

Article

GT Biplot and Cluster Analysis of Barley (*Hordeum vulgare* L.) Germplasm from Various Geographical Regions Based on Agro-Morphological Traits

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Abstract: Barley, an ancient crop, was vital for early civilizations and has historically been served as food and beverage. Today, it plays a major role as feed for livestock. Breeding modern barley varieties for high yield and quality has created significant genetic erosion. This highlights the importance of tapping into genetic and genomic resources to develop new improved varieties that can overcome agricultural bottlenecks and increase barley yield. In the current study, 75 barley genotypes were evaluated for agro-morphological traits. The relationships among these traits were determined based on genotype by trait (GT) biplot analysis for two cropping years (2021 and 2022). This study was designed as a randomized complete block experiment with four replications. The variation among genotypes was found to be significant for all traits. The correlation coefficient and GT biplot revealed that grain yield (GY) was positively correlated with the number of grains per spike (NGS), the grain weight per spike (GW), and the thousand kernel weight (1000 KW). However, the test weight (TW) was negatively correlated with the heading date (HD). Hierarchical analysis produced five groups in the first year, four groups in the second year, and four groups over the average of two years. Genotypes by trait biplot analysis highlighted G25, G28, G61, G73, and G74 as promising high-yielding barley genotypes. This study demonstrated the effectiveness of the GT biplot as a valuable approach for identifying superior genotypes with contrasting traits. It is considered that this approach could be used to evaluate the barley genetic material in breeding programs.

Keywords: barley; GT biplot; correlation coefficients; hierarchical cluster analysis



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1. Introduction

Barley was important as a major crop in Old-World agriculture and is one of the earliest domesticated plants [1]. *Hordeum vulgare* ssp. *spontaneum* (C. Koch) Thell. is one of the progenitors of cultivated barley (*Hordeum vulgare* ssp. *vulgare* L.), and both are diploid species ($2n = 14$) belonging to the family *Poaceae* [2]. It has long been acknowledged that a key hub for the domestication of barley was the Near East Fertile Crescent [3]. Genomic and geographical analyses have identified three distinct genetic clusters of wild barley: (i) the Levant and Southern Turkey cluster, (ii) the Southeastern Turkey cluster, and (iii) the

Eastern and Middle Asian cluster. The Levant cluster stands out for having the highest diversity in the wild *Hordeum vulgare* ssp. gene pool, which underscores its importance in barley's evolutionary history [4]. Regions such as the Himalayas, Eritrea, Ethiopia, and Morocco have also been identified as areas where barley was domesticated and cultivated [5]. Recent molecular evidence indicates that Central Asia, located 1500–3000 km further east from the Fertile Crescent [6], and Tibet in China [7] are additional regions where wild barley domestication may have occurred. This supports the theory that cultivated barley originated from multiple areas and implies that domestication might have occurred independently in various geographical regions [8]. Barley primarily functions as animal feed and is also used in beer production. It is a staple food in regions where other grains may be challenging to cultivate [9,10]. Barley has an extensive geographic range, surpassing nearly every other crop species. Its cultivation extends from the highest arable mountaintops down to coastal regions. The coverage ranges from the northernmost to the southernmost latitudes and from temperate zones to the subtropic regions [11].

Barley is the fourth most important cereal crop worldwide, with cultivation extending across 100+ countries. Over the past decade, Europe has been a major contributor to barley production, accounting for approximately 60% of the global barley tonnage, with Asia (15%) and the Americas (13%) following closely behind. Global barley cultivation spans 47.15 million ha, yielding 154.9 million tons [12]. In 2022, Turkey (5.5%) ranked fifth in barley production globally, followed by European countries, Russia, China, and Canada. The average barley yield is 3285 kg ha⁻¹. Barley in Turkey is approximately 19% lower than the world average barley yield at 2665 kg ha⁻¹. In Turkey, with the development of new cultivars through breeding efforts and improvements in cultivation techniques, the average yield in barley production has surged by 33% over the last 30 years (1992–2022). It has risen from 2010 kg ha⁻¹ to 2665 kg ha⁻¹ [13].

Grain yield (GY), an intricate quantitative trait, is influenced by various characteristics related to yield [14]. Breeders can greatly benefit from examining correlations between these characteristics, especially the correlations between grain yield and other traits. This could help in selecting the optimal features that contribute to increasing yield [15]. One useful approach is correlation analysis, which suggests that improving one feature may improve other positively linked traits as well. The significance of correlation studies in breeding programs is underscored especially by the connection between inheritable traits and crucial characteristics such as yield [16].

Cluster analysis, a multivariate approach, stands out for its widespread ability to depict genetic diversity based on genotype similarities or disparities [17]. The advent of modern agriculture has witnessed the replacement of traditional landraces and diversified germplasm with homogeneous pure-line varieties. This has made crops more susceptible to environmental changes and other stressors. Thoroughly examining the prevalent diversity within the germplasm of different barley collections is imperative to broaden the pool of parental materials for breeding endeavors and to establish effective crop enhancement programs. Agro-morphological characterization has emerged as a pivotal initial step in this process, facilitating the assessment of genetic variability, the differentiation among plant materials from distinct geographical origins, the establishment of core collections, and the prioritization of accessions for strategic inbreeding [18].

The primary aim of crop improvement initiatives is to breed varieties that demonstrate adaptability across diverse ecological conditions. The genotype and genotype by environment (GGE) biplot analysis plays a crucial role in achieving this goal by illustrating genotype stability and adaptability. This analysis proves particularly insightful when a substantial portion of the variance in genotypes (G) and genotype by environment interactions (GEI) can be captured within the first two principal components [19]. Among the various methods of GGE biplot analysis, the genotype by trait (GT biplot) interaction, as introduced by Yan and Rajcan [20], stands out. They suggested that the GT biplot is a suitable and effective tool for discerning interactions between cultivars and traits. The utilization of the GT biplot technique has been used in prior studies to evaluate genotypes across multiple

traits and explore the correlations between yield and various characteristics, including morphological, physiological, and qualitative traits. This was conducted for a variety of crops including wheat [21], barley [22], and sweet corn [23].

In this study, we aimed to

- i: Evaluate the yield and agronomic performance of barley genotypes;
- ii: Examine the relationships among traits;
- iii: Explore the use of GT biplot analysis for barley improvement programs.

2. Materials and Methods

2.1. Experimental Site and Climatic Characteristics

In this study, 75 barley genotypes were identified from plant material collected from six different regions (Table 1). The plant material included barley genotypes from Austria, Bulgaria, Iran, Pakistan, Syria, and Turkey. The trial was conducted in Elmacık village, in the Gumusova district of Duzce province during the 2020–2021 and 2021–2022 cropping years. The trial site was located at approximately 40°50′23.2″ North latitude and 30°58′24.3″ East longitude and was located 160 m above sea level.

