RESEARCH ARTICLE

SABRAO Journal of Breeding and Genetics
58 (5) 2090-2098, 2026
http://doi.org/10.54910/sabrao2026.58.5.9
http://sabraojournal.org/
pISSN 1029-7073; eISSN 2224-8978

DURUM WHEAT (*TRITICUM DURUM*) LODGING RESISTANCE EVALUATION UNDER RAINFED CONDITIONS USING MORPHOLOGICAL TRAITS AND DNA MARKERS

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SUMMARY

The evaluation of lodging resistance in 10 durum wheat (*Triticum durum* Desf.) genotypes under rainfed conditions in Uzbekistan was this study's aim using morphological traits and SSR marker analysis. Field studies revealed considerable variations among genotypes for plant height and spike length, with these traits found in association with lodging susceptibility. The SSR markers' analysis identified three polymorphic markers (Xgwm181, Xgwm312, and Xgwm63) associated with lodging resistance. Favorable alleles of these markers succeeded in their detection in several durum wheat genotypes, particularly in the genotypes D-21 and D-7, indicating their potential for tolerance to lodging. Phylogenetic analysis based on marker polymorphicm revealed a genetic relationship among the studied genotypes. Overall, the results revealed these SSR markers can be reliable tools for confirming lodging resistance in durum wheat genotypes and can be favorably effective in marker-assisted selection for breeding programs.

Keywords: Durum wheat (*T. durum* Desf.), lodging resistance, morphological traits, SSR markers, phylogenetic analysis, rainfed conditions

Key findings: The study identified lodging-resistance-associated alleles in durum wheat (*T. durum* Desf.) genotypes using the SSR markers Xgwm181, Xgwm312, and Xgwm63. The favorable allele Xgwm181 (225 bp) entailed detection in genotypes D-7 and D-21, while Xgwm312 (205 bp) occurred in several other genotypes.

Communicating Editor: Prof. Dr. Zahoor Ahmed Soomro

Manuscript received: March 24, 2026; Accepted: June 28, 2026.
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Citation: Karshiboev KHKH, Amirova MS, Khalikulov DKH, Nakhalbaev JT, Komilov DJ, Abdukarimov U (2026). Durum wheat (*Triticum durum*) lodging resistance evaluation under rainfed conditions using morphological traits and DNA markers. *SABRAO J. Breed. Genet. 58 (5) 2090-2098. http://doi.org/10.54910/sabrao2026.58.5.9*.

INTRODUCTION

Durum wheat (*Triticum durum* Desf.) is one of the most important cereal crops cultivated in diverse semi-arid and rainfed regions worldwide. Durum wheat is a widely used crop for the production of pasta, semolina, and other food products, due to its high-protein content and desirable quality parameters (Alhabbar *et al.,* 2023). In recent decades, enhanced climatic variability, including irregular rainfall patterns, high temperatures, and robust winds, has remarkably affected the wheat production, particularly in dryland cropping systems (Tamrazov, 2022). Under such harsh environmental conditions, lodging has become one of the major constraints limiting durum wheat productivity and grain quality.

Lodging refers to the permanent displacement of plant stems from their upright position and could occur due to a weak-stem strength, an excessive plant height, and unfavorable environmental factors, such as heavy rainfall with wind. Lodging considerably reduces the photosynthetic efficiency, interferes with the nutrient transport system, and makes harvesting more difficult, ultimately resulting in substantial yield losses (Gregersen *et al.,* 2008; Chapman *et al.,* 2021). Previous studies have reported that severe lodging may reduce the durum wheat grain yield up to 30%–50%, depending on the severity and timing of the lodging (Nouri *et al.,* 2011). Therefore, improvement in genotypes' lodging resistance has become an important objective in wheat breeding programs.

Morphological traits, such as plant height, stem diameter, internode length, and spike weight, have an association commonly with lodging resistance in wheat. However, these polygenic traits often bear influences from environmental conditions and may result in considerable variations under rainfed conditions (Meghawal *et al.,* 2021; Mekaoussi *et al.,* 2021). Consequently, phenotypic selection alone may be insufficient to accurately identify the lodging-resistant genotypes. In this context, molecular marker technology provides powerful tools for identifying genetic factors associated with these essential agronomic traits, including lodging resistance (Galal *et al.,* 2023).

