA ATA GGA ACT G



markers (SSRs) were used for diversity analysis of the set, which gave recordable amplicons in the size range of 99-330 bps. Out of forty two SSR markers used, twenty eight (66.6%) markers were polymorphic. The amplified products were resolved on agarose (Figure 1) and PAGE gels (Figure 2). The informativeness of these SSR primers was depicted by various parameters such as number of alleles, major allele frequency, polymorphic information content and gene diversity (Table 3). Highest PIC value of 0.67 was observed in *Xgwm120* while lowest value of 0.19 was observed in *Xgwm155* with an overall average of 0.48. The number of alleles per locus ranged from 2

(*Xgwm46*, *Xgwm155*, *Xgwm47.2*, *Xgwm107* and *Xgwm126*) to 5 (*Xgwm332* and *Xwmc95*) with an average of 3 alleles per locus. The values for major allele frequency ranged from 0.35 (*Xgwm635*) to 0.88 (*Xgwm155*) with an overall average of 0.55. For gene diversity, lowest value of 0.22 was observed for *Xgwm155* while highest value of 0.72 was observed for *Xgwm120* with an overall average of 0.56. Singh *et al.*, (2025) reported genetic diversity of 22 wheat genotypes using 8 SSR markers polymorphic primers with average PIC values of 0.73 per locus were recorded and two markers *wmc398* and *wmc335* were identified as best markers for studying diversity analysis in wheat.

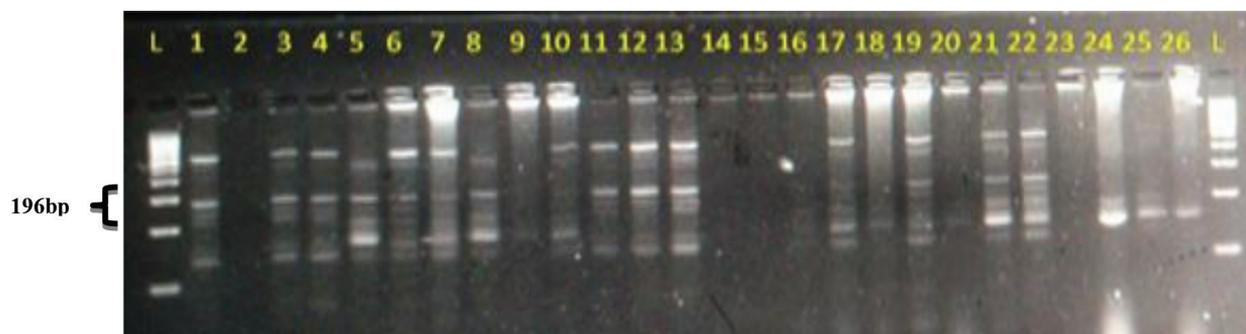


Fig. 1: A representative agarose gel image of 26 wheat genotypes using SSR marker *Xwmc95*

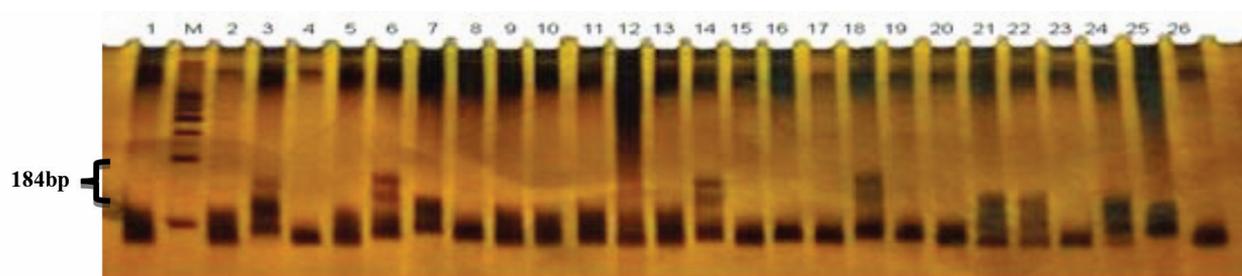


Fig. 2: A representative gel image of PAGE on 26 wheat genotypes using SSR marker *Xwmc160*

Table 3: Parameters depicting informativeness of microsatellite (SSR) primers used for the study