Table 1. Barley genotypes' codes, names, pedigrees, spike type, and origin.

Code	Cultivar	Pedigree	Spike Type (Row)	Origin
G1	Zahir	K10 × Kt 1206	2	Bulgaria
G2	IZ Sayra	Alfa × Nutans 85242/76/ × Yubileı 100	2	Bulgaria
G3	Zemela	F2 2012/01 × Kt 2152 200 Gy—mutant	6	Bulgaria
G4	Alekssan	K-2169-01 × Kt 2145	6	Bulgaria
G5	Odisey	Obzor × Nutans 85242/64/ × Nutans 85242/64 (5)	2	Bulgaria
G6	Orfej	Kjfi × Nutans 8486/40	2	Bulgaria
G7	Dariya	CRT 059 × Lambic	2	Bulgaria
G8	Asparuh	2119Y-75 × Korten	2	Bulgaria
G9	Emon	137HS-21/M-21-H/3/Malta/M-20-H/M-21-H/4/111G-65	2	Bulgaria
G10	Ahil	Clarine 300 Gy—mutant	2	Bulgaria
G11	Deviniya	Tamara × Aster	2	Bulgaria
G12	Vesletc	№102/121 × Karnobat	6	Bulgaria
G13	Zagoretz	4943—41 × 1023K-7	2	Bulgaria
G14	Kuber	2119Y-41 × 2119Y-165	2	Bulgaria
G15	Obzor	28 H 46-10 × TpyMφ	2	Bulgaria
G16	Lardeya	Alfa × Nutans 85242/76/ × Alfa	2	Bulgaria
G17	IZ Bori	K 280-7 NaN3—mutant	6	Bulgaria
G18	Bul Perun	Alpha × Jet	2	Bulgaria
G19	Bozhin	H280—7/NaN3—mutant	6	Bulgaria
G20	Aheloy 2	Hemus × №102/121	6	Bulgaria
G21	Özdemir	CUM/4060//P12-62/P169-2	2	Türkiye
G22	Cumhuriyet 50	No:28 (Kayseri)/Mansholt's-2 Rijige (Holland)	2	Türkiye
G23	İnce-04	4671/Tokak//4648/p12-119/3/WBCB-4	2	Türkiye
G24	Bilgi-91	Selection	2	Türkiye
G25	Boram	Unknown	6	Türkiye

Table 1. Cont.

Code	Cultivar	Pedigree	Spike Type (Row)	Origin
G26	İmbat	80.5064//BOLDO/MJA/3/GEM	6	Türkiye
G27	Hilal	Melusine/Aleli/3/Matico/Jet//Shyri/4/Canela/5/Arupo/ K8755//Mora/3/Canela CBSS 96M00698D-P-5M-1Y-1M-0Y	2	Türkiye
G28	Akhisar 98	GEM*4/PIAST SEA-2636-4S-3S-2S-1S-0S	6	Türkiye
G29	Ayrancı	Osk4.197/12- 84//HB854/Astrix/3/Rod/4/Slad/3/Vict//Yrm/Lhfm	2	Türkiye
G30	Akar	Alpha/Durra//Antares/KY-63-1294/3/Tarm 92	2	Türkiye
G31	Karatay 94	VONTAGE/GÜZAK//TAPLANI/3/REKAL/CUM50/RIGIC	2	Türkiye
G32	Kıral 97	ADAIK/SL//WA1094-67	6	Türkiye
G33	Konevi	CO55/OWB 710-80 (WBCB)	2	Türkiye
G34	Larende	ALM (4652)/TOKAK//342TH/P12-119/3/W.BELT22	2	Türkiye
G35	Yesevi 93	Tokak/local population 4857	2	Türkiye
G36	Burakbey	Coss/OWB 71080-44-1H//Obruk 86	2	Türkiye
G37	Tarm-92	Tokak/local population no 4875	2	Türkiye
G38	Orza 96	Tokak 157-37/4857	2	Türkiye
G39	Zeynel Ağa	Antares/Ky63-1294//Lignee131	2	Türkiye
G40	Tosunpaşa	Atlas/Zarjou	2	Türkiye
G41	Bülbül 89	13GTH/local population	2	Türkiye
G42	Avcı-2002	Sci/3Gi-72AB58, F1//WA1245141	6	Türkiye
G43	Çetin 2000	Star (Iran) /line 4875	6	Türkiye
G44	Aydanhanım	GK Omega/Tarm 92	2	Türkiye
G45	Bozlak	Unknown	2	Türkiye
G46	Vamikhoca 98	GEM*3/3/CR 115/POR//BLANCO MA	6	Türkiye
G47	Bayrak	ARRAYAN/OLMO//LEO- B/3/Lignee527/Aths//Aths/Lignee686	6	Türkiye
G48	Sancak	1861112/ROBUR/7/HLLA/EH 21B/6/MAN/HUIZ//M69.69/3/APAM/RL//H 272/4/CP/BRA/5/JOSO	6	Türkiye
G49	Egebeyi	CEN-B/2*CA-I92//VIRINGA/3/ATACO/4/ Harma-02//11012-2/Cm67/3/Market semple Marageh /5/ROHADES//TB//CHZO/ 3/GL/COPAL/3/BAR/RHODES//GL/COME	6	Türkiye
G50	Bolayır	Osk 4.197/12-84//HB854/Astrix/3/Alpha/Durra	2	Türkiye
G51	Harman	Unknown	2	Türkiye
G52	Sladoran	Introduced from Yugoslavia	2	Türkiye
G53	Hazar	Osk4.39/2-84//Barbe-Rousse	6	Türkiye
G54	Hasat	Rod/Scala	2	Türkiye
G55	Martı	Flam/WM/5/Yky387/3/Api/Cm67//Manc/4/Yrm/Lhfm	6	Türkiye
G56	Çıldır 02	3896/28//284/28/3/Cum- 50/4/624/682/5/WBQT12	2	Türkiye
G57	Erginel-90	Escourgeon Hop 2171 (France)	6	Türkiye
G58	Keser	Unknown	2	Türkiye

Table 1. Cont.

