



Genetic basis and multi-environment GGE-biplot evaluation of common bunt resistance in Turkish bread wheat landraces

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Abstract

Türkiye, one of the primary centers of wheat domestication, harbors a rich landrace diversity adapted to diverse agro-ecological zones. Common bunt (*Tilletia foetida*) is a seed-borne disease that causes substantial yield and quality losses. In this study, the reactions of 80 local bread wheat genotypes collected from five Turkish provinces to common bunt were evaluated over two cropping seasons (2023–2024 and 2024–2025) under artificial epidemic conditions in Kırşehir and Ankara. Genotype main effects and genotype × environment interactions were analyzed using the GGE-biplot approach. Molecular screening with bunt-resistance-associated markers was performed to detect the presence of the *Bt8*, *Bt9*, *Bt10*, and *Bt11* resistance genes. Phenotypic assessments showed that 56% of the genotypes were resistant. The *Bt10* gene was identified in 13 genotypes, *Bt8* in 9 genotypes, *Bt11* in 2 genotypes, and a combined *Bt8+Bt10* gene set in 2 genotypes, whereas *Bt9* was not detected in any genotype. The GGE-biplot explained 98.6% of the total variation and highlighted genotypes 67, 68, and 72 as superior under Ankara conditions and genotype 73 under Kırşehir conditions. Phenotypic resistance consistent with the presence of *Bt8*, *Bt10*, and *Bt11* provides a valuable genetic resource for breeding bunt-resistant wheat lines. Marker-assisted selection, particularly pyramiding *Bt8* and *Bt10*, is recommended to accelerate the development of durable resistance.

Keywords Bread wheat · Landraces · Common bunt · Resistance genes · GGE-biplot analysis

Introduction

Türkiye, as a primary center of wheat (*Triticum* sp.) origin, possesses richness of local varieties and high genetic diversity that provide strategic contributions to modern breeding programs. In a comprehensive survey conducted between 2009 and 2014, spike samples collected from 1,600 farmers' fields across 59 provinces revealed the presence of 95 morphotypes belonging to three species and six subspecies (Kan et al. 2015; Morgounov et al. 2016). Local wheat varieties are considered indispensable genetic resources in both national and international breeding efforts because of their superior traits such as cultivation under minimal inputs, tolerance to regional stress conditions, and yield stability (Akçura and Topal 2006; Jaradat 2013; Salantur et al. 2017). Germplasm originating from Türkiye has been used as parental material in the development of new cultivars in countries such as Russia and the United States, contributing in various ways to wheat production and the food industry (Damania et al. 1996; Qualset et al. 1996; Salantur et al. 2017).

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Common bunt (*Tilletia* spp.) is one of the major fungal stresses threatening global wheat production and quality. In particular, *Tilletia foetida* (Wallr.) Liro [= *Tilletia laevis* Kühn], the causal agent of common bunt, can cause yield losses of 15–20% in untreated fields and up to 96% in highly susceptible cultivars (Aktaş 2001; Dodov and Todorova 1974; İren et al. 1982). The pathogen's brownish-black teliospores produce a characteristic fishy odor due to their trimethylamine content, and infected grains negatively affect flour quality. The primary source of inoculum at national and global scales is the spread of contaminated kernels crushed during harvest. When infected seed is sown without chemical treatment, systemic infection develops under favorable environmental conditions and reaches the developing inflorescence (Wilcoxson 1996). Infected plants may remain morphologically indistinguishable from healthy plants until the milk-dough stage.

Although chemical control is widely preferred for the management of common bunt because of its relatively low cost, ease of application, and high efficacy, improper use can lead to phytotoxicity, increased production costs, negative impacts on human and environmental health. Common bunt poses a serious challenge for organic farming. Consequently, the deployment of resistant cultivars has emerged as a key strategy for sustainable food production. The selection of resistant parents in breeding programs is critical for developing new cultivars with durable and effective resistance. Local wheat landraces represent an important source of genetic resistance to common bunt.

