



Multi-environment trial (MET) evaluation of bread wheat (*Triticum aestivum* L.) genotypes across the highlands of Ethiopia using factor analytic mixed models

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ABSTRACT

This study evaluated 37 multi-environment trials (METs) comprising 787 bread wheat genotypes, using Alpha lattice and partially replicated designs across the highland agro-ecological zones of Ethiopia from 2021 to 2024. The study identified significant phenotypic variation, with grain yields ranging from 1.54 to 7.39 t ha⁻¹ across environmental trials. The genotypes EBW170049 and EBW170074 appeared as superior performers, yielding 5.31 t ha⁻¹ and 5.17 t ha⁻¹, respectively, while 65% of the genotypes exceeded the 4.5 t ha⁻¹ productivity threshold. The superior performance of EBW170049 was complemented by strong grain quality parameters, with HLW of 70.37 kg hl⁻¹ and TKW of 38.46 g, suggesting a stable and efficient grain-filling period. A Factor Analytic Mixed Model (FAMM) captured over 99.9% of the cumulative genetic variance, facilitating a precise decomposition of environmental interactions. Broad-sense heritability reached 99.16% for days to heading and 98.16% for grain yield in high-potential sites, although environmental sensitivity reduced plant height heritability to 52.65% in strained locations. Trait-based environmental clustering helped delineate principal mega-environments and identified specific trial outliers. The finding revealed that early-to-medium maturity demonstrated by superior genotypes serves as a critical terminal stress-escape strategy. Furthermore, top-performing genotypes generally exhibited medium plant height as lodging resistance potential. Regarding biotic stress, genotypes EBW170051 and EBW170058 showed superior yellow rust resistance, while EBW222276 remained resistant to stem rust. Overall, the study confirmed high genetic gain potential and demonstrated that FAMM-based MET analysis is a robust framework for identifying superior genotypes and enhancing productivity and climate resilience in bread wheat breeding programs.

Keywords: Factor analytic model, Genotype-by-Environment interaction, Genetic gain, Rust resistance

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Introduction

Bread wheat (*Triticum aestivum* L.) is a hexaploid cereal (2n = 6x = 42; AABBDD) and the world's most widely grown crop. It accounts for approximately 95% of global wheat production and provides about 20% of the total calories and protein consumed

by humans (Anonymous, 2023; Shewry and Hey, 2015). Although there have been significant advancements in wheat productivity, further efforts are required to address the challenge posed by the continued growth of the global population,

which is projected to reach 9.8 billion by 2050 (Hanafi, 2020). In Ethiopia, wheat is a top-priority food and a strategic crop for national food security and rural livelihoods. Bread wheat now occupies over 90% of the wheat-growing area in Ethiopia, surpassing traditional durum wheat due to improved varieties with higher yield potential and disease resistance (Hodson *et al.*, 2020; Jaleta *et al.*, 2019). It is a significant source of protein, dietary fiber, B vitamins, and minerals (Levy *et al.*, 2020). Beyond its nutritional value, bread wheat is essential for producing various traditional foods, including bread, porridge (genfo), local beer (tela), roasted grain (kolo), boiled grain (nifro), and injera (Geneti *et al.*, 2022; Mullualem *et al.*, 2024). Furthermore, wheat straw is frequently utilized as roofing material and livestock feed, underscoring its socio-economic importance in mixed farming systems (Anteneh *et al.*, 2020). It is cultivated on approximately 2.1 million hectares annually, with 1.7 million hectares under rain-fed conditions and 0.4 million hectares under irrigation (Tadesse *et al.*, 2022). Recently, Ethiopia achieved 100% wheat self-sufficiency with a surplus for export, indicating that the new irrigated wheat initiative has been transformational (Effa *et al.*, 2023). The Ethiopian government continues to enhance production through land expansion, agro-clustering of farmers, and the expansion of irrigation into lowlands (Senbeta and Worku, 2023; Tadesse *et al.*, 2022).

Even though an average wheat yield reached 3.12 t ha⁻¹ in 2024, farmers still face a major gap between actual and potential yield in Ethiopia. This gap is attributed to biotic and abiotic stresses, suboptimal agronomic practices, and limited access to technology. Among biotic constraints, rust diseases—particularly stem rust and stripe rust—pose serious threats (Hodson *et al.*, 2020). Ethiopia is among the most important wheat-producing countries and is highly threatened by cereal rusts, particularly yellow and stem rusts. There exist frequent epidemics of these diseases as a result of the evolution of new races. Regardless of the enormous efforts to develop high-yielding, disease-resistant varieties, the agronomic life span of most varieties is very short. Current commercial bread wheat varieties cultivated in the highlands are susceptible to stem rust and have a long maturity period (Senbeta and Worku, 2023). Therefore,

developing a new rust-resistant, high-yielding, and early-maturing variety is a paramount solution for resource-poor farmers growing wheat in Ethiopia's highlands. To address these challenges, breeding programs focus on developing high-yielding and rust-resistant varieties. However, genotype-by-environment (G×E) interaction complicates the identification of superior genotypes (Zhao *et al.*, 2025). Therefore, Multi-environment trials (METs) are essential for evaluating genotypes across diverse agro-ecologies and identifying adapted varieties (Waters *et al.*, 2023). Linear mixed models, particularly factor analytic mixed models (FAMM), provide a robust framework for MET analysis by capturing G×E covariance structures (Argaw and Demelash, 2025; Smith *et al.*, 2001).

Furthermore, factor analytic mixed models (FAMM), which incorporate spatial analysis, account for field heterogeneity and enhance selection precision (Alemayehu *et al.*, 2025; Argaw *et al.*, 2024). Importantly, combining FA models with spatial models for non-genetic effects greatly improves the analysis of MET datasets, as demonstrated in related studies by Cullis *et al.* (2010) and Kelly *et al.* (2007). FA models are useful not only for accurately estimating and predicting genetic effects but also for estimating their variance and conducting graphical analyses (Smith *et al.*, 2001). By utilizing estimated genetic variance, breeders can identify correlated environments and select genotypes based on best linear unbiased predictions (BLUPs) averaged across these environments (Argaw *et al.*, 2024). Understanding the variance structure related to genotype-by-environment effects is a crucial component of the FA model for METs. The present research was conducted to evaluate the performance of bread wheat genotypes that may be suitable for the diverse agro-ecological conditions of Ethiopia, employing more efficient statistical methods for data analysis. Thus, this study aims to enhance selection strategies in bread wheat breeding by analyzing MET data using a FAMM-based approach to evaluate bread wheat genotypes across the major highland wheat-growing regions of Ethiopia. Hence, the objectives of this study were to evaluate the performance, disease resistance, and stability of bread wheat genotypes across diverse highland environments in Ethiopia using more advanced statistical models.

Materials and Methods

Dataset

The MET datasets were compiled from four trial categories: Observation Nursery (ON; 2021–2022), Preliminary Variety Trials (PVT; 2021–2022), National Variety Trials Stage-I (NVT1; 2022–2023), and National Variety Trials Stage-II (NVT2; 2022–2024), while the NVT trials cover a broader range of environments (Table 1). These trials were conducted across nine representative locations within major high-rainfall bread wheat-growing regions, with Debreziet and Meraro serving specifically as disease screening sites.

A total of 787 unique genotypes, including 2 checks, were evaluated across 37 trial–location–season combinations. For the combined Observation Nursery, Preliminary Variety Trials, and NVT Stage-I (OPN) trials, a partially replicated design was used to accommodate a large number of genotypes while maintaining precision in early-stage evaluations (Piepho *et al.*, 2021). OV, PVT and NVT trials were arranged in row–column alpha lattice designs to account for spatial variability within the field (Butler and Cullis, 2022).

Table 1. Detailed agro-ecological and weather descriptions of the testing location.