The DNA marker technologies have been the widely used method in plant breeding for the identification of genes and quantitative trait loci (QTLs) associated with desirable traits in different crop plants. Various molecular marker systems, including SSR (simple sequence repeat), RAPD (random amplified polymorphic DNA), AFLP (amplified fragment length polymorphicm), and SNP (single nucleotide polymorphicm), have succeeded in their applications in the sustainable improvement of the durum wheat's genetic makeup (Maccaferri *et al.,* 2003; Dagnaw *et al.,* 2023). These markers allow detecting genetic polymorphicm among the genotypes, evaluating genetic diversity, and identifying the marker-trait associations that can be effective in marker-assisted selection (MAS) (Naser *et al.,* 2023).

Several studies have reported that lodging resistance is a complex trait controlled by multiple genes associated with plant architecture and stem strength in wheat. Important dwarfing genes, such as *Rht-B1* and *Rht-D1,* have been successful in their wide usage in wheat breeding to reduce plant height and improve lodging resistance. However, lodging resistance is not solely distinctive of plant height; other traits, such as stem mechanical strength and root anchorage, also play pivotal roles (Richards and Townley-Cmith*,* 1987). Therefore, the identification of molecular markers linked with lodging resistance traits can greatly enhance the efficiency of breeding programs aimed at developing lodging-resistant wheat genotypes.

The molecular markers' application is particularly valuable under rainfed conditions where environmental variability can robustly influence the phenotypic expression. By integration of field evaluation with molecular marker analysis, the researchers can better understand the genetic basis of lodging resistance and also identify promising genotypes with better adaptation to dryland conditions (Meliev *et al.,* 2023). Such approaches may considerably contribute to the

development of improved wheat cultivars that are more stable and productive under challenging environmental conditions.

Despite the importance of lodging resistance in durum wheat production, relatively limited information is available regarding the molecular markers associated with this viable trait under rainfed conditions. Therefore, investigating the genetic diversity of durum wheat genotypes and identifying molecular markers associated with lodging resistance is essential for improving the wheat breeding efficiency (Turaev *et al.,* 2023).

Therefore, based on the aforesaid discussion, the relevant study aimed to evaluate lodging resistance among the durum wheat genotypes grown under rainfed conditions and identify the genetic variability using DNA markers technology. These results could provide useful information for the selection of lodging-resistant genotypes and support the development of improved durum wheat cultivars adapted to dryland environments.

MATERIALS AND METHODS

Plant materials and field experiment

This study used 10 durum wheat (*T. durum* Desf.) lines as experimental plant materials (Table 1). The selected genotypes underwent evaluation to determine their lodging resistance under rainfed conditions. Field experiments conducted took place at the Jizzakh Rainfed Agricultural Research Institute, Uzbekistan. The experiment layout was in a randomized complete block design (RCBD) with three replications. Each durum wheat genotype entailed sowing in separate plots under natural rainfed conditions.

Evaluation of the biometric parameters of the plants in field experiments transpired at the full maturity stage of the wheat. Morphological measurements proceeded on the main stems of 10 randomly selected sample plants from each replicate. Plant height (cm) determination was by measuring the distance from the soil surface (base of the stem) to the topmost tip of the main spike, with the awns excluded from this measurement. The spike length (cm) on the main stem bore detection by measuring the distance from its lowest node (base of the spike) to the tip of the topmost spikelet. Similar to the methodology for measuring plant height, awn length was not considered when measuring spike length. These parameters served to evaluate the lodging resistance potential of the studied durum wheat genotypes and identify the promising lines suitable for cultivation under rainfed conditions.

Weather data

During the experimental year, from October 2024 to June 2025, the weather comprised drought conditions. The annual precipitation was 320.8 mm, which was 41.1 mm below the long-term average. The mean annual air temperature was 14.4 °C, exceeding the long-term average by 1.8 °C. The average relative humidity was 61%. Notably, dry conditions prevailed during the spring and summer months.

DNA extraction and marker selection

Genomic DNA isolation from fresh young leaves of durum wheat (*T. durum* Desf.) plants used a modified CTAB (cetyltrimethylammonium bromide) extraction protocol. Leaf tissues incur grinding in liquid nitrogen to ensure efficient cell disruption and DNA release. The purity and concentration of the extracted DNA samples entailed measurement using a NanoDrop Eight spectrophotometer (Norboyeva *et al.*, 2025). This study selected a set of 15 DNA markers associated with lodging resistance and plant architecture traits in wheat for molecular analysis. These markers' selection depended on previous studies reporting their linkage with the traits associated with plant height, stem strength, and lodging tolerance. The detailed information on the selected DNA markers are available in Table 2.