Marker Name	Number of allele per locus	Major allele Frequency	Polymorphic Information Content (PIC)	Gene diversity
<i>Xgwm160</i>	3	0.58	0.46	0.54
<i>Xgwm332</i>	5	0.67	0.49	0.52
<i>Xgwm130</i>	4	0.50	0.60	0.65
<i>Xgwm124</i>	Monomorphic	-	-	-
<i>Xgwm120</i>	4	0.40	0.67	0.72
<i>Xgwm46</i>	2	0.68	0.34	0.43
<i>Xgwm155</i>	2	0.88	0.19	0.22
<i>Xcfa2135</i>	Monomorphic	-	-	-
<i>Xgwm77</i>	3	0.58	0.43	0.53
<i>Xgwm292</i>	3	0.47	0.45	0.55
<i>Xgwm601</i>	3	0.50	0.55	0.62
<i>Xgwm33</i>	Monomorphic	-	-	-
<i>Xgwm264</i>	3	0.65	0.39	0.48



Xgwm565	3	0.43	0.57	0.64
Xgwm458	Monomorphic	-	-	-
Xgwm71.1	Monomorphic	-	-	-
Xgwm161	Monomorphic	-	-	-
Xgwm47.2	2	0.80	0.27	0.32
Xgwm55.1	3	0.53	0.49	0.58
Xgwm107	2	0.63	0.36	0.47
Xgwm165	Monomorphic	-	-	-
Xgwm608	Monomorphic	-	-	-
Xcfd39	3	0.46	0.52	0.60
Xgwm213	3	0.77	0.33	0.37
Xgwm156	Monomorphic	-	-	-
Xgwm159	Monomorphic	-	-	-
Xcfa2056	3	0.64	0.44	0.51
Xcf15	3	0.42	0.57	0.65
Xgwm52	4	0.53	0.55	0.62
Xgwm126	2	0.52	0.37	0.50
Xgwm617	Monomorphic	-	-	-
Xgwm55	Monomorphic	-	-	-
XWMCIEB	3	0.42	0.58	0.65
Xcfd13	4	0.54	0.56	0.61
Xwmc95	5	0.48	0.64	0.68
Xgwm295	Monomorphic	-	-	-
Xcfd267	Monomorphic	-	-	-
Xgwm325	3	0.42	0.58	0.65
Xgwm302	3	0.67	0.45	0.50
Xgwm635	3	0.35	0.59	0.67
Xgwm539	4	0.45	0.64	0.69
Xgwm30	3	0.55	0.48	0.57
Mean	3	0.55	0.48	0.56

3.2. Phylogenetic Studies

The phylogenetic studies (Figure 3) divided the 26 genotypes into 3 clusters viz., cluster-I, cluster-II (Major cluster) and cluster-III (Minor cluster). Cluster-II and cluster-III were further divided into sub clusters. The results of diversity analysis revealed that the genotypes BISA-3029/ WS-25 (WHEAR/SOKOLL*2//QUAIU #1) and FW-921 (WS-6) as the most diverse genotypes with dissimilarity index of 0.724 followed by Shalimar Wheat-1 (WS-9) and FW-638 (WS-4) with 0.720 and BISA-11047/WS-26 (MUTUS*2/HARIL #1) and *Yr10* reference (WS-7) with 0.711. The genotypes BISA-6012/ WS-15 (KACHU/6/YAR/AE.SQUARROSA (783) /4/ GOV/AZ//MUS/3/SARA/5/MYNA/VUL//JUN) and BISA-5031/ WS-17 (PRL/2*PASTOR//KACHU) were found to be similar with 0.209 dissimilarity index value. The factorial analysis grouped the 26 genotypes under study into 4 clusters (Figure 4).

Genetic diversity in cultivated crops is essential for successful breeding and creation of new cultivars. Estimating the genetic diversity of wheat germplasm can help in identifying diverse parental combinations and creating segregating progeny with high genetic variability for selection (Goyal *et al.*, 2015, Wang *et al.*, 2025). A narrow genetic base for economically important traits increases the risk factor, while the traits with broad genetic base are generally stable. The commercially viable traits like, yield and disease resistance could be improved by expanding their genetic diversity in bread wheat (Nielsen *et al.*, 2014, Wang *et al.*, 2025).