Location	Code	Altitude (m)	Latitude	Longitude	Mean Temp.		Mean Rainfall (mm)
					Min.	Max.	
Kulumsa	KU	2200	08°01'10"N	39°09'11"E	10.5	22.8	820
Bekoji	BE	2780	07°32'37"N	39°15'21"E	7.9	18.6	1010
Koffele	KF	2695	07°00'00"N	38°45'00"E	8.6	19.7	1170
Enawari	EW	2650	09°53'00"N	39°09'00"E	15.0	20.0	878
Arsi Robe	RA	2420	07°53'02"N	39°37'40"E	6.0	22.1	796
Debre Markos	DM	2450	10°19'59"N	37°44'53"E	12.4	23.5	1300
Holletta	HL	2400	09°03'41"N	38°30'44"E	6.1	22.4	1100
Chefe Donsa	CD	2444	08°57'60"N	39°06'28"E	9.8	23.1	1100
Gonder	GD	2,133	12°36'00"N	37°28'00"E	12.3	26.7	937
Debre Zeit	DZ	2050	08°38'08"N	38°30'15"E	11.4	26.8	985
Meraro	MR	3,030	07°24'00"N	39°14'00"E	10.0	17.0	1200

Data collection

Data were recorded on days to heading, plant height, thousand-kernel weight, hectoliter weight, and grain yield. The description of the collected data/traits has been shown as follows:

Days to Heading (DTH): Obtained by recording days from the sowing date to when 75% of spikes have fully emerged.

Plant Height (cm): Measured by averaging the height of five random plants from the ground to the spike tip (excluding awns) after physiological maturity.

Thousand Kernel Weight (g): Determined by counting the seeds with a seed counter and weighing them on a sensitive balance at 12.5% moisture content.

Hectoliter Weight (kg hl⁻¹): Measured as the flour density (kg hl⁻¹) produced from a hectoliter of seed after harvest using a specialized grain analyzer.

Grain Yield (t ha⁻¹): Grain yield in grams was obtained from the entire plots harvested after physiological maturity, weighed, and converted to tons per hectare at 12.5% moisture content.

Trial designation

Table 2 summarizes the 37 Trials, including trial design and trait measurements. Each trial was identified by a name (e.g., 21BWOLBE) combining the last two digits of the trial year (21= 2021), two letters of the crop under study (BW = Bread Wheat), the first letters of the trial series (OL = Observation nursery for Late maturity), and the testing site/location code (BE = Bekoji). Table 2 also provides the number of replications, the dimensions of each trial (rows × columns), and the average trait values across trials. In this study, the terms “environment” and “trial/site” are used interchangeably.

Table 2. Summary of trial scheme and average trait measurements across trials.

Trial	No. of Reps	No. of Rows	No. of Columns	No. of Genotypes	Average measurement				
					GYLD	PHT	HLW	DTH	TKW
21BWOLBE	2	30	10	200	1.54	93.83	59.54	82.47	24.95
21BWOLKF	2	30	10	200	1.66	89.78	52.81	78.87	26.10
21BWPLBE	3	26	10	127	2.18	91.35	62.11	80.32	26.99
21BWPLKF	3	26	10	127	2.29	86.00	57.01	NA	29.57
22BWNLBE	3	15	10	50	3.65	92.4	68.80	81.83	38.29
22BWNLCD	3	15	10	50	5.20	86.23	72.21	71.29	50.86
22BWNLDM	3	15	10	50	4.69	94.62	NA	67.00	47.75
22BWNLEW	3	15	10	50	6.99	90.70	83.60	78.83	NA
22BWNLGD	3	15	10	50	4.92	94.03	NA	72.21	41.43
22BWNLHL	3	15	10	50	2.97	88.55	74.02	63.87	30.99
22BWNLKF	3	15	10	50	3.17	91.05	58.71	NA	NA
22BWNLKU	3	15	10	50	5.02	93.19	63.62	62.27	35.39
22BWNLRA	3	15	10	50	5.15	93.89	70.77	68.35	44.81
22BWOLBE	2	30	10	200	2.42	103.46	62.35	83.61	32.71
22BWOLKF	2	30	10	200	2.20	99.55	55.68	NA	26.26
22BWPLBE	3	28	8	112	2.83	91.30	67.78	81.79	33.66
22BWPLDM	3	28	8	112	4.26	96.90	NA	69.24	NA
22BWPLEW	3	28	8	112	7.39	93.37	81.96	81.07	49.29
22BWPLHL	3	28	8	112	2.55	88.33	74.77	64.98	29.72
22BWPLKU	3	28	8	112	4.63	97.21	61.55	63.90	27.62
22BWPLRA	3	28	8	112	4.07	90.21	69.57	71.24	43.24
23BWNLBE	3	10	9	30	4.15	85.26	72.15	83.84	43.16
23BWNLDM	3	10	9	30	5.15	85.70	NA	66.10	NA
23BWNLHL	3	10	9	30	4.62	90.14	NA	62.63	41.49
23BWNLRA	3	10	9	30	2.89	87.58	67.14	65.48	33.07
23BWOPNLKU	3	42	13	325	3.67	86.92	64.96	67.02	32.51
23BWPLBE	3	14	12	84	2.79	87.85	69.45	85.83	39.31
23BWPLDM	3	14	12	84	3.61	90.12	NA	64.23	NA
23BWPLHL	3	14	12	84	4.19	88.02	NA	62.09	39.55
23BWPLRA	3	14	12	84	2.53	94.88	66.79	65.00	33.60
24BWOPNLBE	3	35	9	169	2.84	87.49	59.70	82.30	28.84
24BWOPNLKU	3	35	9	169	4.74	102.55	66.00	72.95	27.38

GYLD= Gain yield, PHT=Plant height, DTH= Days to heading, TKW=Thousand kernel weight, HLW = Hectoliter weight, NA= Not available, O = Observation nursery, L= Late maturity groups, P = Preliminary variety trails, N= National variety trails

Statistical analysis

The Multi-Environment Trial (MET) analysis was performed for trials evaluated at high-altitude areas of Ethiopia during the 2021 to 2024 cropping seasons. Genetic variance, error variance, heritability, and clustering across trials were analyzed using advanced statistical approaches, including Factor-analytic mixed models and genotype-by-environment interaction, implemented in ASReml and the Metan R package (Olivoto *et al.*, 2023). The ASReml-R statistical software package was used to fit all models analyzed in this study (Butler *et al.*, 2009).

Results and Discussion

Mean performances of genotypes across environments

A comprehensive analysis of 37 trials conducted during the 2021 to 2024 main

cropping season revealed a significant phenotypic range across all traits (Table 2). Using a Factor Analytic Mixed Model (FAMM) to evaluate bread wheat genotypes enabled precise modeling of G×E interactions and spatial variability, facilitating the identification of stable, high-performing genotypes (Smith *et al.*, 2001; Argaw and Demelash, 2025). The mean grain yield (GYLD) across the study sites was 3.78 t ha⁻¹; this average masked a nearly five-fold discrepancy between the lowest-yielding trial, 21BWOLBE (1.54 t ha⁻¹), and the highest, 22BWPLEW (7.39 t ha⁻¹). Such fluctuations in mean yield are frequently attributed to varying moisture availability and heat stress during the critical flowering stage (Mondal *et al.*, 2020), a trend reflected in the lower performance of the 2021 trial series compared to the 2022 and 2023 cohorts. Grain yield varied significantly across genotypes, ranging from

2.39 t ha⁻¹ to 5.47 t ha⁻¹, with EBW170049 (5.31 t ha⁻¹) and EBW170074 (5.17 t ha⁻¹) as the top performers, followed closely by elite genotypes EBW222923 (5.06 t ha⁻¹) and EBW170051 (5.03 t ha⁻¹). Such a vast gap suggested that the yield potential was profoundly influenced by localized environmental factors or Genotype by Environment (G × E) interactions. The grain yield results were further elucidated by grain quality parameters, with Hectoliter Weight (HLW) averaging 66.52 kg hL⁻¹ and Thousand Kernel Weight (TKW) averaging 35.50 g. The significant range—from 24.95 g (21BWOLBE site) to 50.86 g (22BWNLCD site) for TKW—directly reflects the observed trends in total grain yield. As grain weight is a primary determinant of cereal yield potential (Slafer *et al.*, 2022), the higher performance of genotypes at the 22BWNLCD site highlights the critical role of grain density in driving overall productivity.

