



Herbage Quality of Eight Native *Hordeum* Ecotypes Collected From Natural Grassland & Pasture Ecology of Southeastern Anatolia

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Abstract: In this study, plant samples of eight different ecotypes of three distinct species of the genus *Hordeum* were collected at the anthesis stage of the plants in the 2023 spring in Southeastern Anatolia's natural grassland and pasture ecology. The quality analyses of the herbage samples of *Hordeum bulbosum*, *H. murinum*, and *H. spontaneum* ecotypes collected from five different locations (Karacadağ-I, Batman-1, Diyarbakır-6, Diyarbakır-8, and Diyarbakır-13) were determined by NIRS analyzer. Crude protein (CP), dry matter (DM), acid detergent insoluble fiber (ADF), neutral detergent insoluble fiber (NDF), acid detergent insoluble protein (ADP), Ca, K, Mg, P, Ca/P, K/(Ca+Mg), digestible dry matter (DDM), dry matter intake (DMI) and relative feed values (RFV) were determined. The values were determined between 8.2-23.4% for CP; 92.1-93.4% for DM; 19.3-36.2% for ADF; 26.2-71.9% for NDF; 0.16-0.71% for ADP; 60.7-73.9% for DDM; 1.67-4.58% for DMI; 78.6-262.8 for RFV; 0.30-0.42% for P; 1.72-2.84% for K; 0.12-1.62% for Ca; 0.17-0.30% for Mg; 0.33-3.84 for Ca/P; 2.76-4.77 for K/(Ca+Mg). In conclusion, the CP, ADF, NDF, DMI, RFV, and Ca/P values were found very variable in collected ecotypes and can be used for forage barley breeding purposes.

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1. Introduction

An environment that is both demanding and unpredictable poses a threat to global food security. An effective and sustainable approach to enhancing food productivity stability is through selecting crops to exhibit high resilience to abiotic stressors. The use of the gene pools of the agricultural plant's wild relatives is a commonly used strategy for improving crop genetics (Chen et al., 2008).

One of the most important cereals in the world and one of the earliest domesticated crops is barley (*Hordeum vulgare* subsp. *vulgare*). The wild progenitor of barley, *H. vulgare* subsp. *spontaneum*, is considered as a significant source for barley improvement in response to changing climatic conditions. Both species are interfertile, primarily self-pollinating, and diploid ($2n = 14$). There are various phenotypic differences between cultivated and wild barley, which are combined to form the domestication syndrome. From the coasts of the eastern Mediterranean to the semideserts of Afghanistan, wild barley grows naturally in Southwest Asia (Jakob et al., 2014). *H. spontaneum*, the

parent plant of cultivated barley, is a self-fertilizing annual grass that is primarily found in the Mediterranean and Irano-Turanian regions. It can also be found in desert regions, where it can sustain steady populations. There are colder climates where wild barley grows, like in Tibet. Rich genetic diversities for drought, salt, and cold resistances have been collected in wild barley as a result of its adaption to both cold and desert environments (Chen et al., 2008).

Since many of the taxa that make up the *Hordeum*, such as the majority of tetra- and hexaploid species, are of hybrid origin, it is not simple to determine the relationships between its members. Within the *Triticeae* family, *Hordeum* is one of the largest genera, containing over 30 species distributed worldwide. These two species do not appear to be related, based on the observed dramatic variations in the number and distribution of the repeat sequences that were analyzed in *H. vulgare* and *H. bulbosum*. Conversely, no distinctions were found between *H. vulgare*'s two subspecies (subsp. *vulgare* and subsp. *spontanum*). Concerning the *H. murinum* complex, the results support the idea that *H. glaucum* and two extinct species were respectively the diploid donors of the subgenomes Xu, Xv, and Xw present in polyploids (Jouve et al., 2018).

Particularly in colder climates where other feed grain crops like wheat, sorghum, and maize are difficult to grow, barley is an extremely important feed grain (Poulsen, 2020). One of the few essential crops that were domesticated during the beginning of agriculture in the Fertile Crescent was barley, and it is still widely used in farming today in the region. According to Russell et al. (2011), barley serves as a model species for research on the evolution, adaptability, and dissemination of major crops worldwide. In the Fertile Crescent, the taxon makes up a significant annual component of the open herbaceous and park-like vegetation. It is mostly limited to artificial (secondary) habitats outside of this area, and only widely dispersed populations are found there, especially in the east of its distribution area. Extensive serrated lemma awns and the ear's ability to break apart after ripening, releasing spikelets resembling arrows, offer superior adaptability to epizoochory and colonization. Uncertainty surrounds whether wild barley populations in Morocco, Ethiopia, and Tibet are naturally occurring, human-introduced, or wild variations of farmed barley (Molina-Cano et al., 2005). The spike structure separates the two-rowed and six-rowed barley. Hulless barley is defined as having neither the palea nor the lemma attached to the seed. Compared to hulled barley, naked barley yields less and has a higher protein content. While naked barley is an old food crop, most farmed varieties of barley are hulled. By manipulating inflorescence, there is a great potential to increase its yields (Li, 2020).