Polymorphism among individual genotypes is observed due to variation in DNA sequences present in their chromosomes. Higher polymorphic bands of primers indicate their efficiency to study genetic diversity and discrimination of genotypes (Pradhan *et al.*, 2004). Simple sequence repeats or microsatellites are used in a lot of research because of their locus specificity, ease of use,



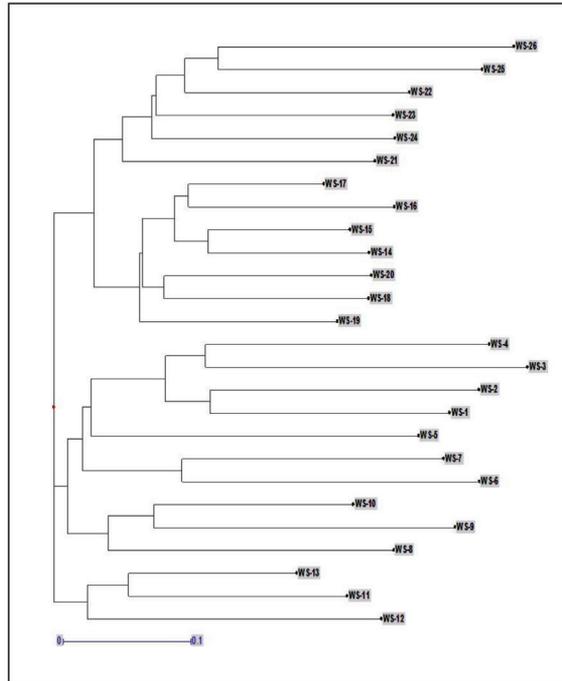


Fig. 3: Dendrogram showing clustering of different wheat genotypes based on SSR data