Code	Cultivar	Pedigree	Spike Type (Row)	Origin
G59	Yerçil 147	Strengs Frankengerste from Germany	2	Türkiye
G60	Hamidiye 85	Tokak mutant 173 TH/Tokak	2	Türkiye
G61	Rihane-03	As46//Avt/Aths	6	Syria
G62	Yerli siyah	Landrace	2	Türkiye
G63	Pamir-009	Unknown	6	Pakistan
G64	Ünver	YEA389-3/YEA475-4//97-98DH8	2	Türkiye
G65	Sahra	LB.Iran/Una8271//Gloria"s"/Com"s"	2	Iranian
G66	Khatem	LB Iran /Una8271//Gloria"s"/Com"s"/3/Kavir	2	Iranian
G67	Jolgeh	Makoe//Zarjow/80-5151	2	Iranian
G68	Nik	Lignee 527/NK1272//JLB70-63	2	Iranian
G69	Bahman	WA 2196-68/NY6005-18, F1//Scotia I	2	Iranian
G70	Behrokh	Novosadski-444	2	Iranian
G71	Goharan	Rhn-03//L.527/NK1272	2	Iranian
G72	Tokak 157/37	Selection from Landraces	2	Türkiye
G73	Arcanda	Unknown	2	Austria
G74	Finola	Unknown	6	Austria
G75	Alena	Unknown	2	Austria

Duzce province is in the Western Black Sea Region and has a typical Black Sea climate. Additionally, the region exhibits a transitional climate and has the characteristics of both Mediterranean and Continental climates (Figure S1). The average temperature and precipitation during the experimental years (2021 and 2022) and the long-term averages over time are presented in Figure 1. The experimental area was characterized by clay soil with a slightly acidic pH. The soil was found to be salt-free with shallow lime content. The soil exhibited limited phosphorus availability for plants, low potassium levels, and high nitrogen and organic matter content (Table 2).

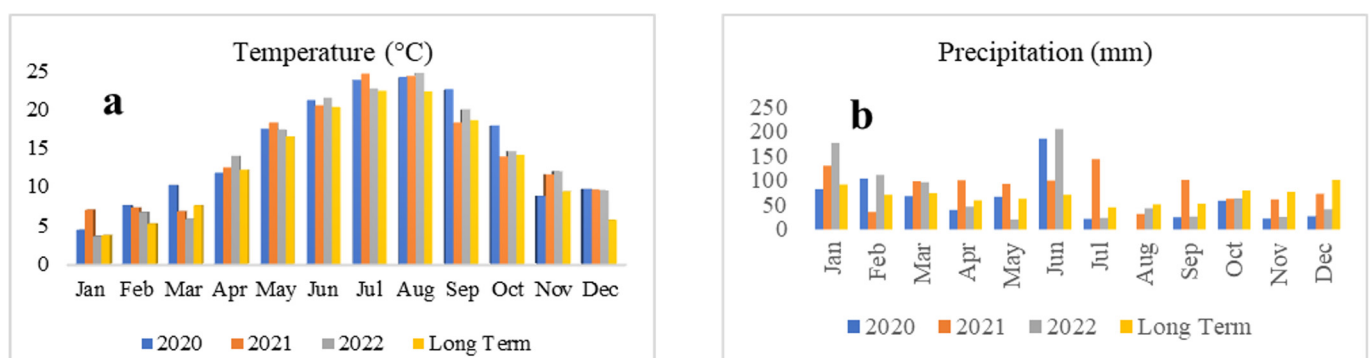


Figure 1. The average climate data for the experimental years; (a) Temperature; (b) Precipitation.

Table 2. The soil characteristics of the experimental field.

Texture	pH	EC (dS m ⁻¹)	Lime (%)	Organic Matter (%)	Total N	P	K
Clay (86)	6.41	0.033	0.000	4.874	0.244	0.57	26

The experiment was carried out in a randomized complete block design (RCBD) with four replications. The sowing density was 450 seeds per m². The plots were five meters long

and contained six rows 20 cm apart ($5 \text{ m} \times 1.2 \text{ m} = 6 \text{ m}^2$). Sowing was performed in the first week of November in both years. Then, 50 kg ha^{-1} N and P_2O_5 fertilizers were applied during planting. As a top dressing, 90 kg ha^{-1} of N fertilizer was applied at tillering and 60 kg ha^{-1} of N fertilizer was applied at joining. Weed management was conducted manually and there was no treatment for pest control. The harvest was conducted in the first week of July for both cropping seasons.

2.2. Investigated Traits

The heading date (HD) was recorded when approximately 50% of heads had fully emerged. Ten plants were selected randomly from each plot to record agronomic traits like plant height (PH), spike length (SL), grain number per spike (NGS), grain weight per spike (GW), thousand-kernel weight (1000 KW), and test weight (TW). The total harvested area was 6 m^2 ($5 \times 1.2 \text{ m}$). Grain yield (GY) was determined by weighing the plot yields and was converted to yield per hectare (kg ha^{-1}).

2.3. Statistical Analysis

Variance analysis was performed for each year and combined data and comparison of the means were conducted using the Duncan method. Pearson correlation analysis was utilized to examine the relationship between the traits. Cluster analysis was used to uncover similarities or differences among groups within a population and to demonstrate taxonomic relationships among populations [24]. The genotype by trait interaction was determined using the method proposed by Yan [25], as outlined in the equation below, as follows:

$$\frac{\alpha_{ij} - \beta_j}{\sigma_j} = \sum_{n=1}^2 \lambda_n \xi_{in}^* \eta_{jn}^* + \varepsilon_{ij} = \sum_{n=1}^2 \xi_{in}^* \eta_{jn}^* + \varepsilon_{ij}$$

where α_{ij} represents the average amount of genotype i for every trait j , β_j denotes the average amount of all genotypes for the traits, σ_j indicates the standard deviation of trait j in the average genotypes, ε_{ij} represents the amount of genotype i remaining in trait j , λ_n signifies a certain amount for the main element (PCn), ξ_i denotes the amount of PCn for genotype i , and η_{jn} represents the amount of PCn for genotype j . We standardized the traits and eliminated the influence of different traits. Subsequently, a biplot method was constructed for all the scored traits for each genotype using the Genstat 14th (Copyright 2011, VSN International Ltd.) release software program.

3. Results

According to the variance analysis, significant differences were observed between years for all studied traits. Statistically significant differences were noted in all examined traits for both genotype and genotype $\times$ year interactions at the $p \leq 0.01$ level (Table 3). The data were graphically analyzed and interpreted using GT biplot software. Six GT biplot diagrams (Figures 2–5, S2 and S3) were used in this study to rank and group genotypes based on traits, evaluate genotype stability, determine ideal genotypes, and classify genotypes according to investigated traits. The cluster is presented in Figure 6.

Table 3. Variance analysis for the investigated traits.