SSR analysis and gel electrophoresis

The SSR marker analysis proceeded using the polymerase chain reaction (PCR) technique

Table 1. Triticale cultivars and their origins used in the study.

No.	Cultivar Name	Designation
1	Leukurum-3	D-1
2	Javohir	D-2
3	Yoqut-2014	D-4
4	Billurdon	D-5
5	Kumushdon	D-6
6	Tilladon	D-7
7	RNS-2024/9	D-9
8	RNS-2024/12	D-12
9	RNS-2024/19	D-19
10	RNS-2025/21	D-21

Table 2. Panel of DNA markers associated with lodging resistance in durum wheat.

No.	Marker name	Primer sequence (5'–3')	Chr.	References
1	Xgwm247	F: GCAATCTTTTTTCTGACCACG R: ATGTGCATGTCGGACGC	6B	Röder, 1998; Somers *et al*., 2004
2	Xgwm340	F: GCAATCTTTTTTCTGACCACG R: ACGAGGCAAGAACACACATG	3D	Röder *et al*., 1998; Dodig *et al*., 2010
3	Xgwm547	F: GTTGTCCCTATGAGAAGGAACG R: TTCTGCTGCTGTTTTCATTTAC	2B	Röder *et al*., 1998; Somers *et al*., 2004
4	Xbarc77	F: GCGTATTCTCCCTCGTTTCCAAGTCTG R: GTGGGAATTTCTTGGGAGTCTGTA	3B	Somers *et al.* 2004; Dodig *et al*., 2010
5	Xgwm285	F: ATGACCCTTCTGCCAAACAC R: ATCGACCGGGATCTAGCC	3B	Röder *et al*., 1998
6	Xgwm174	F: GGGTTCCTATCTGGTAAATCCC R: GACACACATGTTCCTGCCAC	5A	Röder *et al*., 1998; Dodig *et al*., 2010
7	Xgwm181	F: TCATTGGTAATGAGGAGAGA R: GAACCATTCATGTGCATGTC	3B	Röder *et al*., 1998
8	Xgwm368	F: CCATTTCACCTAATGCCTGC R: AATAAAACCATGAGCTCACTTGC	5B	Röder *et al*., 1998; Somers *et al*., 2004
9	Xgwm257	F: AGAGTGCATGGTGGGACG R: CCAAGACGATGCTGAAGTCA	2A	Röder *et al*., 1998; Somers *et al*., 2004
10	Xgwm63	F: TCGACCTGATCGCCCCTA R: CGCCCTGGGTGATGAATAGT	7A	Röder *et al*., 1998
11	Xgwm577	F: TGCCTGAGGATGAGGTTGAG R: GACAGGGTAGGAAGACGAGC	7B	Röder *et al*., 1998; Somers *et al*., 2004
12	Xgwm630	F: GGTGCTCAGATGAGGTTGTT R: ACTGCTGTTGCTGTTGCTTC	7D	Röder *et al*., 1998
13	Xgwm312	F: CGTGGATGAGGACGTTGATG R: TGGATGAGGACGTTGATGGC	2D	Röder *et al*., 1998; Somers *et al*., 2004
14	Xgwm296	F: AGGCTGTTGAGGACGTTGAG R: CGTGAGGCTGTTGAGGACGT	7D	Röder *et al*., 1998
15	Xgwm219	F: GTTGACGTTGAGGACGTTGA R: CAGGCTGTTGAGGACGTTGA	6B	Röder *et al*., 1998; Somers *et al*., 2004

following standard molecular protocols (Erjigitov *et al.,* 2025). In the presented study, 15 SSR markers previously reported to be associated with lodging resistance and plant architecture traits in wheat were choices for genotyping (Table 2). These markers reached selection based on their linkage with genomic regions controlling plant height, stem strength, and other traits associated with lodging tolerance.

Performing the PCR amplification used a thermal cycling program comprising an initial denaturation step, followed by 35 amplification cycles. Each cycle included denaturation at 94

°C for 20 s, primer annealing at temperatures ranging from 55 °C to 60 °C for 30 s depending on the primer pair, and an extension at 72 °C for 50 s.