Resistance to both common bunt and dwarf bunt in wheat is controlled by a series of *Bt* resistance genes (*Bt1*–*Bt15* and *Btp*), which may be present either singly or in combination within individual genotypes. Wheat lines carrying specific *Bt* genes have been developed and are routinely employed as reference materials for the identification and characterization of pathogen races (Hoffman and Metzger 1976; Goates 2012).

Multi-site trials provide critical information not only on the mean performance of genotypes but also on rank shifts attributable to genotype $\times$ environment ($G \times E$) interactions. Because such interactions are inherently complex, their effective visualization and interpretation require robust analytical tools. The GGE biplot methodology, which

simultaneously incorporates genotype main effects and the $G \times E$ component, offers a powerful framework to address the fundamental question of which genotype performs best in which environment.

In the present study, 80 single-spike derived local bread wheat genotypes collected from diverse regions of Türkiye were evaluated for their reactions to common bunt across multiple environments. Year-site combinations were treated as distinct environments, and the GGE biplot approach was employed to jointly assess genotype main effects and $G \times E$ interactions. The boundaries of resistance reaction groups relevant to breeding were examined, and the proportion of variation explained by the GGE biplot was quantified. Furthermore, the presence of the *Bt8*, *Bt9*, *Bt10*, and *Bt11* resistance genes was investigated using molecular markers (Table 1). The concordance between molecular marker profiles and phenotypic resistance group was evaluated to identify genotypes that exhibit environment-specific resistance, as well as stable lines with consistent performance across environments, which could be prioritized for incorporation into resistance breeding programs.

Materials and methods

Plant materials and inoculum source

In this study, 80 bread wheat landraces derived from single spikes collected from the five provinces of Türkiye including Denizli (3), Eskişehir (6), Kahramanmaraş (8), Konya (42), and Kütahya (21) were analyzed (Table S1). Additionally, the PI178383 genotype containing the *Bt8*, *Bt9*, and *Bt10* resistance genes was used as a resistant control (McIntosh et al. 2013), while the Little Club and Yakar-99 genotypes were used as susceptible controls (Akçura and Akan 2018). Both the susceptible check variety and the inoculum source of *Tilletia foetida* (syn. *T. laevis*) used in the field experiments were obtained from the Department of Plant Protection, Kırşehir Ahi Evran University. The virulence profile of this inoculum (Vr: *Bt0*, *Bt2*, *Bt3*, *Bt4*, *Bt6*, *Bt7*) was previously characterized by Akçura and Akan (2018) using a differential set. This set comprised the following varieties: Red Bobs (*Bt0*), Sel2092 (*Bt1*), Sel1102 (*Bt2*),

Table 1 The molecular markers used in the current study for detection of *Bt* resistance genes

Gene	Marker type	Marker	Sequence (5'→3')	Annealing Temp (°C)	Expected fragment (bp)	References
<i>Bt8</i> <i>Bt10</i> <i>Bt11</i>	SSR	Xgwm 114	ACAAACAGAAAATCAAAACCCG ATCCATCGCCATTGGAGTG	58 °C	180/ 160 /120	(Cichy and Goates 2009)
<i>Bt9</i>	SSR	Xgpw 7433	GTACATGGAAAGAGACCAACACCA CGCTGAGCAAGGACGATAG	60 °C	296	(Steffan et al. 2017)
<i>Bt10</i>	SCAR	FSD/RSA	GTTTATCTTTTATTTC CTCCTCCCCCA	44 °C	275	(Laroche et al. 2000)

Ridit (*Bt3*), Turkey 1558 (*Bt4*), Hohenheimer (*Bt5*), Rio (*Bt6*), Sel50077 (*Bt7*), M78-9496 (*Bt8*), M82-2098 (*Bt9*), M82-2102 (*Bt10*), PI178383 (*Bt8,9,10*), M82-2123 (*Bt11*), PI119333 (*Bt12*), PI181463 (*Bt13*), Doubbi (*Bt14*), and Carleton (*Bt15*).