Source of Variance	DF	GY	HD	PH	SL	NGS	GW	TW	1000 KW
Year (Y)	1	107,382.5 **	17.3 **	4261.3 **	1.6 *	860.4 **	0.33 *	16.006 **	2986.2 **
Genotype (G)	74	258,276.06 **	236.4 **	569.7 **	17.4 **	1430.7 **	2.33 **	159.5 **	210.6 **
Y $\times$ G	74	30,721 **	2.36 **	255.1 **	2.32 **	57.8 **	0.27 **	17.27 **	85.3 **
CV %	-	7.57	0.41	3.60	8.06	10.20	14.08	0.88	3.60
R ²	-	0.95	0.99	0.90	0.87	0.94	0.88	0.98	0.96

*, **: significant at the %5 and %1 level. GY: Grain yield, HD: Heading date, PH: Plant Height, SL: Spike length, NGS: Grain number per spike, GW: Grain weight per spike, TW: Test weight, 1000 KW: Thousand kernel weight.

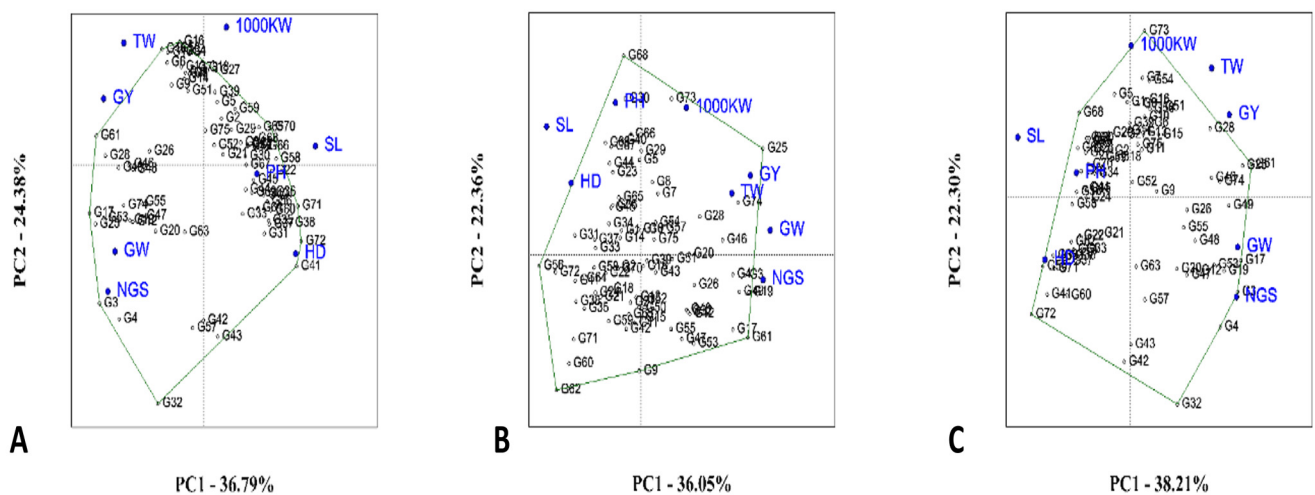


Figure 2. Polygon view of the GT biplot of barley genotypes; (A) First year; (B) Second year; (C) Average of two years.

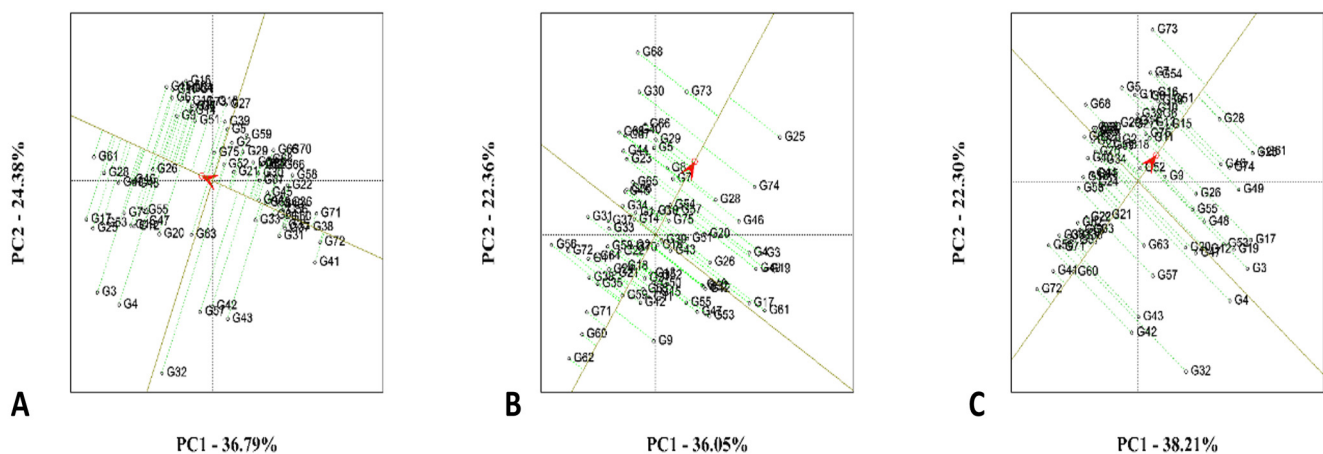


Figure 3. Ranking of barley genotypes based on stability in the (A) first year, (B) second year, and (C) average of two years.

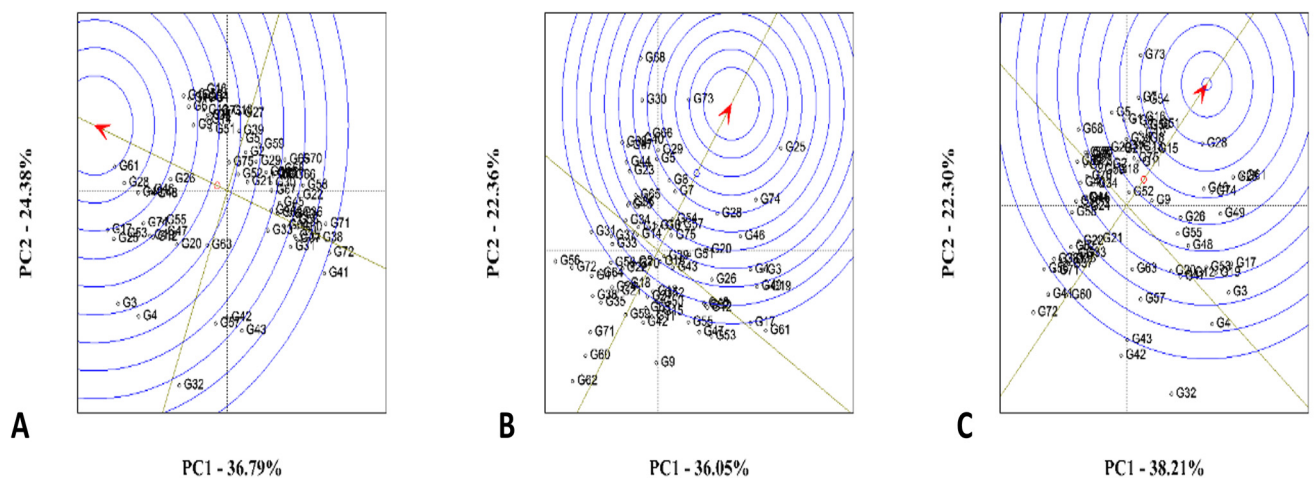


Figure 4. Ranking of barley genotypes based on ideal genotype in the (A) first year, (B) second year, and (C) average of two years.