Separating the amplified PCR products used a 2.5% agarose gel electrophoresis to determine the size of DNA fragments. The gels obtained staining with ethidium bromide before visualization using a GelDoc Go Gel Imaging System (Bio-Rad, USA). Fragment sizes estimations utilized the AmpliSize Molecular Ruler (Bio-Rad, USA) as a DNA ladder. The obtained SSR profiles further served for genetic analysis and phylogenetic tree construction using MEGA 11 software to evaluate the genetic relationships among the studied durum wheat genotypes (Erjigitov *et al.,* 2025).

Statistical analysis

Based on various variables, the recorded data's assescment through analysis of variance (ANOVA) evaluated the differences among the durum wheat genotypes. The SSR data sustained analysis for polymorphicm and genetic relationships at $p \leq 0.05$.

RESULTS AND DISCUSSION

Plant height is considerably one of the important morphological traits determining lodging resistance in wheat. Generally, excessively tall genotypes appeared to be more susceptible and vulnerable to lodging due to increased mechanical pressure on the stem, especially under rainfed conditions where robust winds and irregular precipitation patterns frequently occur. In the promising study, the durum wheat genotypes revealed considerable variations for plant height. Overall, the genotypes ranged from 82.3 to 104.2 cm for plant height. The tallest plants recorded were in the genotype D-21 (104.2 ± 1.75 cm), followed by D-7 (103.8 ± 1.03 cm) and D-19 (99.9 ± 0.99 cm). In contrast, the shortest plants resulted in the genotype D-9 (82.3 ± 1.57 cm).

Durum wheat genotypes D-21 and D-7 exhibited the tallest plant height, suggesting that these lines may be more prone to lodging than with medium-height genotypes (Figure 1). The analysis of variance showed significant ($P \leq 0.001$) differences among the genotypes for plant height. The results revealed a substantial genetic variability among the studied durum wheat lines for plant height. Previous studies have also reported that a moderate plant height can play a vital role in improving lodging resistance while maintaining grain yield potential (Khanishova and Azizov*,* 2023). Similarly, studies conducted on durum wheat have demonstrated that plant height occurred to be closely associated with lodging susceptibility and overall plant stability (Luche *et al*., 2015; Mekaoussi *et al.,* 2021).

Spike length may indirectly influence the lodging susceptibility in wheat because longer spikes often contain more spikelets and grains, increasing spike weight and mechanical pressure on the stem. Under rainfed conditions, this additional grain weight could accelerate the risk of lodging, particularly in taller genotypes with weaker stem strength. Among the studied durum wheat genotypes, the analysis of spike length revealed considerable variations. According to results, durum wheat genotypes ranged from 7.3 to 10.0 cm in spike length.

The longest spikes recorded resulted in genotype D-19 (9.9 cm), followed by genotype D-7 (9.4 cm). However, the moderate spike length was evident in the genotypes D-12 and D-21 (8.3–8.4 cm), whereas the shortest spikes recorded arose in the genotypes D-1, D-2, and D-9, ranging from 7.3 to 7.5 cm (Figure 2). In the presented study, genotypes D-19 and D-7 exhibited the lengthiest spike, revealing their potential as promising breeding materials for grain yield formation. Previous studies have also reported that spike length showed a positive association with the number of spikelets and grains per spike (Dagnaw *et al.,* 2023).

Molecular analysis

Based on the obtained results, 15 DNA markers associated with lodging resistance received evaluation using PCR analysis. Among these markers, seven markers were identifiably as polymorphic, indicating the considerable

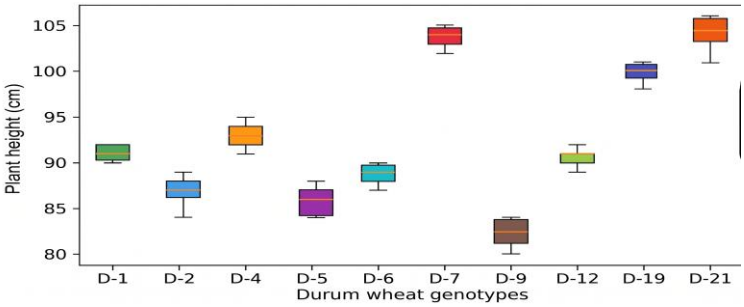

Figure 1. Variation of plant height among the durum wheat genotypes.