Structural and phenological traits also exhibited distinct environmental patterns. Significant variation was observed among genotypes, with DTH ranging from 67.78 to 80.12 days and PHT varying from 85.67 to 105.1 cm, suggesting a heterogeneous genetic background for height-reduction genes. This implied that Plant Height (PHT) remained relatively stable with a mean of 91.64 cm, while Days to Heading (DTH) showed a substantial 24-day gap between the earliest and latest trial sites. Hence, optimizing the heading date is essential for escaping terminal stress (Tadesse *et al.*, 2021), suggesting that trials reaching the 72-day mean likely benefited from a more favorable grain-filling gap than those with extreme early or late maturity. The majority of high-yielding genotypes headed between 69 and 72 days, identifying early-to-medium maturity as a key adaptive trait. The early heading recorded for genotypes such as EBW212354 (67.78 days) and EBW212116 (69.03 days) represented a "terminal stress escape" strategy, which allowed the crop to complete anthesis before the onset of frost and the rapid depletion of residual soil moisture common in the highland areas of Ethiopia. Furthermore, the dominance of semi-dwarf types (90–95 cm) among high-yielding lines, such as EBW170074, reflected an optimized Harvest Index (HI). While ultra-short plants may lack carbohydrate storage capacity, taller genotypes often prioritize vegetative biomass over reproductive sinks, explaining their lower yield performance (Xue *et al.*, 2022).

In general, the identification of a "high-yield, early-maturity" bread wheat genotypes—specifically EBW170049, EBW170074, EBW212401, EBW212116, and EBW212354—provided a robust strategy for highland conditions, where frost, low temperatures, and high rainfall limit the growing season. These genotypes reached yields above 4.5 t ha⁻¹ by demonstrating enhanced cold tolerance during flowering and optimized biomass accumulation under low-temperature conditions (Xue *et al.*, 2022). Quality parameters further supported these findings, as EBW212354 recorded the highest TKW (45.58 g). Ultimately, the superior performance of these elite genotypes confirmed that high kernel weight, hectoliter weight (HLW), and optimized plant height were the primary drivers of genetic gain. Consequently, this study demonstrated that incorporating FAMM effectively partitioned genetic and environmental variance, thereby pinpointing superior bread wheat germplasm. These results provided valuable resources for breeding programs aimed at enhancing climate resilience and yield stability in the rain-fed highlands of Ethiopia.

Variance components and heritability estimates for each trait

Estimates of genetic variance, error variance, and heritability for each trait across trials are presented in Table 3. Understanding variance components and heritability is essential for effective selection in plant breeding (Cullis *et al.*, 2010). These metrics provided insights into the potential response to selection for the key traits measured across the highland rain-fed areas of Ethiopia. Hence, the evaluation of Bread wheat genotypes across multiple highland areas revealed a substantial range of genotypic variance, a fundamental requirement for any successful breeding program. In almost all environments, the genetic variance for grain yield (GYLD) was significantly higher than the environmental variance. Grain yield is a typical polygenic trait, yet the data revealed an unexpectedly robust genetic architecture across the tested environments. This suggested the presence of stable genetic markers that could be highly valuable for developing crops that perform reliably in different climates. Hence, genetic variance for Grain Yield fluctuated significantly, peaking at 3.78 in the 22BWNLCF environment, while the corresponding error variance remained

negligible at 0.22, resulting in a high broad-sense heritability of 98.16%, indicating that in specific high-potential highland environments, the genetic signal for yield was highly detectable. Conversely, in more restrictive environments like 22BWPLEW, heritability dipped to 68.45%, with error variance (0.53) exceeding genetic variance (0.34). Recent literature emphasizes that, while yield is highly influenced by Genotype $\times$ Environment interactions, the high heritability observed in many Ethiopian wheat-growing sites indicates that substantial genetic gains could still be achieved through direct selection (Tadesse *et al.*, 2022). Furthermore, integrating genomic selection and high-throughput phenotyping can accelerate these gains by more effectively accounting for complex Genotype $\times$ Environment interactions than traditional methods alone (Merrick *et al.*, 2022).

The phenological development of the crop, represented by days to heading, showed the most consistent and highest heritability estimates across all trials, frequently exceeding 95% (e.g., 99.16% at the 23BWNLHL site). The genetic variance values for days to heading were substantially larger than their error variance counterparts in nearly every instance, indicating that time to flowering is under strict genetic control and minimally susceptible to minor environmental fluctuations common in highland agro-ecologies. In the context of contemporary climate-resilient breeding, the high heritability of days to heading enables breeders to precisely select maturity groups that align with specific rainfall patterns in the highlands (Mwadingeni *et al.*, 2021). Furthermore, plant height exhibited a wide range of genetic variance, reaching as high as 89.39 at the 22BWOLKF site. While heritability remained generally high (averaging between 80% and 95%), there were notable exceptions, such as 22BWPLDM, where heritability fell to 52.65%. In this specific case, the error variance (19.75) was nearly quadruple the genetic variance (5.07), indicating that environmental factors unique to certain highland sites—such as soil fertility gradients, high-altitude moisture availability, or plant competition—significantly influenced stem elongation. Moreover, modern wheat breeding prioritizes semi-dwarf architectures to improve harvest index and lodging resistance, which is vital

in high-input highland systems. Similarly, recent findings have shown that high heritability of morphological traits facilitates selection for optimal biomass distribution, enabling the development of varieties that can support high grain loads without lodging, even under high-input conditions (Voss-Fels *et al.*, 2019).

On the other hand, the traits associated with grain physical quality, such as thousand-kernel weight (TKW) and hectoliter weight (HLW), exhibited high genetic variance relative to environmental variance, with heritability often exceeding 90%. The high heritability for TKW was recorded at the 21BWPLBE site (94.06%), driven by a genetic variance of 30.47 against a low error variance of 4.99. Likewise, HLW followed a similar trend, showing higher heritability across most highland sites with 97.49% at 22BWNLHL site. This indicated that the density and size of bread wheat grain are primarily determined by the varieties' inherent genetic makeup rather than by site-specific soil or weather conditions. Recent global meta-analyses of wheat quality noted that TKW and HLW are critical components of both milling yield and market value (Guzmán *et al.*, 2022). Therefore, the high heritability values found in this dataset implied that these traits are less sensitive to Genotype $\times$ Environment interactions than grain yield itself. Consequently, selection for high TKW and HLW can be conducted with high confidence across diverse highland environments, providing a reliable pathway to improve the overall yield performance of bread wheat genotypes in Ethiopia.

Overall, specific highland locations, such as 24BWOPNLBE and 22BWNLHL, appeared to be "gold standard" environments for selection because they consistently provided the ideal balance of high genetic variance and high heritability. Hence, optimal selection environments demonstrated high genetic variance and low environmental error, supporting the advancement of climate-resilient cultivars. These superior environments maximize the potential for genetic gain by ensuring that selection pressure was applied to phenotypic differences that are almost entirely attributable to the underlying genotype rather than environmental noise (Cullis *et al.*, 2010). By prioritizing genotypes that excel at these high-potential highland sites, breeders can capture the highest possible gains for both grain yield and quality traits

in a single selection cycle. Generally, Multi-environment trials across the Ethiopian highlands revealed high heritability for grain yield, days to heading, and grain quality, enabling effective selection of bread wheat genotypes. Therefore, the overall trend across the 37 highland site-year

combinations confirmed that most of the genotypes under study possessed high genetic gain potential, establishing them as excellent candidates for promotion to advanced variety trials across the highlands of Ethiopia.

Table 3. Estimates of genetic variance, error variance and heritability for each trait across trials.