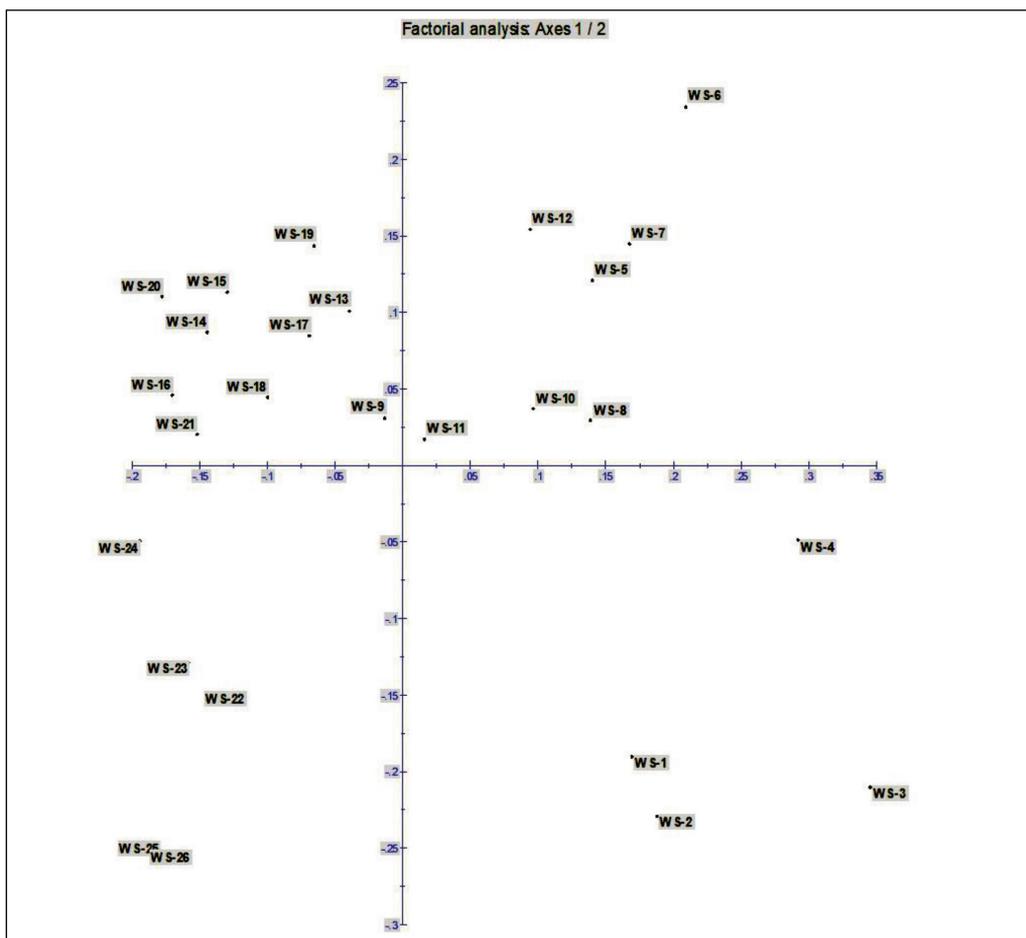


Fig. 4: Factor analysis of twenty six (26) wheat genotypes into different co-ordinates showing relationships among different wheat genotypes.



codominance, and high polymorphism (Roder *et al.*, 1998; Landjeva *et al.*, 2007; Laido *et al.*, 2013, Singh *et al.*, 2025). Microsatellite markers are useful for marker-assisted selection, identifying quantitative trait loci, genetic diversity and labeling of stress-tolerant genes in wheat or wild relations (Landjeva *et al.*, 2007; Ijaz and Khan, 2009). SSR markers have a high level of PIC in comparison with other types of molecular markers (Gupta *et al.*, 2008, Singh *et al.*, 2025). In the previous studies (Henkrar *et al.*, 2016) it was reported that PIC values in wheat ranged between 0.07 to 0.84 with an average genetic diversity of 0.48 in Moroccan wheat cultivars. Mattar *et al.*, (2009) employed eleven microsatellites generating 44 alleles with an average of 4 alleles per locus comparable to the value of 3.17 alleles per locus. The phylogenetic tree divided the 26 genotypes into 3 clusters viz., cluster-I, cluster-II (Major cluster) and cluster-III (Minor cluster). Cluster-II and cluster-III were further divided into sub clusters. The factorial analysis grouped the 26 genotypes under study into 4 clusters (Figure 4). Based on previous literature, these studies have been reported for diversity analysis and determination of genetic variability in bread wheat (Tilahun *et al.*, 2016, Choudhary *et al.*, 2022, Singh *et al.*, 2025). Singh *et al.*, (2025) reported genetic diversity of 22 wheat genotypes for 13 traits using eight SSR markers. The study identified SSR markers *wmc398* and *wmc335* as best markers for diversity analysis.

Similar study was reported for Turkish bread wheat genotypes in which the genetic diversity was estimated based on morphological and molecular observations and cultivars were clustered into two groups (Erayman *et al.*, 2016). This facilitates an important criterion for identification of diverse genotypes so that they can be incorporated into future breeding programmes. The study has identified two genotypes namely BISA-3029 (WS-25) and FW-921 (WS-6) as the most diverse genotypes based on the molecular characterization. The identification of the highly polymorphic SSRs and diversity of twenty six genotypes based on their molecular characterization shall facilitate the understanding of these diverse genotypes which can be helpful for selection of parental lines to breed for agronomic traits. However, there is further need of involving large number of markers as well as genotypes to have a better picture of germplasm structure for its exploitation in crop improvement programs of India.

The estimation of genetic diversity in crop plants is essential for successful breeding and creation of new cultivars. This can help in identifying diverse parental combinations and creating segregating progenies with high genetic variability for efficient selection. The work presented here inferred the identification of the highly polymorphic SSRs and diversity of pre-selected twenty six genotypes based on molecular characterization. Such study facilitates the understanding of these diverse genotypes which can be helpful for selection of parental lines to breed for agronomic traits. However, there is further need of involving large number of markers as well as genotypes to have a better picture of germplasm structure to exploit for crop improvement programs. The present study identified that the two genotypes namely BISA-3029 (WS-25) and FW-921 (WS-6) are the most diverse genotypes based on the molecular characterization.

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Authors' contributions

RRN, MAK and RRM- compiled and edited the manuscript; SK helped in the statistical analysis of the findings and in editing of the manuscript.

Conflict of interest

The authors declare that they have no conflict of interest.

Ethical Approval

The article doesn't contain any study involving ethical approval.

Use of Generative AI or AI assisted technologies

Authors declare that no Generative AI or AI assisted technologies have been used in preparation of this manuscript



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