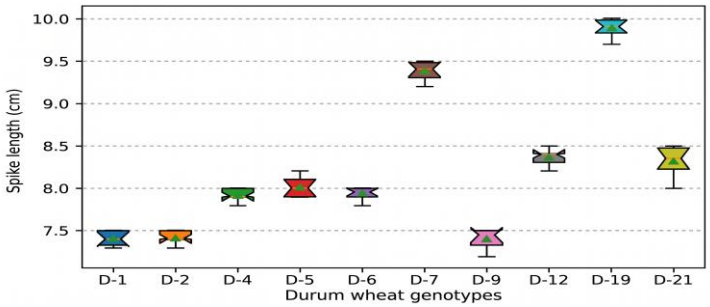

Figure 2. Variation of spike length among the durum wheat genotypes.

Table 3. The SSR markers and their polymorphicm information content (PIC) and heterozygosity (He) values.

No.	DNA markers	Molecular weight (pair bases, pb)	Specific pb.	PIC	He
1	Xgwm181	200, 225	125	0.56	0.59
2	Xgwm312	175, 190, 205	205	0.61	0.66
3	Xgwm63	160, 185, 205, 265	265	0.76	0.81

presence of genetic variability among the studied durum wheat genotypes. The polymorphic markers showed different levels of polymorphicm information content (PIC), reflecting their effectiveness in recognizing genetic diversity in the durum wheat genotypes. Dodig *et al.* (2010) also reported several SSR markers located on different wheat chromosomes displayed significant polymorphicm, which were successfully used for evaluating important agronomic traits, including lodging resistance.

Among the identified polymorphic markers, three markers exhibited PIC values higher than 0.5, indicating the highest level of polymorphicm. The markers Xgwm181 (0.56), Xgwm312 (0.61), and Xgwm63 (0.76) showed the highest PIC values among the tested markers (Table 3). The results suggested these markers occurred to be highly informative and suitable for identifying the genetic variations associated with lodging resistance in durum wheat genotypes. Previous studies have also stated that SSR markers with higher PIC values were more effective in detecting genetic diversity and identifying important agronomic traits in wheat breeding programs. Similarly, Somers *et al.*'s (2004) findings exhibited that SSR markers from the Xgwm312 marker series were highly informative for detecting genetic variations in wheat germplacm For the marker Xgwm181, two alleles with 225 and 200 bp were distinct among the studied durum wheat genotypes. The 225 bp allele appeared to be.

associated with lodging resistance in the genotypes D-7 and D-21. In contrast, the 200 bp allele was evidently in most of the other durum wheat genotypes and had no association with lodging resistance. The results enunciated that the marker Xgwm181 can be useful for distinguishing certain genotypes that may possess lodging resistance.

For the marker Xgwm312, three alleles detected were with 205, 190, and 175 bp. Among these alleles, the 205 bp allele was notably in association with lodging resistance and was existent in the genotypes D-1, D-5, D-6, D-9, D-12, D-19, and D-21. The 190 and 175 bp alleles detected were in other durum wheat genotypes but showed non-association with lodging resistance. The relatively wide distribution of the 205 bp allele suggested the marker Xgwm312 was an informative marker for identifying lodging-resistant wheat genotypes.

For the marker Xgwm63, four alleles entailed detection with 265, 205, 185, and 160 bp. The 265 bp allele was apparently in association with lodging resistance in the genotypes D-1, D-2, D-4, D-5, D-6, D-7, D-9, D-19, and D-21. However, the only genotype D-12 did not possess this allele. The results exhibited that the marker Xgwm63 contains one of the most frequent favorable alleles among the studied markers and may therefore be an important marker for identifying lodging-resistance in durum wheat genotypes (Figure 3).

Overall, molecular marker analysis revealed that genotype D-21 possessed all three resistance-associated alleles (Xgwm181_225, Xgwm312_205, and Xgwm63_265), suggesting that it could be one of the most promising genotypes for lodging resistance among the studied breeding materials. Previous studies have also reported that SSR markers were recognizably as effective molecular tools for detecting genetic diversity in wheat and for studying important agronomic traits. In particular, markers belonging to the Xgwm series have been successful in their wide usage in wheat genetic studies (Röder *et al.,* 1998). Similarly, Dodig *et al.'s* (2010) findings disclosed that SSR markers can be effectively beneficial in the genetic analysis of stem structure and plant architecture traits in wheat. Therefore, the identified favorable alleles confirmed the usefulness of these markers for molecular evaluation of lodging resistance in durum wheat genotypes.