Site	GYD			DTH			PHT			TKW			HLW		
	Gvar	Evar	H ²	Gvar	Evar	H ²	Gvar	Evar	H ²	Gvar	Evar	H ²	Gvar	Evar	H ²
21BWOLBE	0.93	0.04	97.03	7.62	1.06	94.75	17.99	19.75	68.75	41.44	6.73	91.89	29.20	3.08	83.67
21BWOLKF	1.54	0.46	90.80	11.47	1.21	94.83	18.99	25.55	66.12	35.15	28.32	85.12	26.95	13.44	75.46
21BWPLBE	1.10	0.08	97.62	10.63	0.76	96.83	20.91	23.43	75.62	30.47	4.99	94.06	14.14	3.62	90.15
21BWPLKF	1.74	0.64	92.74	NA	NA	NA	26.89	45.58	69.56	41.59	12.67	91.81	24.86	9.08	89.21
22BWNLBE	0.65	0.39	91.93	6.17	1.09	96.07	23.23	24.61	95.34	20.83	9.03	92.67	6.69	4.86	89.34
22BWNLCD	0.08	0.48	74.36	12.86	1.57	98.14	20.95	18.79	87.23	14.24	5.24	94.24	0.43	0.51	88.26
22BWNLDM	0.75	0.51	91.76	12.02	1.55	99.13	18.76	9.49	97.14	NA	NA	NA	NA	NA	NA
22BWNLEW	0.13	0.36	83.90	7.80	1.80	96.10	15.30	8.15	91.93	9.39	11.41	90.87	1.65	0.66	92.03
22BWNLGD	0.26	0.44	85.16	17.77	2.50	98.16	10.40	6.58	96.78	20.80	6.97	94.57	20.29	3.05	95.87
22BWNLHL	0.60	0.18	94.30	24.51	2.47	98.43	18.60	12.83	96.64	35.16	4.32	96.74	48.23	5.46	97.49
22BWNLKF	3.78	0.22	98.16	NA	NA	NA	21.53	25.44	89.43	NA	NA	NA	NA	NA	NA
22BWNLKU	0.80	0.39	92.28	11.39	1.62	97.31	21.49	14.34	93.33	14.85	9.39	92.94	7.29	4.44	92.57
22BWNLRA	1.04	0.25	94.55	28.38	4.56	96.98	20.81	21.43	97.15	21.87	4.16	94.82	3.72	1.38	92.55
22BWOLBE	1.41	0.19	92.38	19.58	1.58	94.46	61.79	62.25	83.55	30.09	11.52	85.67	27.91	6.29	88.58
22BWOLKF	2.26	0.16	95.60	NA	NA	NA	89.39	25.15	88.12	36.56	15.52	83.97	39.70	9.07	87.56
22BWPLBE	0.45	0.25	87.59	8.42	1.50	93.51	13.37	15.67	71.51	14.62	11.44	85.77	8.88	5.13	87.94
22BWPLDM	0.60	0.42	89.32	12.80	2.30	96.83	5.07	19.75	52.65	NA	NA	NA	NA	NA	NA
22BWPLEW	0.34	0.53	68.45	15.31	2.58	96.20	10.36	7.07	80.78	8.17	9.58	71.55	3.19	0.54	92.15
22BWPLHL	0.29	0.10	91.04	28.28	1.82	98.38	11.16	17.93	86.00	23.80	5.08	92.00	12.76	2.90	91.54
22BWPLKU	1.36	0.30	91.57	19.38	2.49	97.02	19.30	21.01	83.99	29.29	8.14	90.87	13.26	5.95	88.20
22BWPLRA	1.35	0.36	90.65	20.56	6.68	94.34	12.06	25.30	78.54	29.26	9.06	89.59	8.25	3.66	84.83
23BWNLBE	0.76	0.20	94.74	2.33	0.65	96.84	19.17	17.49	87.65	26.66	6.22	95.00	9.51	1.60	96.93
23BWNLDM	0.21	0.55	87.00	2.93	1.08	97.89	8.54	43.74	93.04	NA	NA	NA	NA	NA	NA
23BWNLHL	0.15	0.21	89.98	12.79	0.52	99.16	29.65	9.74	96.75	9.52	4.25	93.78	NA	NA	NA
23BWNLRA	0.45	0.23	94.74	14.78	1.37	98.46	23.12	29.87	93.24	34.35	7.83	94.44	18.89	4.28	95.15
23BWOPNLKU	0.80	0.61	76.45	52.44	3.29	96.70	57.88	34.20	83.50	16.02	8.18	77.92	6.02	1.76	83.36
23BWPLBE	0.85	0.37	90.48	1.48	0.84	91.05	62.06	19.10	94.18	35.23	6.53	94.14	18.63	2.56	96.04
23BWPLDM	0.40	0.44	76.12	3.77	2.43	89.96	NA	NA	NA	NA	NA	NA	NA	NA	NA
23BWPLHL	0.34	0.25	85.16	13.29	1.01	97.39	80.93	14.67	96.26	12.32	7.60	83.80	NA	NA	NA
23BWPLRA	0.47	0.36	87.86	17.24	1.34	97.28	48.76	32.04	89.07	15.13	11.24	80.56	9.33	3.71	89.10
24BWOPNLBE	2.04	0.19	94.63	17.26	0.90	97.00	66.60	23.16	88.37	54.74	5.38	94.13	72.99	10.17	89.30
24BWOPNLKU	1.00	0.33	84.89	35.15	2.12	96.78	55.89	24.64	87.93	17.64	7.59	82.01	12.87	3.50	88.01
24BWPNLDM	0.23	0.26	82.41	6.38	0.86	97.99	52.01	13.09	97.16	NA	NA	NA	NA	NA	NA
24BWPNLHL	0.27	0.16	88.39	19.14	1.47	98.15	53.09	20.13	95.76	16.44	5.38	92.23	4.75	2.96	90.28
24BWPNLRA	0.59	1.17	79.61	19.19	2.06	97.66	68.79	30.58	97.58	17.84	12.88	89.95	1.83	3.56	88.06
23BWOLBE	NA	NA	NA	11.54	1.60	95.03	42.14	25.86	82.86	41.68	7.04	88.33	24.45	3.40	87.52
23BWOLDM	NA	NA	NA	24.84	8.31	95.60	28.17	16.13	84.10	NA	NA	NA	NA	NA	NA

GYLD = Grain Yield, TKW = Thousand kernel weight, HLW = Hectoliter Weight, PHT= Plant height, DTH = Days to Heading, Gvar = Genetic variance, Evar = Error variance and H²=Heritability

Genetic correlations and trait-based environmental clustering

The implementation of Factor Analytic (FA) models and hierarchical clustering facilitated a robust partitioning of the Genotype by Environment (G×E) setting within the high-altitude wheat-growing regions of Ethiopia. Dendrograms based on the dissimilarity matrices derived from the final FA models illustrated that environmental complexity was inherently trait-dependent (Fig. 1). Based on the Dendrograms, 9 distinct groups developed for grain yield (GYLD), while four were

identified for hectoliter weight (HLW), three for thousand kernel weight (TKW), and two each for days to heading (DTH) and plant height (PHT). These groupings represented sets of correlated environments in which genotype performance was relatively consistent within each cluster, allowing accurate ranking of genotypes based on Best Linear Unbiased Prediction (BLUP) values.

The hierarchical clustering for grain yield revealed a significant spatial contrast within the testing network. At a 0.4 threshold, the environments partitioned into nine distinct clusters, reflecting the high spatial

variability of Ethiopia's high-altitude regions. A critical finding was the consistent isolation of the Enawari environment (22BWPLEW), which appeared as a standalone outlier at the highest dissimilarity level (~0.8), representing a unique ecological niche. On the other hand, major environments such as Bekoji (21BWOLBE, 24BWPNLBE) and Kulumsa (22BWPLKU) showed strong genetic associations despite their altitudinal gradient (2780 m vs. 2200 m), whereas the unique temperature-moisture regime at the Enawari site induced a distinct physiological response. This isolation underscored the necessity of a specific adaptation-breeding strategy for the Enawari highlands to avoid the masking effect of average performance in more consistent Arsi-zone sites like Arsi-Robe (22BWNLRA).