Site description and experimental procedure

The bread wheat landraces were evaluated at two Turkish sites: one in the Ankara Polatlı (39°03'04.5" N, 32°08'06.4" E) and other in the Kırşehir Center (39°08'47.8" N, 34°06'49.0" E). The experiments were carried out at both sites during the growing seasons 2023–2024 and 2024–2025. Briefly, in the experiment, 2–3 g of seed from each genotype and each differential line were placed in separate paper envelopes and inoculated immediately prior to planting at a rate of approximately 0.05% (teliospore: seed, w/w) (Akan et al. 2005). In both years, seeds were sown in October and the experimental plots consisted of 1 m rows (80–100 seeds per row) with 30–35 cm row spacing and a seeding depth of 5–7 cm, arranged in three replications. To observe the disease pressure, one row of the susceptible control cultivar Little Club (LC), inoculated with the same teliospore mixture, was inserted after every 10 local bread-wheat genotypes. The differential set was planted under the same conditions. In addition, around the perimeter of each trial, four rows of the susceptible controls Yakar-99 and LC, and two rows of the resistant control PI178383, were sown. No organic or chemical fertilizers or irrigation were applied, and weed control was performed mechanically.

Disease evaluation

The number of healthy and infected spikes was recorded for each tested genotype at the maturity stage in July 2024 and 2025, in both the experimental material and the differential set. Disease incidence (%) was calculated using the following formula (Akçura and Akan 2018; Aydogdu and Kaya 2020; Dumlalısova and Bartos 2010):

$$\text{Infection rate (\%)} = \frac{(\text{Number of infected spikes} \times 100)}{\text{Total number of spikes}} \quad (1)$$

For evaluation of the results, genotypes were classified according to the highest infection percentage recorded at each environment (year × site combination). To establish clear selection thresholds, reaction classes were defined as follows: 0% (immune), 0.1–10.0% (resistant), 10.1–25.0% (moderately resistant), 25.1–40.0% (moderately susceptible), 40.1–70.0% (susceptible), and >70.1% (highly susceptible) (Akçura and Akan 2018).

For the differential set, mean infection percentages were calculated for each genotype, and reaction was classified following Hoffman and Metzger (1976) as “avirulence” (0–10%, or “virulence” (10.1–100%). Variation in disease severity of the experimental material across environments was expressed using mean common bunt scores and genotype rankings across all test environments. In the GGE-biplot analysis was applied to identify environment-specific genotypes, and mean-vs-stability plots were used to jointly evaluate average performance and stability.

Extraction of genomic DNA

Genomic DNA was extracted from 100 mg of bread wheat genotypes and control varieties at the three-leaf stage (Zadoks 13) following the CTAB protocol (Doyle and Doyle 1987). The extracted DNA was dissolved in 100 µL of Tris-EDTA (TE) buffer, and its quality and concentration were assessed by 1% agarose gel electrophoresis using a DNA standard. The DNA was then diluted with TE buffer (pH 8.0) to a final concentration of 50 ng/µL for PCR amplification.

Molecular detection of *Bt* genes

Molecular analysis was performed using PCR amplification with three different molecular markers linked to *Bt8*, *Bt9*, *Bt10*, and *Bt11* to determine the presence or absence of these resistance genes in the bread wheat landraces. Details of these markers are provided in Table 1. The total PCR reaction volume was 15 µL, containing 100 ng of DNA template, 1× PCR buffer (Thermo Fisher Scientific, Waltham, MA, USA), 1.5 mM MgCl₂ (Thermo Fisher Scientific, Waltham, MA, USA), 0.2 mM dNTPs (Thermo Fisher Scientific, Waltham, MA, USA), 1 µM forward primer, 1 µM reverse primer, and 1 U Taq DNA polymerase (Thermo Fisher Scientific, Waltham, MA, USA). Amplifications were carried out in a thermal cycler (T100, Bio-Rad, Hercules, CA, USA) under the following conditions: initial denaturation at 94 °C for 5 min; 35 cycles of denaturation at 94 °C for 1 min, annealing at 44–60 °C (Table 1) for 1 min, and extension at 72 °C for 2 min; followed by a final extension at 72 °C for 7 min. Amplified products were electrophoresed on a 2% (w/v) agarose gel in 1× TBE (Tris-borate-EDTA) buffer (pH 8.3) using a horizontal electrophoresis system (Bio-Rad, Hercules, CA, USA) at 5 V/cm for 40–60 min. A 1 kb DNA ladder (Thermo Fisher Scientific, Waltham, MA, USA) was used to determine the sizes of the amplicons. Gel visualization was performed under UV light using a gel imaging system (UVsolo touch, Analytik Jena, Jena, Germany) after staining with ethidium bromide.