Barley, both wild and cultivated, has been collected across its range in the past century, and seed samples have been preserved and kept in ex-situ gene banks. Approximately 470 000 barley accessions are kept in more than 200 collections across the globe (Knüpffer, 2009). According to Russell et al. (2011), this stock of accessions serves as the primary source of plant materials used in investigations into many facets of genetic variation in barley.

Annual forages are well suited to semiarid climates. In dryland systems, annual cereal forages are more resilient than grain crops in terms of water use (WU), water use efficiency (WUE), and weed control. Many types of fodder can survive in semiarid conditions, including warm- and cool-season grasses as well as mixes of legumes and grasses. Dryland cropping systems can be effectively diversified by using any of these annual forages (Lenssen et al., 2015).

For livestock producers in ecologies similar to Montana (USA), barley cut for hay is a major source of winter feed. Barley is a widely accessible and reasonably priced feed ingredient. As it grows and is harvested similar to legume forages, forage barley yields well and is beneficial to producers. Feed value can be maximized by careful harvesting. Compared to other minor grains, barley has been found to have a lower fiber concentration and a higher nutritional value. Awns on some barley cultivars, nevertheless, are harsh when chopped for hay. Cereal grains with rough or barbed awns can irritate the mouth and reduce palatability (Todd et al., 2003). Despite the significance of fodder quality, forage barley breeding programs often choose new barley lines primarily on yield and awnless features (Surber et al., 2004). The barley feed's palatability may be impacted by the awns. The forage's ingestion potential is inversely connected with neutral detergent fiber. Acid detergent fiber is a measure of the portion of fiber that is less digestible and has a negative correlation with the animal's potential for digestion. Barley hay is usually fed in the winter and spring, when the animals' protein needs may be at their peak, hence its nitrogen concentration is crucial. Particular varieties of hay barley are more likely to accumulate nitrates, which can make them harmful to cattle that consume them. It would be preferable if the nitrate accumulation potential was low. Thus, it would be preferable to have higher CP and digestibility and

reduced accumulation of NDF, ADF, and NO₃-N. Although measuring these traits is not difficult, breeding programs do not employ them as selection criteria (Surber et al., 2001).

According to Liancourt et al. (2013), an ecotype is a population (or subspecies, or race) that has adapted to the environmental conditions of its particular location and is distinguished by particular physiological and morphological traits. The response of species to the environment is "ecotype-specific". Climate change effects may be mitigated or obscured by prevailing sources of variance affecting plant performance which exist in ecotypes.

In this study, plant samples of eight different ecotypes of three distinct species of the genus *Hordeum* were collected at the anthesis stage of the plants in Southeastern Anatolia's natural grassland and pasture ecology. The quality analyses of the herbage samples of *Hordeum bulbosum*, *H. murinum*, and *H. spontaneum* ecotypes collected from five different locations (Karacadag, Batman, Diyarbakır-1, Diyarbakır-2, and Diyarbakır-3) were determined in the study.

2. Material and Methods

Plant samples of eight ecotypes from three distinct species of the genus *Hordeum* constitute the research material. The samples were obtained in 2023 from various sites (Karacadag, Batman, Diyarbakır-1, Diyarbakır-2, and Diyarbakır-3 locations) in the Southeastern Anatolia Region of Türkiye. Table 1 provides information on the locations, dates, and geographic coordinates of the collected plants.

Tablo 1. Species, locations, collection dates, and geographic coordinates of the plant samples

Species	Locations	Latitude	Longitude	Altitude (m)	Date
<i>H. bulbosum</i>	Karacadag	37.77°	39.78°	1469	21.05.2023
<i>H. bulbosum</i>	Diyarbakır-1	37.91°	40.27°	652	15.05.2023
<i>H. bulbosum</i>	Diyarbakır-2	38.21°	39.27°	1113	10.05.2023
<i>H. murinum</i>	Diyarbakır-2	37.91°	40.27°	652	15.05.2023
<i>H. murinum</i> ssp. <i>leporinum</i>	Batman	37.95°	41.36°	572	07.05.2023
<i>H. spontaneum</i>	Batman	37.95°	41.36°	572	07.05.2023
<i>H. spontaneum</i>	Diyarbakır-1	37.91°	40.27°	652	15.05.2023
<i>H. spontaneum</i>	Diyarbakır-2	38.19°	39.36°	982	10.05.2023

Samples of plants and herbarium specimens from *Hordeum* species were collected at the anthesis stage of the plants. Determination of species was conducted by Prof. Dr. Selçuk ERTEKİN from Dicle University, Faculty of Science, Department of Biology (Diyarbakır, Türkiye). Approximately 200 g of green grass samples from each species were cut from the the plants at soil level. The samples were dried in a drying cabinet (Memmert ULM 800) at 70 °C for 48 hours (Anonymous, 2021). The dried samples were ground in a laboratory type mill (IKA, A11), then sieved in a 1 mm diameter sample sieve (Retsch, DIN-ISO 3310/2), and made ready for analysis.