For phenological and growth traits, the analysis revealed a substantial temporal shift between the 2021–2023 baseline and the 2024 cropping season. The high Agglomerative Coefficients (0.89 for Plant Height and 0.90 for Days to Heading) indicated a highly structured dataset where the 2024 groups formed a distinct "outgroup." According to Jenks (2025) and Boyko and Tkachyk (2023), the tight clustering of the 2021–2023 samples (merge heights below 0.2) suggested a period of relative environmental stability. The 2024 environments were isolated at a dissimilarity height of 0.69, suggesting a major climatic or unconditional shift likely resulted from varying rainfall patterns in the Ethiopian highlands. Such "temporal G×E" highlighted the importance of multi-year testing to ensure that phenological stability and lodging resistance were maintained under shifting climatic cycles (Bruzzese and Vistocco, 2025). Moreover, the analysis of Thousand Kernel Weight (TKW) and Hectoliter Weight (HLW) reinforced the

vigorous clustering patterns, with Agglomerative Coefficients of 0.81 and 0.84, respectively. While the core environments of the Arsi highlands formed cohesive groups, Enawari (22BWPLEW) was again identified as the primary outlier, merging with the rest of the dataset only at maximum dissimilarity levels (0.78 for TKW and 0.80 for HLW). As discussed by Bruzzese and Vistocco (2025), this "outgroup" status for Enawari underscored a fundamental divergence in grain-filling conditions. The specific higher altitude-rainfall combination at 2650 m potentially extended the grain-filling duration, which fundamentally distinguished its study population from relatively high altitude sites. Supported by the evolutionary visualization frameworks of Müller *et al.* (2024), these findings ensured that regional breeding partitions were grounded in the landscape's inherent mathematical and ecological realities. Consequently, genotypes with superior "stay-green" traits showed specific adaptation to the Enawari niche, whereas they were penalized in environments where terminal stresses were more predominant.

The structural contradiction identified across all traits provided a clear statistical basis for re-evaluating wheat breeding sub-zones in Ethiopia. The high genetic diversity observed between the outlying Enawari cluster and the central Arsi-zone clusters offered a strategic opportunity to design a breeding program that can enhance the production and productivity of bread wheat in high-altitude areas of the country. By identifying these natural divisions without pre-specifying cluster counts, this study ensured that the resulting partitions reflected the data's inherent biological relationships, facilitating the selection of genotypes precisely tailored to the diverse high-altitude agro-ecologies of Ethiopia.

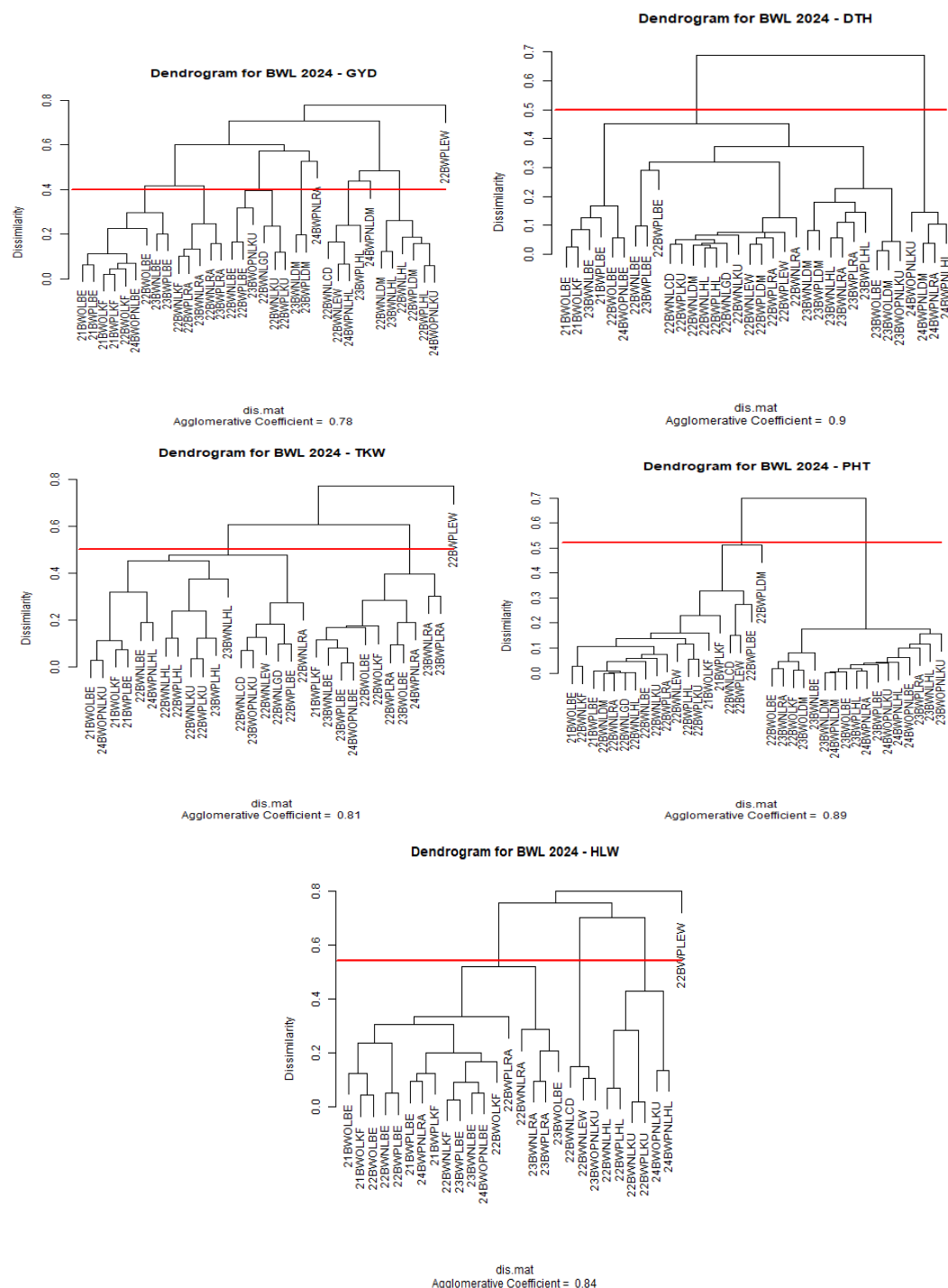


Fig 1. Dendrogram of the dissimilarity matrix from the final FA models.

In this study, heatmaps of genetic correlations were generated to visualize relationships among environments (Fig. 2) for each trait, with the color gradient indicating the strength and direction of environmental similarity. Deep red cells indicated strong positive correlations, signifying environments with similar genetic responses, while deep blue cells highlighted strong negative correlations, indicating contrasting growing conditions. As noted

by [Smith and Cullis \(2018\)](#), these visual tools were essential for Multi-Environment Trial (MET) analysis, clarifying the correlation structures that inform regional breeding decisions. Hence, based on the results of this study, the genetic correlation analysis for GYLD revealed a complex landscape of environmental interactions across the network of high-altitude bread wheat trial sites. Thus, distinct clusters of high positive correlations were identified,

particularly among sites such as 21BWOLBE, 21BWPLBE, and 21BWOLKF, where correlation coefficients reached near-unity. These clusters defined specific "mega-environments" in which genotype-by-environment interaction (GEI) is minimized, showing that such alignment enables the consolidation of breeding resources by selecting representative sites to predict performance across the group. The consistency in these highland clusters suggests that physiological mechanisms, such as nutrient uptake efficiency and frost resilience, remain stable (Bornhofen *et al.*, 2021). Conversely, low-to-moderate correlations between distant clusters—notably between 23BWNLRA and the EBWOLBE group—indicated significant environment-specific genetic expression. These findings implied that different genetic makeup enhanced yield under varying local stressors, such as moisture fluctuations or soil acidity, which are common in Ethiopian highland vertisols (Zemedu *et al.*, 2024). For the national program, these low-correlation zones necessitate continued multi-environment trials (METs) to capture crossover interactions. Success in stable environments did not necessarily translate to these variable locations, reinforcing the need for site-specific selection (Tolessa *et al.*, 2024; Tadesse *et al.*, 2025).