Table 2 Reactions of all bread wheat landraces used in the current study to common bunt disease in the Ankara and Kırşehir sites

Site	Resistant groups		Susceptible groups			Total
	Resistant	Moderately resistant	Moderately susceptible	Susceptible	Highly susceptible	
Ankara	58 (73%)	14 (18%)	3 (4%)	3 (4%)	2 (3%)	80
Kırşehir	25 (31%)	20 (25%)	1 (1%)	3 (4%)	31 (39%)	80

Table 3 Reactions of bread wheat landraces collected from five different provinces of Türkiye to common bunt disease in Ankara site

Province	Resistance reactions		Susceptible reactions			Total
	Resistant	Moderately resistant	Moderately susceptible	Susceptible	Highly susceptible	
Denizli	2 (2.5%)	1 (1.25%)	0	0	0	3 (3.75%)
Eskişehir	4 (5.0%)	2 (2.5%)	0	0	0	6 (7.5%)
Kahramanmaraş	3 (3.75%)	4 (5.0%)	0	1 (1.25%)	0	8 (10%)
Konya	33 (41.25%)	4 (5.0%)	2 (2.5%)	2 (2.5%)	1 (1.25%)	42 (52.5%)
Kütahya	16 (20.0%)	3 (3.75%)	1 (1.25%)	0	1 (1.25%)	21 (26.25%)
Total	58 (72.5%)	14 (17.5%)	3 (3.75%)	3 (3.75%)	2 (2.5%)	80 (100%)

Data analysis

Analysis of variance (ANOVA) was applied to determine whether interactions between genotypes and environments were statistically significant. Prior to ANOVA, the reaction scores were arcsine-transformed to stabilize the variance. To further assess the response of bread wheat landraces to common bunt, we used genotype plus genotype-by-environment (GGE) biplot analysis, a widely recognized multivariate technique for studying genotype-environment interactions (Yan et al. 2007). This method condenses multi-environment trial data into a single matrix through singular value decomposition (SVD). SVD generates a set of eigenvalues, of which the two largest - those explaining the greatest proportion of the total variation - are chosen and displayed in a two-dimensional biplot (Yan and Kang 2002). The resulting plot simultaneously displays genotypes and test environments, enabling direct comparisons among environments and facilitating the identification of genotypes that either perform consistently well across all conditions or are best suited to specific sites. (Kaplan et al. 2017).

In this study, we examined the common bunt responses of 80 wheat varieties grown at two sites, Ankara and Kırşehir. The GGE biplot was based on the following statistical model:

$$Y_{ij} - \mu - \beta_j = \lambda_1 \xi_{i1} \eta_{1j} + \lambda_2 \xi_{i2} \eta_{2j} + \varepsilon_{ij}. \quad (2)$$

where Y_{ij} = represents the expected common bunt value of genotype i in environment j , μ is the overall mean across all genotype environment combinations, β_j is the main effect of environment j , λ_1 and λ_2 are the singular values of the first and second largest principal components (PC1 and PC2), ξ_{i1} and ξ_{i2} are the genotype eigenvectors, and η_{1j} and η_{2j} are the environment eigenvectors for PC1 and PC2, respectively, and lastly, ε_{ij} is the residual not captured by the first two principal components.

Results

Evaluation of bread wheat landraces for resistance to common bunt

During the study, reactions to common bunt, one of the major fungal diseases of wheat, were evaluated at two sites: Kırşehir (Central district) and Ankara (Polatlı) (Table 2). Across all sites, the susceptible control genotypes LC and Yakar-99 exhibited disease reaction rates ranging from 90% to 100%. This finding confirms the successful implementation of the inoculation procedure and demonstrates the reliability of the disease reaction assessments.