The analysis of variance discovered that the frequency of lodging-resistance alleles did not significantly ($P \leq 0.418$) differ among the plant height groups. The results further revealed that the association of favorable alleles with lodging resistance was not critically dependent on plant height among the studied durum wheat genotypes. Although genotypes with short plant stature generally seemed more resistant to lodging, and the molecular analysis suggested the lodging resistance could also

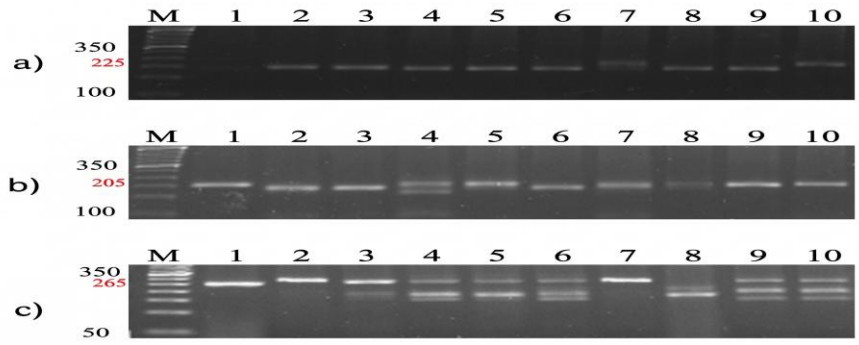

Figure 3. Electropherogram of PCR amplicons for polymorphic DNA markers associated with lodging resistance in durum wheat samples. M - Molecular weight marker (50 bp). 1-D-1, 2-D-2, 3-D-4, 4-D-5, 5-D-6, 6-D-7, 7-D-9, 8-D-12, 9-D-19, and 10-D-21. a- Xgwm181, b- Xgwm312, and c- Xgwm63.

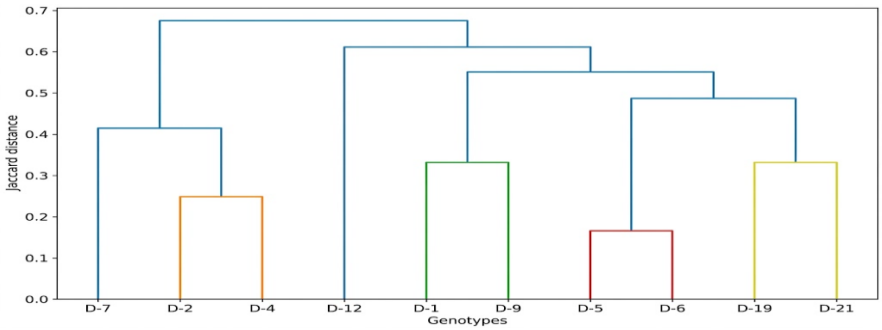

Figure 4. Phylogenetic dendrogram of durum wheat genotypes based on polymorphicm of SSR markers associated with lodging resistance.

have the control of other genetic factors independent of plant height. In contrast, a significant ($P \leq 0.002$) effect was noticeable among the evaluated SSR markers. The result enunciated the markers differed in their ability to detect the lodging-associated alleles. Among the analyzed markers, Xgwm63 and Xgwm312 showed a higher frequency of favorable alleles, indicating these markers may be particularly informative for identifying lodging-resistant genotypes.

Phylogenetic analysis based on SSR marker data showed that the durum wheat genotypes bore groupings according to the similarity of lodging-resistance alleles. The closest genetic relationship was evident between the genotypes D-5 and D-6. Similarly, the genotypes D-19 and D-21 formed a closely related group, revealing a higher similarity in alleles associated with lodging resistance (Figure 4). Genotypes D-1 and D-9 also came together, while lines D-2 and D-4 created another group. In contrast, genotype D-7 appeared more distant from the other durum wheat genotypes, signifying differences in its marker profile. Overall, the dendrogram revealed that the durum wheat genotypes entailed partial groupings according to their lodging-resistance-related genetic similarity.

CONCLUSIONS

This timely study evaluated lodging resistance in 10 durum wheat (*T. durum*) genotypes using morphological traits and SSR marker analysis. Significant variations were evident among the durum wheat genotypes for plant height and spike length. Molecular analysis showed the SSR markers Xgwm181, Xgwm312, and Xgwm63 were polymorphic and effective for identifying the lodging-resistance–associated alleles. Among the studied genotypes, D-21 and D-7 showed the presence of favorable alleles associated with lodging resistance, indicating their potential as promising genotypes for future breeding programs. The results confirmed the selected SSR markers can serve as reliable tools for the molecular evaluation of lodging resistance in durum wheat genotypes.

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