Moreover, genetic correlation for DTH indicated high environmental uniformity across most sites, particularly for the 2024 trials. A central cluster, including 22BWNLCD, 22BWPLKU, 22BWNLHL, and 22BWPLHL, exhibited a near-perfect positive correlation, indicating that genotype rankings for flowering time were highly stable, allowing selection at one site to effectively predict outcomes at others. Such high correlations are typical of phenological traits, which generally exhibit higher heritability and lower GEI than polygenic traits such as yield (Kabir *et al.*, 2024). However, peripheral sites such as 24BWPNLKU and 21BWOLBE showed significantly lower correlations, indicating greater environmental sensitivity. These likely represented unique agro-climatic zones where specific cues, such as photoperiod or temperature variations at different elevations, caused divergent genetic pathways (Masawi *et al.*, 2025). Generally, the genetic control of flowering time remained remarkably consistent across the highland environments tested in Ethiopia, despite specific outliers that responded differently to local conditions. For PHT, the

genetic correlation matrix indicated significant clustering and varying degrees of stability. High positive correlations, approaching unity, were observed in a cluster comprising 21BWOLBE, 22BWNLRA, 22BWPLRA, and 22BWPLHU, facilitating predictable selection outcomes (Begna and Begna, 2025). Conversely, lower correlation values (0.0–0.5) were observed at more recent sites such as 24BWPNLDM and 23BWOPNLKU. This divergence highlighted substantial GEI, suggesting that shifting climatic factors or management practices were introducing new sources of variation for plant height. As proposed by Lee *et al.* (2025), advanced spatial factor-analytic approaches are essential for accurately partitioning these shifting genetic effects in modern breeding cycles, especially since plant architecture directly influences lodging resistance.

Regarding TKW, most sites exhibited a complex landscape of interactions, with most site-to-site comparisons showing robust positive correlations, consistent with Cooper *et al.* (2021), who noted that yield components such as kernel weight generally exhibit lower GEI than total grain yield. However, site 22BWPLBW was a significant outlier, displaying near-zero correlations. This suggested unique stressors, likely terminal frost or other stresses, which altered the typical grain-filling process in that specific highland zone (Crespo-Herrera *et al.*, 2021). Moreover, the correlation matrix for hectoliter weight (HLW) revealed high genetic similarity across most environments, with strong correlations (>0.70) in the primary cluster. Nevertheless, a second cluster exhibited significant crossover GEI, particularly at 22BWNLCD and 22BWNLHL, with near-zero or negative correlations relative to the primary cluster, indicating significantly altered genotype rankings. These interactions necessitate partitioning the bread wheat breeding program into target populations of environments (TPEs) to maximize local yield potential and ensure regional food security (Mondal *et al.*, 2024). Generally, genetic correlation analysis revealed that while heading date and kernel weight remained stable across environments, grain yield and plant height exhibited site-specific variability. Hectoliter weight was generally consistent but varied across locations (Worede *et al.*, 2025). Therefore, these findings necessitated the use of Target Populations of Environments (TPEs) to tailor breeding strategies effectively across highland.

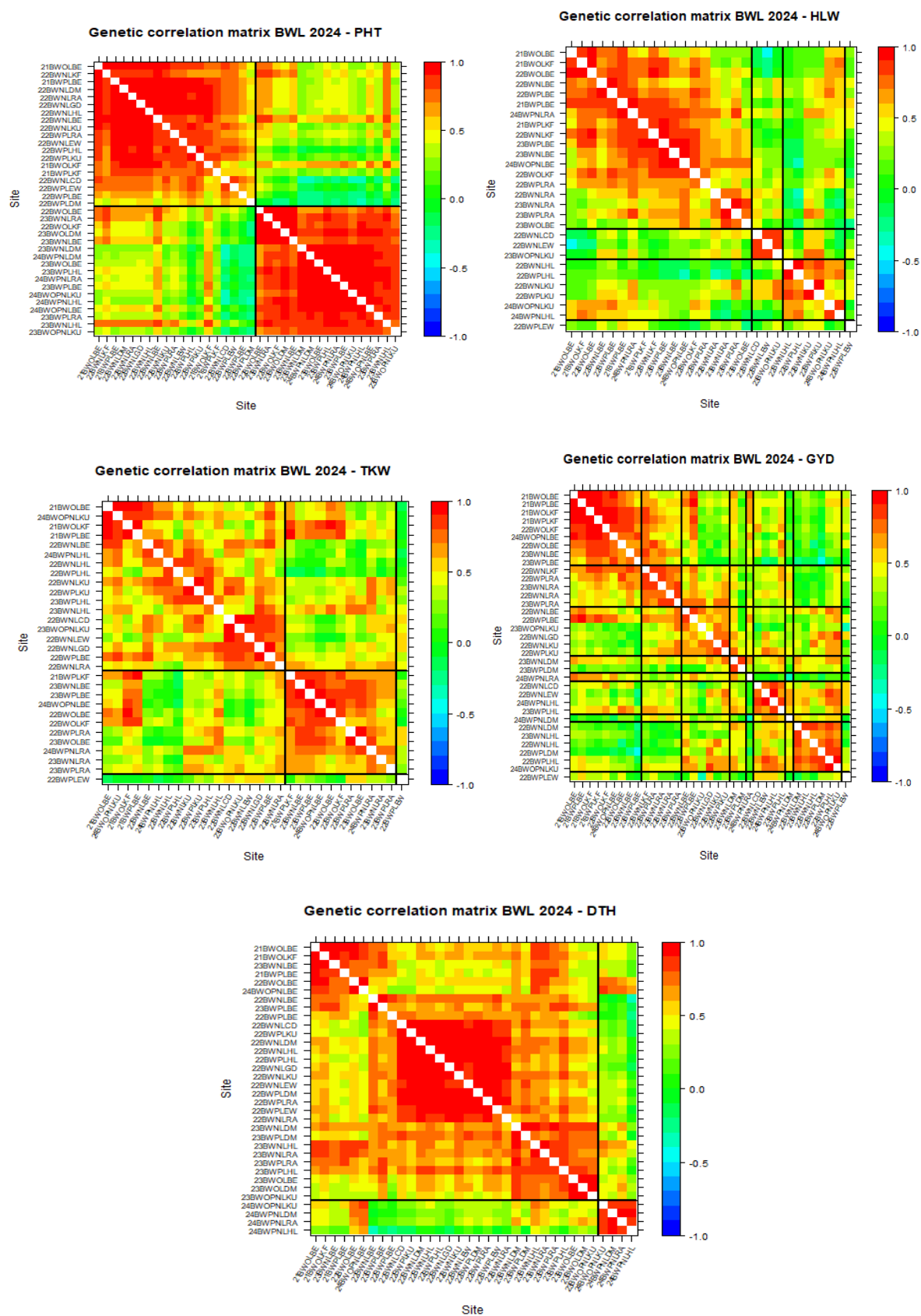


Fig 2. Heat map representation of the genetic correlation matrix from the final FA models.

Factor analysis

Factor analytic (FA) models were employed to analyze G×E interactions while preserving spatial variability from individual trials. Model adequacy could be assessed by the proportion of G×E variance explained by factor components, both within individual trials and across all trials (Smith *et al.*, 2018). Thus, applying an eight-factor analytic (FA_8) model to the multi-environment trial (MET) data yielded a granular decomposition of genetic variance, as summarized in Table 4. Hence, in this study, the FA models effectively estimated and predicted G×E effects and quantified G×E variance. Similar results have been reported for common bean (Argaw *et al.*, 2024), finger millet (Tesfaye *et al.*, 2023), and durum wheat (Tadese *et al.*, 2021), confirming the efficiency of FA models in MET genetic analyses. The model revealed unique expressive power, with the cumulative explained variance across the eight latent factors typically exceeding 99.9%. This indicated that the preferred FA framework was sufficient to capture the vast majority of the complex covariance structure within the genotype-by-environment interaction (Smith *et al.*, 2015). Hence, the results identified a distinct group of environments in which the first underlying factor (FA_1) was the dominant driver of genetic variation. In the 2021 and 2022 trials, sites such as 21BWOLKF (89.57%), 21BWPLKF (91.98%), and 22BWNLFK (86.37%) exhibited high FA_1 loadings, suggesting a strong genetic correlation and minimal rank change among genotypes at these locations (Beeck *et al.*, 2010). Moreover, such a trend continued into the 2024 season with 24BWOPNLBE (74.09%), indicating that these environments constituted the core of the Target Population of Environments (TPE), where selection for broad adaptation and yield stability is most effective (Cullis *et al.*, 2010).