Over two growing seasons and across the two sites, statistically significant differences in common bunt reactions were detected among the 80 local lines tested. Disease incidence ranged from 0.0% to 72.0% in the Ankara site and from 2.0% to 93.0% in the Kırşehir site.

Analysis of the data revealed that none of the tested local bread wheat genotypes fell into the immune (0%) category. However, in Kırşehir, 25 genotypes (31.25%) were classified as resistant, 20 (25.0%) as moderately resistant, 1 (1.25%) as moderately susceptible, 3 (3.75%) as susceptible, and 31 (38.75%) as highly susceptible. Overall, 45 of the tested local bread wheat genotypes (56.25%) were grouped as resistant, while 35 genotypes (43.75%) were categorized as susceptible (Table 2).

For both sites, the highest disease score recorded among replications was used as the disease reaction value for that site. Disease reactions were classified as immune (0.0%), resistant (0.1–10.0%), moderately resistant (10.1–25.0%), moderately susceptible (25.1–45.0%), susceptible (45.1–70.0%), and highly susceptible (> 70.1%) following Akçura and Akan (2018).

In the Ankara site, 72 of the tested genotypes (90%) were placed in the resistant category. Conversely, eight (10%) in Ankara (Table 3) and 35 (44%) in Kırşehir were classified

as susceptible. Detailed results for the Ankara site showed that among the materials collected from different provinces: three from Denizli (3.75%) were resistant and none were susceptible; six from Eskişehir (7.55%) were resistant and none were susceptible; seven from Kahramanmaraş (8.75%) were resistant and one (1.25%) was susceptible; 37 from Konya (46.25%) were resistant and five (6.25%) were susceptible; and 19 from Kütahya (23.75%) were resistant while two (2.5%) were susceptible (Table 3). Overall, among the 80 entries tested in Ankara, 72 (90%) were classified as resistant and 8 (10%) as susceptible.

In the Kırşehir site, among the tested materials, two from Denizli (2.5%) were resistant and one (1.25%) was susceptible; four from Eskişehir (5%) were resistant and two (2.5%) were susceptible; two from Kahramanmaraş (2.5%) were resistant and 6 (7.5%) were susceptible; 27 from Konya (33.75%) were resistant and 15 (18.75%) were susceptible; and 10 from Kütahya (12.5%) were resistant while 11 (13.75%) were susceptible (Table 4). Overall, of the 80 materials tested in Kırşehir, 45 (56%) were classified as resistant and 35 (44%) as susceptible.

Evaluation of the GGE-biplot analysis

The first and second principal components accounted for 79.71% and 18.88% of the total variation, respectively, explaining 98.59% when combined. This high cumulative proportion indicates that the variation in bunt reactions of the genotypes across different environments is statistically well represented on a two-dimensional plane (Fig. 1).

The polygon view of the GGE biplot clearly illustrates how the environments discriminate among genotypes. The Kırşehir environments from 2024 to 2025 are positioned close to each other on the plot, indicating a positive and significant relationship between these two testing years. Similarly, the 2024 and 2025 disease data from the Ankara site show a positive and significant association. In contrast, the Ankara and Kırşehir environments are clearly separated. The long distances of the environment vectors from the origin indicate that both sites possess strong discriminatory ability.

The polygon view of the GGE biplot also highlights environment-specific superiority. Genotypes 67, 68, and

72, which align at narrower angles with the Ankara vectors, are identified as having low disease reactions specific to the Ankara environment. In the lower-left sector, genotype 73, located nearest to the Kırşehir 2024 and 2025 vectors, stands out for the Kırşehir environment. Genotypes such as 26, 20, 24, and 17, positioned close to the origin, contribute minimally to the total variation and, although considered stable, are not recommended for any specific environment based on disease reaction.