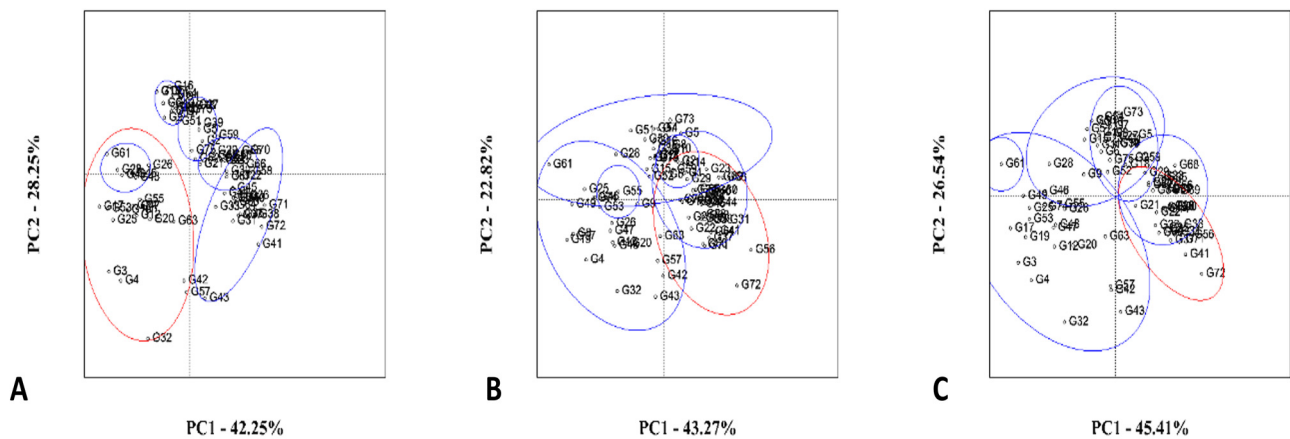


Figure 5. Grouping of barley genotypes based on traits in the (A) first year; (B) second year; and (C) average of two years.

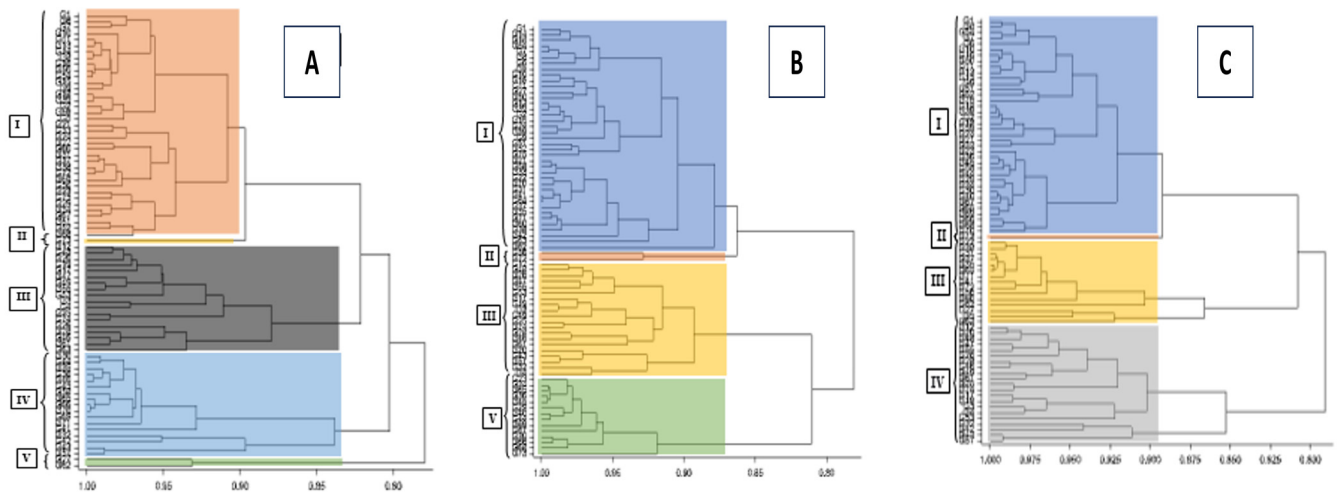


Figure 6. Cluster describing variation among barley genotypes in the (A) first year, (B) second year, and (C) average of two years.

3.1. Agro-Morphological Evaluation

The mean performance of different barley genotypes demonstrated considerable variability across various traits, with recorded ranges in the first year as follows: HD (109.0–126.0 days), PH (88.25–126.50 cm), SL (4.05–11.70 cm), NGS (20.25–76.50), GW (0.825–3.278 g), 1000 KW (26.55–53.42 g), TW (51.52–72.77 kg hL⁻¹), and GY (2797.00–10,858.20 kg ha⁻¹); in the second year: HD (105.75–126.00 days), PH (91.50–139.50 cm), SL (4.85–11.15 cm), NGS (15.00–67.50), GW (0.812–2.932 g), 1000 KW (25.55–56.45 g), TW (50.52–71.52 kg hL⁻¹), and GY (1703.50–11,274.50 kg ha⁻¹); and based on the averages over both years: HD (107.37–126.00 days), PH (89.87–129.00 cm), SL (5.08–10.90 cm), NGS (17.62–70.12), GW (0.908–3.105 g), 1000 KW (30.56–53.05 g), TW (52.44–71.31 kg hL⁻¹), and GY (2659.87–11,066.50 kg ha⁻¹), as presented in Table S1.