In contrast, a significant subset of environments displayed more complex variance structures, where secondary factors (FA_2 through FA_8) were hidden or

matched the primary factor. For the 2022 trials, a major shift toward Factor 2 was observed in 22BWNLHL (63.54%), 22BWNLU (63.60%), and 22BWPLDM (62.85%) trials, showing that these high F2 loadings signify a distinct environmental stimulus—potentially related to seasonal rainfall patterns or soil characteristics—that differentiated the trials from the FA_1-dominant group (Gogel *et al.*, 2018). Similarly, 22BWPLEW was defined exclusively by FA_5 (44.00%), illustrating the model's ability to isolate niche-specific genetic responses. The 2023 and 2024 data further highlighted this environmental divergence. In site 23BWPLDM, FA_6 became the primary source of variation (63.77%), while 23BWOPNLKU was heavily influenced by FA_4 (37.87%). In the 2024 trials, 24BWPNLRA and 24BWPNLHL were primarily driven by FA_4, accounting for 39.17% and 35.02% of the variance, respectively. These findings suggested that higher-order factors are not merely noise but represent specific physiological constraints or environmental categories that require targeted selection strategies to maximize genetic gain (Piepho *et al.*, 2012). A notable finding was the discrepancy in total explained variance for site 22BWNLRA, which reached only 85.67%. This indicated a substantial proportion of "specific" or residual genetic variance (approximately 14.33%) that remains independent of the common factors shared by the rest of the testing network (Kelly *et al.*, 2007). Such environments represent unique microclimates where genotype performance may be entirely decoupled from the broader TPE. Ultimately, the distribution of loadings across the eight factors revealed a highly segmented environmental landscape. By quantifying the exact contribution of each latent factor to the total genetic variance at every site, the FA model provided a statistically rigorous foundation for identifying genotypes with either robust stability across the FA_1 dominant sites or specialized adaptation to the niches defined by factors FA_2 through FA_8 (Smith *et al.*, 2019).

Table 4. Results from fitting the factor analytic model.

Site	FA_1	FA_2	FA_3	FA_4	FA_5	FA_6	FA_7	FA_8	Total
21BWOLBE	71.86	1.15	19.36	1.92	2.34	0.27	0.41	2.68	99.99
21BWOLKF	89.57	6.00	3.80	0.00	0.00	0.00	0.38	0.24	99.99
21BWPLBE	80.33	0.02	7.79	0.33	8.57	2.59	0.18	0.18	99.99
21BWPLKF	91.98	4.10	1.81	0.80	0.58	0.16	0.55	0.03	100.01
22BWNLEB	45.55	25.04	3.28	6.16	0.00	16.85	0.99	2.13	100.00
22BWNLCD	24.04	3.31	1.25	25.85	4.37	0.43	33.30	7.44	99.99
22BWNLDM	4.89	45.93	18.69	4.57	17.82	0.10	2.67	5.34	100.01
22BWNLEW	15.66	48.32	0.03	18.15	10.91	0.13	5.39	1.42	100.01
22BWNLGD	21.20	46.01	0.20	0.29	5.72	2.18	23.30	1.11	100.01
22BWNLHL	5.24	63.54	15.31	0.23	10.04	0.54	0.00	5.09	99.99
22BWNLKF	86.37	0.01	7.22	0.74	4.67	0.03	0.92	0.05	100.01
22BWNLKU	16.06	63.60	0.71	12.39	0.02	1.61	0.06	5.55	100.00
22BWNLRA	45.92	7.52	23.96	0.39	1.84	1.91	4.05	0.08	85.67
22BWOLBE	68.04	9.68	1.10	8.21	3.68	2.09	0.26	6.94	100.00
22BWOLKF	83.13	6.10	4.04	1.62	0.34	1.45	2.02	1.30	100.00
22BWPLBE	74.10	4.07	0.71	6.63	5.34	5.30	3.85	0.00	100.00
22BWPLDM	1.21	62.85	8.60	15.81	0.82	3.43	7.28	0.02	100.02
22BWPLEW	11.99	0.01	2.30	25.45	44.00	0.42	7.06	8.77	100.00
22BWPLHL	23.70	52.68	13.78	6.29	0.09	1.78	1.24	0.44	100.00
22BWPLKU	29.47	47.61	4.21	2.60	8.70	6.44	0.91	0.06	100.00
22BWPLRA	67.40	0.00	24.51	4.31	1.43	0.40	1.30	0.63	99.98
23BWNLEB	70.56	6.82	2.03	0.22	3.93	0.21	14.83	1.40	100.00
23BWNLDM	41.21	18.09	5.15	0.03	0.11	26.26	0.80	8.34	99.99
23BWNLRA	58.92	2.06	29.68	0.64	0.54	5.92	0.46	1.78	100.00
23BWOPNLKU	13.42	32.41	2.84	37.87	1.23	2.93	0.20	9.11	100.01
23BWPLBE	49.42	27.96	3.96	2.46	12.96	0.30	1.91	1.03	100.00
23BWPLDM	7.65	10.87	5.82	1.76	9.53	63.77	0.54	0.06	100.00
23BWPLHL	45.99	2.99	0.46	28.06	0.28	12.81	7.72	1.69	100.00
23BWPLRA	49.34	13.98	17.73	17.00	0.04	0.96	0.75	0.20	100.00
24BWOPNLBE	74.09	11.15	9.36	3.19	2.05	0.06	0.07	0.03	100.00
24BWOPNLKU	35.99	43.47	5.85	10.97	0.32	0.14	2.48	0.77	99.99
24BWPNLDM	9.21	27.61	16.15	15.91	3.77	8.82	9.05	9.48	100.00
24BWPNLHL	21.78	22.97	0.00	35.02	3.76	2.78	13.34	0.37	100.02
24BWPNLRA	32.38	0.04	12.65	39.17	5.06	10.59	0.11	0.00	100.00

Disease reaction and mean performance of advanced genotypes

The evaluation of advanced bread wheat genotypes and two commercial checks revealed significant genetic variability in resistance to yellow rust (Yr) and stem rust (Sr), as well as in mean trait performance across two testing environments (Table 5). At the Meraro site, the lowest yellow rust scores were recorded for genotypes EBW170051 (1.47) and EBW170058 (1.54), both of which performed comparably to the resistant commercial check, Kulumsa (1.55). These high levels of resistance were notably associated with longer phenological cycles; for example, EBW170058 recorded the maximum of 77.49 days to heading (DTH), suggesting a possible correlation between extended vegetative growth and rust resilience in this specific germplasm. Conversely, genotypes EBW233267 and EBW233059 displayed high susceptibility to yellow rust with scores of 7.06 and 6.81, respectively. Notably, the earliest maturing genotype, EBW233267 (70.19 DTH), exhibited the highest susceptibility, indicating that early heading did not serve

as an effective escape mechanism against yellow rust in this environment. Regarding stem rust (Sr) at the Debrezeit site, reactions were more uniform among the genotypes, though distinct levels of terminal resistance were observed. Thus, genotype EBW222276 demonstrated the most robust resistance at this location with a score of 5.01. In contrast, the majority of the EBW1700 series reached a susceptibility peak of 6.80 at Debrezeit, suggesting a potential breakdown of resistance under the high inoculum pressure characteristic of that specific site. These results align with findings by [Figueroa *et al.* \(2018\)](#) and [Singh *et al.* \(2020\)](#), which emphasized that multi-location screening is essential for identifying genotypes with broad-spectrum and durable resistance against evolving rust races.