Molecular detection of common bunt resistance genes

In this study, molecular markers linked to the *Bt8*, *Bt9*, *Bt10*, and *Bt11* resistance genes were employed to determine whether the tested local bread wheat varieties carried these genes. PCR analyses using the Xgwm114 primer, which is associated with the *Bt8*, *Bt10*, and *Bt11* resistance genes, produced DNA fragments of 180, 160, and 120 bp, respectively (Figure S1). The *Bt9* resistance gene was analyzed with the Xgwp7433 molecular marker; however, it was not detected in any of the tested materials (Figure S2). For the identification of the *Bt10* resistance gene, primers specific to the FSD/RSA SCAR marker were used (Laroche et al. 2000). PCR analysis with this marker generated bands of 275 bp (Figure S3).

According to the molecular analyses, the *Bt8* resistance gene was detected in nine bread wheat landraces, the *Bt10* in 13 of them, and *Bt11* in two genotypes. Given that both Xgwm114 and FSD/RSA markers have been reported to be associated with the *Bt10* gene, a comparative analysis of the results obtained from these two markers was conducted. The analysis revealed that only one genotype was positive for both markers, indicating concordant results. However, discrepancies were observed among the remaining genotypes, where the presence of the diagnostic fragment detected by one marker was not confirmed by the other. In contrast, none of the tested varieties carried the *Bt9* resistance gene. Furthermore, a gene combination, namely *Bt8*+*Bt10* was detected in two genotypes (Table 5).

Table 4 Reactions of bread wheat landraces collected from five different provinces of Türkiye to common bunt disease in Kırşehir site

Province	Resistance reactions		Susceptible reactions			Total
	Resistant	Moderately resistant	Moderately susceptible	Susceptible	Highly susceptible	
Denizli	0	2 (2.5%)	0	0	1 (1.25%)	3 (3.75%)
Eskişehir	2 (2.5%)	2 (2.5%)	0	0	2 (2.5%)	6 (7.5%)
Kahramanmaraş	1 (1.25%)	1 (1.25%)	0	0	6 (7.5%)	8 (10%)
Konya	15 (18.75%)	12 (15.0%)	1 (1.25%)	1 (1.25%)	13 (16.25%)	42 (52.5%)
Kütahya	7 (8.75%)	3 (3.75%)	0	2 (2.5%)	9 (11.25%)	21 (26.25%)
Total	25 (31.25%)	20 (25.0%)	1 (1.25%)	3 (3.75%)	31 (38.75%)	80 (100%)

Table 5 Molecular detection results of the bread wheat landraces carrying *Bt* resistance gene(s)