The earliest heading was found in genotypes G44 (126.0 days), G45 (125.0 days), and G36 (125.0 days), while the latest heading was observed in genotypes G61 (107.4 days), G28 (109.0 days), and G27 (125.0 days). The highest PH was found in genotypes G40 (129.0 cm), G30 (127.4 cm), G72 (123.9 cm), and G44 (123.5 cm), while the lowest PH was recorded in genotypes G60 (89.9 cm), G53 (93.2 cm), G52 (95.2 cm), and G62 (95.5 cm). The genotypes with the highest SL values were G69 (10.90 cm), G56 (10.71 cm), and G68 (10.50 cm), while those with the lowest SL values included G19 (5.09 cm), G17 (5.11 cm), G61 (5.16 cm), and G4 (5.19 cm). Genotypes G32 (70.12), G3 (67.81), and G25 (64.62) had the highest NGS, while genotypes G62 (17.62), G27 (23.50), and G10 (23.56) showed the

lowest NGS. The highest GW was obtained in genotypes G25 (3.11 g), G74 (2.82 g), and G57 (2.81 g), while the lowest GW was found in genotypes G62 (0.90 g), G21 (0.99), and G24 (1.02 g). In terms of 1000 KW, G68 (53.05 g), G73 (52.82 g), and G66 (50.28 g) showed the highest values, whereas the lowest values were seen in G42 (30.56 g), G53 (30.90 g), G12 (31.45), and G41 (31.72 g). Genotypes G15 (71.31 kg hl⁻¹), G73 (70.75 kg hl⁻¹), and G11 (70.10 kg hl⁻¹) showed the highest TW, while genotypes G62 (55.01 kg hl⁻¹), G72 (54.46 kg hl⁻¹), and G56 (52.43 kg hl⁻¹) had the lowest TW. Regarding GY, the genotypes G25 (10,858.25 kg ha⁻¹, 11,274.75 kg ha⁻¹, 11,066.50 kg ha⁻¹), G73 (10,833.50 kg ha⁻¹, 10,078.00 kg ha⁻¹, 10,455.75 kg ha⁻¹), and G74 (10,459.25 kg ha⁻¹, 10,406.75 kg ha⁻¹, 10,412.75 kg ha⁻¹) exhibited superior grain yields in the first and second years, as well as in the overall mean, respectively. In contrast, the genotypes G27 (3100.25 kg ha⁻¹), G71 (2797.00 kg ha⁻¹), and G72 (3181.50 kg ha⁻¹) recorded the lowest yields in the first year, and G62 (1703.50 kg ha⁻¹), G63 (3019.50 kg ha⁻¹), and G72 (2138.25 kg ha⁻¹) in the second year, with the overall averages showing that G72 (2659.88 kg ha⁻¹), G62 (2853.00 kg ha⁻¹), and G71 (3161.13 kg ha⁻¹) had the lowest grain yields across both years (Table S1).

3.2. GT Biplot Model

The GT biplot generated from the GT data shows that the total variation among the traits was 61.17% in the first year, 58.41% in the second year, and 60.51% for the average over two years (Figures 2–5). The information about the “which won where/what” pattern of genotypes based on traits is conveyed in Figure 2. In the first year of the trial, GY, TW, and 1000 KW were grouped together, GW and GNS formed another group, and HD, PH, and SL constituted a separate group. In the second year of the trial, GY, TW, GW, and NGS formed one group; HD, PH, and SL formed another group; and 1000 KW comprised a separate third group. For the two-year average, GY, 1000 KW, and GW were in one group; NGS and GW were in another group; and HD, PH, and SL were in a third group. During the first year, notable genotypes included G3 and G4 for NGS; G65 and G70 for SL; G16 for 1000 KW; and G41 for HD. In the second year, the G25 genotype stood out for GY and GW; G73 for 1000 KW; and G3, G4, and G19 for NGS. For the two-year average, the G25 and G61 genotypes stood out for GY and TW, while the G3 genotype stood out for NGS and GW. The G68 genotype stood out for HD, PH, and SL. Even though the G32 genotype is situated in the diagonal part of the polygon, the absence of any traits in its section suggests it lacks ideal or high values, as shown in Figure 2.

In Figure S2, the rankings of barley genotypes are visualized based on their average performance across all traits. In the first year of the study, genotypes G61, G28, G17, and G25 exhibited the highest performance while G41, G72, and G38 genotypes showed the lowest performance for the examined traits. During the second year of the trial, genotypes G25, G78, and G68 displayed the highest performance whereas G62, G60, and G71 had the lowest performance across traits. Over the two-year average, genotypes G73, G61, G28, and G25 had the highest performance while G72, G41, and G60 had the lowest performance for the pursuant traits.

Figure 3 visualizes the stability of the genotypes according to the various traits. During the initial year of the study, the G26, G61, G28, G46, G48, and G30 genotypes were determined to be the most stable genotypes and were above average. The G32, G57, G42, and G43 genotypes were determined to be unstable and below-average. Although the G26 genotype was more stable than the G61 genotype, the G61 genotype performed higher than the G26 genotype. During the second year of the trial, among the genotypes showing above-average performance, G54, G75, G7, G8, G5, and G57 emerged as the most stable, while G25, G73, and G68 demonstrated the highest performance. Conversely, G61, G17, G53, and G19 were identified as the most unstable genotypes, with G62, G60, and G71 exhibiting the lowest performance. For the average over two years of the experiment, the G73, G61, G28, G25, and G54 genotypes had the highest performance. However, the G52, G15, G11, G13, and G51 genotypes were the most stable. The G32, G4, G3, G42, G17, and

G19 genotypes were the most unstable. The G72, G41, G60, and G42 were the genotypes with the lowest performance.

The GT biplot comparison ranks genotypes according to the hypothetical (ideal) genotype (Figure 4). The genotypes found in the first concentric circle were considered ideal genotypes. When examining the comparison biplot graph representing the genotypes' status in the first year, the G61 and G28 genotypes were the closest to the center and represented the ideal genotype. The G72 and G41 genotypes were the farthest away. In the second year, the G73 and G25 genotypes were the closest to the ideal center, while the G62 and G60 genotypes were the farthest away. Looking at the average over the two years, the G28 and G54 genotypes were the closest to the ideal center, while G72 and G41 were the farthest away (Figure 4).

A correlation diagram analysis was conducted to investigate the correlation between the traits in this research (Figure S3). When analyzing the interrelationships among traits in the first year of the study, the angles between vectors representing GY, TW, and 1000 KW were less than 90 degrees. This signified a positive correlation among these traits and was determined among PH, SL, and HD. Additionally, there was a high correlation between NGS and GW traits. There was also a negative correlation between GY, HD, and PH. In the second cropping season, a positive correlation was observed between HD and SL as well as between HD and PH. GY exhibited positive correlations with TW, GW, NGS, and 1000 KW. For the two-year average, a strong correlation was observed among PH, SL, and HD. There was a high correlation between GY, TW, and 1000 KW and a high positive correlation between GW and NGS (Figure S3).