On the other hand, mean grain yield (GYLD) across the genotypes was highly variable, ranging from 3.02 t ha⁻¹ to 5.31 t ha⁻¹. The advanced genotypes EBW170049 (5.31 t ha⁻¹) and EBW170074 (5.17 t ha⁻¹) achieved the highest mean yield, followed closely by EBW222923 (5.06 t ha⁻¹) and EBW170051 (5.03 t ha⁻¹). The superior performance of

EBW170049 was complemented by strong grain quality parameters, including a Hectoliter Weight (HLW) of 70.37 kg hl⁻¹ and a Thousand Kernel Weight (TKW) of 38.46 g, suggesting a stable and efficient grain-filling period. On the other hand, the check Danda'a and the genotype EBW233267 recorded the lowest yields of 3.02 t ha⁻¹ and 3.10 t ha⁻¹, respectively. This reduction in yield appeared to correlate directly with higher disease pressure and lower TKW. Additionally, genotype EBW170074 was particularly notable for producing the highest TKW in the study (41.25 g), outperforming both commercial checks and all other entries. Such variations in yield components are typical in advanced wheat germplasm, where high kernel weight often compensates for fewer tillers or shorter grain-filling durations (Gazzola *et al.*, 2021).

Furthermore, phenological development and plant architecture also showed significant differences ($P < 0.05$) among the tested genotypes. Days to heading (DTH) ranged from a minimum of 70.19 days in EBW233267 to a maximum of 77.49 days in EBW170058. While early heading is often viewed as an escape mechanism for terminal stresses, in this study, it was associated with lower yield potential and higher disease susceptibility. In terms of plant height (PHT), the check Danda'a and genotype

EBW160066 reached the maximum height of 101.42 cm, while the shortest stature was observed in EBW222919 at 87.82 cm. Genotype EBW222276 also maintained a semi-dwarf stature of 88.68 cm while providing robust resistance to stem rust. The integration of reduced plant height in these genotypes suggests improved lodging resistance, which remains a key objective in modern wheat breeding for high-productivity systems (Würschum *et al.*, 2018). Ultimately, the EBW1700 series—specifically EBW170049 and EBW170074—performed as the most promising candidates, successfully combining high grain yield (up to 5.31 t ha⁻¹) and superior kernel weight (41.25 g) with competitive resistance against yellow and stem rust profiles and stable phenological development. Moreover, EBW222276 was also identified as a critical genetic resource for its robust terminal stem rust resistance and desirable semi-dwarf architecture. The integration of high yield potential, superior kernel weight, and broad-spectrum rust resistance makes these advanced genotypes the best candidates for variety development. Consequently, they are the most promising selections for formal release or as parental lines in future breeding programs designed to boost productivity in rust-prone highland areas of Ethiopia.

Table 5. Disease reaction and mean traits of advanced genotypes with two checks.

Genotypes	Yellow Rust (Yr)		Stem Rust (Sr)	Mean of traits				
	Meraro	Bekoji		GYLD	DTH	HLW	PHT	TKW
EBW202276	3.49	4.47	6.70	4.91	71.9	70.70	88.96	35.48
EBW212274	2.79	3.98	5.90	4.34	72.98	66.68	90.33	37.94
EBW160066	1.92	3.42	5.18	4.32	70.81	69.83	101.4	38.86
EBW222276	3.13	4.30	5.01	4.58	73.95	72.04	88.68	39.19
EBW233267	7.06	7.01	5.24	3.10	70.19	64.17	90.64	31.87
EBW170049	1.68	3.27	6.80	5.31	76.63	70.37	98.24	38.46
EBW170051	1.47	3.15	6.80	5.03	77.43	70.64	94.71	37.94
EBW170052	1.82	3.35	6.80	4.63	74.11	70.71	94.87	38.68
EBW170057	2.24	3.60	6.80	4.55	72.66	70.46	94.59	37.95
EBW170058	1.54	3.16	6.80	4.91	77.49	71.00	97.41	39.95
EBW170074	2.60	3.89	6.10	5.17	71.04	69.96	95.92	41.25
EBW222919	3.04	4.21	5.57	4.11	71.71	67.30	87.82	40.83
EBW222923	2.15	3.53	5.94	5.06	73.9	69.75	87.87	38.15
EBW233059	6.81	6.85	6.08	3.63	72.38	64.79	88.06	35.64
EBW232809	2.92	4.06	5.48	4.18	72.52	70.06	92.12	38.05
EBW232499	1.75	3.32	5.48	4.01	73.33	68.65	89.82	36.83
Danda'a	4.57	5.28	6.16	3.02	76.91	60.34	101.42	32.03
Kulumsa	1.85	3.84	6.93	4.47	73.12	71.02	94.46	40.27

Conclusion

The evaluation of bread wheat trials across the Ethiopian highlands from 2021 to 2024 established that productivity was fundamentally governed by complex Genotype by Environment (G×E) interactions, with grain yields fluctuating significantly between 1.54 and 7.39 t ha⁻¹ across environmental sites. Using a Factor Analytic Mixed Model (FAMM), the study partitioned 99.9% of the genetic variance and identified 65% of elite genotypes that consistently surpassed the 4.5 t ha⁻¹ threshold across diverse agro-ecologies such as 24BWOPNLBE and 22BWNHLHL. The top performing genotypes EBW170049 (5.31 t ha⁻¹) and EBW170074 (5.17 t ha⁻¹), followed closely by EBW222923 (5.06 t ha⁻¹) and EBW170051 (5.03 t ha⁻¹), achieved stability through a high-yield, early-maturity ideotype, characterized by a semi-dwarf architecture and a precise day to heading frame, which served as a critical stress-escape strategy. Hierarchical clustering and Factor_1 loadings exceeding 85% at sites such as Bekoji and Kulumsa confirmed the Arsi highlands as the core Target Population of Environments (TPE), where high broad-sense heritability of phenological traits enabled breeders to maintain a stable maturity window despite varying altitudes and seasonal rainfall. Hence, environmental clustering successfully partitioned the test sites into distinct mega-environments based on their discriminatory power and representativeness, highlighting that while most Arsi highland locations were highly correlated, specific seasons exhibited outlier behaviors due to erratic rainfall distributions that disrupted traditional genetic rankings. The identification of ecological outliers such as Enawari (22BWPLEW) and Arsi-Robe (22BWNLRA) highlighted unique physiological constraints that were often obscured by regional averages.

Furthermore, correlation analysis and disease screening revealed that while yield was positively associated with quality parameters such as Hectoliter Weight (HLW > 78 kg hL⁻¹) and Thousand Kernel Weight (TKW), it showed a strong negative correlation with rust severity, particularly in hotspots like Meraro. The EBW1700 series genotypes, notably EBW170049 and EBW170074, demonstrated significant genetic progress by combining high productivity with broad-spectrum rust resistance, achieving yields up to 5.31 t ha⁻¹ and a TKW of 41.25 g, representing a 9–14% advantage over standard checks.

Conversely, early-maturing genotypes such as EBW233267 failed to escape virulent rust races in high-pressure areas, proving that phenological escape must be integrated with robust genetic resistance to prevent kernel shriveling and yield loss. Ultimately, while EBW22276 offered valuable specific stem rust resistance, the EBW1700 series and related elite genotypes were designated the highest priority for variety advancement due to their superior stability, milling quality traits, and their ability to maintain high harvest indices under the combined pressures of biotic stress and shifting climatic patterns across the moisture-variable and disease-prone environments of wheat belt highland areas of Ethiopia. In summary, the study successfully applied FAMM-based MET analysis to identify top-performing genotypes, thereby offering breeding programs a proven methodology to accelerate genetic gains and secure climate-resilient wheat production under highland agroecologies of Ethiopia.

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