No.	Province	Local name	Resistance gene			
			Bt8	Bt9	Bt10	Bt11
1	Denizli	Polatlı buğday1	–	–	–	+
2	Denizli	Polatlı buğday2	–	–	–	+
3	Denizli	Polatlı buğday3	–	–	–	–
4	Eskişehir	Ak buğday1	+	–	–	–
5	Eskişehir	Ak buğday2	–	–	+	–
6	Eskişehir	Külümbür1	–	–	–	–
7	Eskişehir	Külümbür2	–	–	+	–
8	Eskişehir	Zincirli-Külümbür1	–	–	+	–
9	Eskişehir	Zincirli-Külümbür2	–	–	+	–
10	Kahramanmaraş	Beyaz buğday1	–	–	+	–
11	Kahramanmaraş	Beyaz buğday2	–	–	+	–
12	Kahramanmaraş	Elbistan yazlığı1	–	–	+	–
13	Kahramanmaraş	Elbistan yazlığı2	–	–	+	–
14	Kahramanmaraş	Polatlı yazlığı1	–	–	+	–
15	Kahramanmaraş	Polatlı yazlığı2	+	–	–	–
16	Kahramanmaraş	Yerli buğday1	+	–	–	–
17	Konya	Yerli buğday2	–	–	–	–
18	Konya	Akbaş1	–	–	–	–
19	Konya	Akbaş2	–	–	–	–
20	Konya	Akbaş3	–	–	+	–
21	Konya	Akbuğday1	–	–	–	–
22	Konya	Akbuğday2	–	–	+	–
23	Konya	Akbuğday3	–	–	–	–
24	Konya	Alabuğday1	–	–	–	–
25	Konya	Alabuğday2	–	–	–	–
26	Konya	Alabuğday3	–	–	–	–
27	Konya	Beyaz Kelle1	–	–	–	–
28	Konya	Beyaz Kelle2	–	–	–	–
29	Konya	Beyaz Kelle3	–	–	–	–
30	Konya	Dede buğday1	+	–	+	–
31	Konya	Dede buğday2	–	–	–	–
32	Konya	Dede buğday3	–	–	–	–
33	Konya	Göremez1	–	–	–	–
34	Konya	Göremez2	+	–	–	–
35	Konya	Göremez3	+	–	–	–
36	Konya	Kamçı1	–	–	–	–
37	Konya	Kamçı2	+	–	–	–
38	Konya	Kamçı3	+	–	–	–
39	Konya	Karabuğday1	–	–	–	–
40	Konya	Karabuğday2	–	–	–	–
41	Konya	Karabuğday3	–	–	–	–
42	Konya	Beyaz Buğday1	–	–	–	–
43	Konya	Beyaz Buğday2	–	–	–	–
44	Konya	Beyaz Buğday3	–	–	–	–
45	Konya	Kırmızı buğday1	–	–	–	–
46	Konya	Kırmızı buğday2	–	–	–	–
47	Konya	Kırmızı buğday3	–	–	–	–
48	Konya	Kızılca1	–	–	–	–
49	Konya	Kızılca2	–	–	–	–
50	Konya	Kızılca3	–	–	–	–
51	Konya	Morbuğday1	–	–	–	–
52	Konya	Morbuğday2	–	–	–	–
53	Konya	Morbuğday3	–	–	–	–
54	Konya	Sarı buğday1	+	–	+	–

Table 5 (continued)

No.	Province	Local name	Resistance gene			
			Bt8	Bt9	Bt10	Bt11
55	Konya	Sarı buğday2	–	–	–	–
56	Konya	Sarı buğday3	–	–	–	–
57	Konya	Sert buğday1	–	–	–	–
58	Konya	Sert buğday2	–	–	–	–
59	Konya	Sert buğday3	–	–	–	–
60	Kütahya	Ak buğday1	–	–	–	–
61	Kütahya	Ak buğday2	–	–	–	–
62	Kütahya	Ak buğday3	–	–	–	–
63	Kütahya	Akçalıbasan1	–	–	–	–
64	Kütahya	Akçalıbasan2	–	–	–	–
65	Kütahya	Akçalıbasan3	–	–	–	–
66	Kütahya	Delihüseyin buğday1	–	–	–	–
67	Kütahya	Delihüseyin buğday2	–	–	–	–
68	Kütahya	Delihüseyin buğday3	–	–	–	–
69	Kütahya	Gülümbür1	–	–	–	–
70	Kütahya	Gülümbür2	–	–	–	–
71	Kütahya	Gülümbür3	–	–	–	–
72	Kütahya	Akkobak buğday1	–	–	–	–
73	Kütahya	Akkobak buğday2	–	–	–	–
74	Kütahya	Akkobak buğday3	–	–	–	–
75	Kütahya	Kobak buğday1	–	–	–	–
76	Kütahya	Kobak buğday2	–	–	–	–
77	Kütahya	Kobak buğday3	–	–	–	–
78	Kütahya	Sümter1	–	–	–	–
79	Kütahya	Sümter2	–	–	–	–
80	Kütahya	Sümter3	–	–	–	–

*Presence (+) or absence (–) of the *Bt* genes according to the obtained molecular data

is known to carry not only *Bt8*, *Bt9*, and *Bt10* but also an unidentified resistance gene (Hoffman and Metzger 1976).