The grouping of genotypes according to traits is illustrated in Figure 5. The study divided genotypes into six groups during the first growing season, the second growing season, and the average across two growing seasons. During the first year of the investigation, the genotypes G61, G28, and G26 stood out in terms of stability and as the ideal genotypes. It is worth noting that G61, G28, and G26 were in the same group. The genotypes G72, G71, G38, and G41, which were undesirable in terms of stability and were considered not an ideal genotype, were also in the same group. During the second year of the investigation, the genotypes G73, G25, G66, G68, and G9, which were desirable in terms of stability and represented an ideal genotype, were grouped in the same cluster. The genotypes G60, G61, and G72, which had low performance, were grouped in the same cluster. G55, G46, and G74 were grouped into three clusters. For the two-year average, the group containing only G61 was found within a large cluster containing the genotypes G25, G46, and G74. Genotypes G41, G60, and G72 were in the same group and were considered undesirable genotypes for the examined characteristics.

3.3. Cluster Analysis

The dendrogram illustrating the clustering of barley genotypes utilized in the study is presented in Figure 6, while the grouping of barley genotypes according to cluster analysis is detailed in Table 4. In the first year of the study, five groups were formed: Group 1 comprised 37 genotypes, Group 2 consisted of one genotype, Group 3 had 18 genotypes, Group 4 included 17 genotypes, and Group 5 comprised two genotypes. In the second year of the study, four clusters were formed: Group 1 comprised 39 genotypes, Group 2 had two genotypes, Group 3 contained 20 genotypes, and Group 4 comprised 14 genotypes. For the two-year average, four clusters were formed: Group 1 consisted of 38 genotypes, Group 2 had 1 genotype, Group 3 contained 15 genotypes, and Group 4 comprised 21 genotypes (Figure 6; Table 4).

Table 4. The groups of barley genotypes according to cluster analysis.

Year	Group	Genotypes
First Year	I	G1, G2, G5, G6, G7, G8, G9, G10, G11, G13, G14, G15, G16, G18, G21, G22, G23, G24, G29, G31, G33, G34, G37, G38, G39, G50, G51, G52, G54, G56, G59, G60, G64, G67, G68, G72, G75
	II	G73
	III	G3, G4, G12, G17, G19, G20, G25, G26, G28, G46, G47, G48, G49, G53, G55, G61, G63, G74
	IV	G30, G32, G35, G36, G40, G41, G42, G43, G44, G45, G57, G58, G65, G66, G69, G70, G71
	V	G27, G62
Second Year	I	G1, G2, G5, G6, G7, G8, G9, G10, G11, G13, G14, G15, G16, G18, G21, G22, G24, G27, G31, G33, G35, G37, G38, G39, G41, G42, G50, G51, G52, G54, G58, G59, G60, G62, G63, G64, G70, G71, G75
	II	G56, G72
	III	G3, G4, G12, G17, G19, G20, G25, G26, G28, G32, G43, G46, G47, G48, G49, G53, G55, G57, G61, G74
	IV	G23, G29, G30, G34, G36, G40, G44, G45, G65, G66, G67, G68, G69, G73
Average of Two Year	I	G1, G2, G6, G7, G8, G9, G10, G11, G13, G14, G15, G16, G18, G21, G23, G24, G29, G30, G33, G34, G36, G39, G40, G44, G45, G50, G51, G52, G54, G58, G59, G65, G66, G67, G68, G69, G70, G75
	II	G73
	III	G5, G22, G27, G31, G35, G37, G38, G41, G56, G60, G62, G63, G64, G71, G72
	IV	G3, G4, G12, G17, G19, G20, G25, G26, G28, G32, G42, G43, G46, G47, G48, G49, G53, G55, G57, G61, G74

3.4. Pearson's Correlation

Pearson's correlation analysis is presented in Table 5. It reveals notable negative correlations between GY and both HD and SL. GY displayed an insignificant negative association with PH. Conversely, there were significant positive correlations observed between GY and NGS, GW, and TW. A significant positive correlation was noted between GY and 1000 KW. Significant positive correlations were also found between HD and PH and between HD and SL. HD had non-significant and negative correlations with NGS, GW, and 1000 KW. A noteworthy inverse relationship was observed between HD and TW. PH showed significant positive correlations with SL, as well as positive correlations with GW and 1000 KW. PH had non-significant negative relationships with both NGS and TW. SL had significant negative correlations with both NGS and GW. A significant negative correlation existed between SL and TW, while there was a significant positive correlation between SL and 1000 KW. A significant and positive correlation was observed between NGS and GW along with a significant negative correlation between NGS and 1000 KW and a positive but non-significant correlation between NGS and TW. The correlation between GW and 1000 KW, as well as TW, was positive but not statistically significant. There was a notable positive correlation between 1000 KW and TW.

Table 5. Pearson's correlation coefficient between traits of 75 barley genotypes.

Traits	GY	HD	PH	SL	NGS	GW	1000 KW
HD	−0.3131 **						
PH	−0.1107	0.3893 **					
SL	−0.2959 *	0.4673 **	0.51 **				
NGS	0.3425 **	−0.1294	−0.0844	−0.5865 **			
GW	0.4411 **	−0.1804	0.0502	−0.4700 **	0.9082 **		
1000 KW	0.2396 *	−0.1608	0.1645	0.2655 *	−0.2725 *	0.0419	
TW	0.6631 **	−0.4612 **	−0.0669	−0.2739 *	0.0671	0.1904	0.3395 **

*, **: significant at the %5 and %1 level, respectively.

4. Discussion

Grain yield is a complex quantitative trait influenced by a variety of genetic and environmental factors [26]. This emphasizes the necessity of conducting a comprehensive evaluation of its interactions with yield-related traits and agronomic characteristics to enhance the effectiveness of plant selection in breeding programs [27]. The average values

for the studied traits across two cropping seasons involving 75 barley genotypes are presented in Table S1. A significant variation was observed among the analyzed traits across the study. Genotype G25 exhibited superior performance, achieving the highest grain weight and grain yield, positioning it as a leading candidate for yield-related traits. Additionally, genotypes G73 and G74 were among the top performers in average grain yields over the two-year period, further emphasizing their high yield potential (Table S1).