According to the differential set results, the inoculum used in this study was virulent to *Bt0*, *Bt2*, *Bt3*, *Bt4*, *Bt6*, and *Bt7*, but avirulent to *Bt1*, *Bt5*, *Bt8*, *Bt9*, *Bt10*, *Bt11*, *Bt12*, *Bt13*, *Bt14*, and *Bt15*. These findings closely align with earlier study (Akçura and Akan 2018). International research has likewise shown that the effectiveness and distribution of *Bt* resistance genes vary geographically: *Bt8*, *Bt9*, *Bt10*, and *Bt11* were reported to be effective in Kazakhstan, whereas different *Bt* genes were found effective in Iraq and Syria (Al-Maaroof and Mahmood-Aziz 2016; Madenova et al. 2021). In the United States, *Bt6*, *Bt9*, *Bt11*, *Bt12*, *Bt13*, *Bt15*, and *Btp* have been reported as effective (Mourad et al. 2018). Conversely, Goates (2012) noted that some new races can overcome *Bt8*, *Bt9*, *Bt10*, *Bt11*, and *Bt12*, and that *Bt14* and *Bt15* may be influenced by environmental conditions.

The GGE-biplot analysis highlighted two major points. First, the primary source of environmental variation was attributed to site, whereas the year factor had a relatively limited effect. Second, the presence of genotypes that performed best in environments matching their origin suggests that the panel includes candidates with broad adaptation

potential. These genotypes are likely to exhibit stable and durable performance if repeatedly tested across diverse environments. Moreover, the length and angular relationships of the environment vectors reflected epidemiological and climatic differences in the field; therefore, the selection of reference environments should be based not only on discriminating ability but also on representativeness.

The use of molecular markers has emerged as an important tool in breeding programs aimed at developing disease-resistant cultivars (Ciucă 2011). The increasing adoption of marker-assisted selection in recent years has accelerated the mapping of common bunt resistance and its integration into breeding. However, the lack of specific markers capable of reliably detecting all *Bt* resistance genes and ongoing debates about the specificity of some markers remain challenges (Poyraz and Gümüç 2016; Wang et al. 2009). The practical value of marker-assisted selection has been emphasized in studies from Canada, Kazakhstan, and Türkiye (Madenova et al. 2019; Randhawa et al. 2013; Tekin 2023). Furthermore, although *Bt9* and *Bt10* are located on the same chromosome, mapping and functional analyses have shown that they represent distinct loci (Hiebert et al. 2011; Steffan et al. 2017).

In conclusion, when evaluated together with similar studies, this research demonstrates that local Turkish bread wheat germplasm harbors valuable sources of resistance that can serve as parents in breeding for common bunt resistance. Molecular screening revealed the presence of *Bt8*, *Bt9*, *Bt10*, and *Bt11* in local material, suggesting that appropriate combinations of these genes (gene pyramiding) could effectively control the disease and reduce infection levels.

Conclusion

In this study, 56% of the evaluated Turkish bread wheat landraces were grouped as the resistant demonstrating their potential use as donor parents in breeding programs. The differential set analysis showed that the inoculum used was virulent to the *Bt0*, *Bt2*, *Bt3*, *Bt4*, *Bt6*, and *Bt7* resistance genes, whereas molecular screening revealed the presence of *Bt8*, *Bt10*, and *Bt11* in the local material however, *Bt9* was not found by markers in the material. Notably, the presence of both *Bt8* and *Bt10* in genotypes 30 and 54 suggests that these genotypes may be considered as potential candidates for future gene-pyramiding.

GGE-biplot analysis accurately captured the spatial and temporal patterns of common bunt resistance, with PC1 and PC2 together explaining 98.59% of the total variance, indicating that a two-dimensional biplot provides sufficient decision support. Genotypes 67, 68, and 72 were identified as superior and relatively stable for the Ankara environment, whereas genotype 73 was outstanding for the Kırşehir environment. Genotypes 26, 20, 24, and 17, originating from environments close to their collection sites, exhibited broad adaptation potential and were incorporated into the core candidate pool.

These findings underscore the strategic importance of complementing classical selection with molecular markers in the development of bunt-resistant cultivars. They further emphasize the need to prioritize region-specific cultivar recommendations, conserve local genetic diversity, and continue multi-year, multi-site characterization efforts.

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Declarations

Competing interests There is no conflict of interest between authors.

Ethical approval The article does not require any ethical committee decision.

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