The genotype by trait biplot (GT Biplot), a component of the GGE biplot methodology, serves as a valuable instrument for analyzing datasets that encompass multiple traits and is helpful for identifying genotypes relative to these traits. It provides a visual representation of the correlations between traits across genotypes and highlights the trait profiles of the evaluated genotypes [23]. The GT biplot derived from the GT data shows that there was 61.17% total variation among the traits in the first cropping season, 58.41% in the second cropping season, and 60.51% on average across the two cropping seasons (Figures 2–6). Dyulgerov and Dyulgerova [28] reported the total variation in the GT biplot analysis as 73.86%, while Ebadi-Segherloo et al. [29] reported it as 55%. The polygon view of the biplot aids in identifying genotypes that exhibit the highest values for one or more traits. Genotypes positioned at the vertices of the biplot can be distinguished by their specific traits, while those located near the origin are considered to possess a broader range of traits [30]. The biplot presented as a polygon view illustrates data from 75 barley genotypes across eight different traits (Figure 2). In Figure 2, the traits were grouped as follows: HD, PH, and SL; NGS and GW; and 1000 KW, TW, and GY. Genotypes G25 and G61 exhibited superior performance in GY and TW, while G3 and G4 demonstrated notable characteristics for NGS. Genotype G73 showed outstanding performance in 1000 KW. The polygon view of the GT biplot model effectively facilitated the identification of top-performing genotypes across multiple traits, offering a detailed visualization of the interactions between traits and genotypes [31].

The study employed the Average Tester Coordination (ATC) view to evaluate genotypes based on GT data, assessing overall superiority and stability across all traits (Figures 2 and 3). Genotypes were compared relative to the horizontal stability axis and vertical mean axis established from average values. Genotypes above the vertical axis were considered preferable, while those below were less favorable. Stability was determined by proximity to the center of the horizontal line, with deviations indicating increasing instability [20]. In the first cropping season, G61, G28, G17, and G25 showed the highest performance, and G26, G61, G28, G46, G48, and G30 were the most stable. In the second season, G25, G78, and G68 had the highest performance, while G54, G75, G7, G8, G5, and G57 were the most stable. Across both seasons, G73, G61, G28, and G25 demonstrated the highest performance, with G52, G15, G11, G13, and G51 being the most stable. These findings align with previous studies by Tsige [32] and Mariey et al. [22], which also used this method to identify genotypes with superior stability and performance.

The ideal genotype is positioned within the inner circle and closest to the center arrowhead of the biplot, making these genotypes the most preferred [33]. The comparison biplot revealed that genotypes G61 and G28 were nearest to the ideal in the first year, while G73 and G25 were closest in the second year. Over the two-year average, genotypes G28 and G54 demonstrated the closest proximity to the ideal genotype (Figure 4). Similar studies comparing genotypes with the ideal have been reported in various crops, including barley [22], sweet corn [23], wheat [34], and sunflower [35].

The Pearson correlation analysis (Table 5) revealed significant negative correlations between GY and HD, as well as between GY and SL. While the correlation between GY and PH was not significant, significant positive correlations were observed between GY and NGS, GW, TW, and 1000 KW. The GT biplot provided additional insights into these relationships, helping to identify key traits for improving grain yield in breeding programs. In the biplot, the cosine of the angle between trait vectors represents the correlation coefficient (Figure S3), where acute angles indicate positive correlations and obtuse angles indicate negative correlations. The analysis of Figure S3 showed a consistent

positive correlation between GY and TW across both experimental years and the two-year average. A positive correlation was also observed among HD, PH, and SL, as well as between NGS and GW. Conversely, GY exhibited a negative correlation with HD, PH, and SL, which corroborated the Pearson correlation results. Naser et al. [36] reported a significant positive relationship between GY and 1000 KW, as well as between NGS and GY. Dyulgerov and Dyulgerova [37] identified a negative correlation between GY and HD, along with significant positive correlations between GY and NGS, GW, and 1000 KW. These results are consistent with previous studies that have reported similar trait relationships.

The genotype grouping diagram enables the evaluation of genotypes based on their stability and yield across multiple traits, organizing them into groups accordingly [38]. In the first growing season, genotypes G61, G28, and G26 stood out for their stability and were considered ideal, forming the same group. In the second season, genotypes G73, G25, G66, G68, and G9, also desirable for their stability, were grouped together as ideal genotypes. For the two-year average, G61 was initially grouped alone but later included in a larger group with genotypes G25, G46, and G74 (Figure 5). Shojei et al. [38] and Stansluos et al. [23] also utilized grouping diagrams to evaluate genotypes based on their performance across various traits.

Cluster analysis, a multivariate analysis, is extensively used to characterize genetic diversity by assessing genotype similarities or differences [17]. The resulting cluster of the barley genotypes in this study is presented in Figure 6 and Table 4. The analysis revealed five distinct groups in the first year, four in the second year, and four groups on average across the two-year period. The first group, containing the largest number of genotypes, was primarily composed of barley genotypes from Bulgaria, while genotypes of Iranian origin were closely related to those from Turkey. In both the first year and the two-year average, G73 formed a distinct group. High-yielding genotypes such as G25, G28, G61, and G74 consistently clustered together, with G25 and G74 exhibiting a particularly close relationship. Breeding for high-yield varieties often reduces genetic diversity, potentially altering gene frequencies within plant populations [39]. The success of crop improvement strategies depends on the effective identification and integration of genetic diversity from various sources, including existing cultivars, landraces, wild relatives, and germplasm collections containing elite and mutant lines [40]. Cluster analysis elucidated the relationships among barley genotypes from six different regions, revealing that high-yielding genotypes consistently formed distinct clusters.

5. Conclusions

Breeding programs aim to identify superior genotypes and develop high-yield high-quality cultivars to ensure sustainable production. In this study, the GT biplot technique, derived from the GGE biplot method, was applied to select appropriate barley genotypes. An analysis of data from each growing season and the two-year average revealed that genotype G25 exhibited the highest yield among all the genotypes studied. The relationships between traits identified through GT biplot analysis were consistent with Pearson's correlation coefficients, showing positive correlations between GY and NGS, GW, 1000 KW, and TW. Moreover, GT biplot analysis identified G25, G28, G61, G73, and G74 as promising genotypes for high-yield barley, offering valuable genetic resources for future breeding efforts. The results demonstrated that GT biplot analysis effectively represented genotype-trait relationships, enabled comparisons, and revealed interconnections among various traits and genotypes. These findings provide important insights that can guide future barley breeding initiatives.

Supplementary Materials: The following supporting information can be downloaded at <https://www.mdpi.com/article/10.3390/agronomy14102188/s1>: Table S1: Mean values of yield, yield components, and quality traits across 75 barley genotypes; Figure S1: Turkey's regional climatic differences; Figure S2: Ranking of barley genotypes based on traits in the (A) first year, (B) second year, and (C) average of two years; Figure S3: Analysis of correlations between traits in the (A) first year, (B) second year, and (C) average of two years.

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