Quality analyses of the grass samples were conducted with a NIRS analyzer (Near Infrared Spectroscopy-Foss Model 6500) in the laboratory of Dicle University, Science and Technology Application and Research Center (Diyarbakır, Türkiye). Crude protein (CP), acid detergent insoluble fiber (ADF), neutral detergent insoluble fiber (NDF), acid detergent insoluble protein (ADP), Ca, K, Mg, P, Ca/P, and K/(Ca+Mg) values were determined via analysis. Additionally, digestible dry matter (DDM), dry matter intake (DMI), and relative feed values (RFV) were calculated with the help of the determined ADF and NDF values. The following equations were used for calculations (Morrison, 2003).

$$\text{DDM} = 88.9 - (0.779 \times \text{ADF}) \quad (1)$$

$$\text{DMI} = 120 / \text{NDF} \quad (2)$$

$$\text{RFV} = (\text{DDM} \times \text{DMI}) / 1.29 \quad (3)$$

A comparison of the quality of the samples with reference standards was conducted according to the classification method of Lacefield (1988) given for legumes, grasses, and legume + wheatgrass mixtures (Table 2).

Table 2. Reference quality standards for legumes, grasses, and legume + wheat mixtures (Lacefield, 1988)

Quality standards	CP (%)	ADF (%)	NDF (%)	DDM (%)	DMI (%)	RFV
P	>19	<31	<40	>65	>3.0	>151
1	17-19	31-35	40-46	62-65	3.0-2.6	151-125
2	14-16	36-40	47-53	58-61	2.5-2.3	124-103
3	11-13	41-42	54-60	56-57	2.2-2.0	102-87
4	8-10	43-45	61-65	53-55	1.9-1.8	86-75
5	<8	>45	>65	<53	<1.8	<75

2.1. Evaluation of data

The produced data regarding the searched features were analyzed according to the one-way ANOVA test in the Jump-Pro13 statistical package program. Differences between means were compared with the Tukey HSD test. According to the scatter plot model, the principal components analysis was conducted in the Genstat (2009) 12th (Copyright 2011, VSN International Ltd) statistical package program and the scatterplot matrix was made in the Jump-Pro13 statistical package program. The research results were presented in a table using the one-way ANOVA test.

3. Results

The results of one-way variance analysis of the examined quality characteristics of *Hordeum* species and the resulting groups are given in Tables 3, 4, and 5.

Table 3. Comparisons of species according to one-way ANOVA using Tukey HSD test

Species	Location	Crude protein				
		Mean	Std Dev	Std Err Mean	Lower 95%	Upper 95%
<i>H. bulbosum</i>	Karacadag	10.09 d	0.20	0.12	9.60	10.59
<i>H. bulbosum</i>	Diyarbakır-3	8.15 f	0.22	0.13	7.60	8.70
<i>H. bulbosum</i>	Diyarbakır-2	11.47 c	0.16	0.09	11.06	11.88
<i>H. murinum</i>	Diyarbakır-2	13.85 b	0.21	0.12	13.33	14.38
<i>H. murinum</i> ssp. <i>leporinum</i>	Batman	11.93 c	0.05	0.03	11.79	12.07
<i>H. spontaneum</i>	Batman	9.19 e	0.22	0.12	8.66	9.73
<i>H. spontaneum</i>	Diyarbakır-3	9.72 d	0.18	0.10	9.27	10.17
<i>H. spontaneum</i>	Diyarbakır-1	23.38 a	0.04	0.02	23.27	23.48
Significance	**					
Coefficient variance (%)	1.40					
		Dry matter				
<i>H. bulbosum</i>	Karacadag	93.14 bc	0.064	0.037	92.982	93.300
<i>H. bulbosum</i>	Diyarbakır-3	92.09 f	0.098	0.057	91.847	92.335
<i>H. bulbosum</i>	Diyarbakır-2	92.88 d	0.057	0.033	92.735	93.018
<i>H. murinum</i>	Diyarbakır-2	93.35 a	0.002	0.001	93.350	93.357
<i>H. murinum</i> ssp. <i>leporinum</i>	Batman	93.09 bc	0.066	0.038	92.927	93.256
<i>H. spontaneum</i>	Batman	92.98 cd	0.050	0.029	92.856	93.104
<i>H. spontaneum</i>	Diyarbakır-3	93.23 ab	0.046	0.027	93.119	93.349
<i>H. spontaneum</i>	Diyarbakır-2	92.31 e	0.085	0.049	92.099	92.519
Significance	**					
Coefficient variance (%)	0.06					
		ADF				
<i>H. bulbosum</i>	Karacadag	30.86 c	0.595	0.344	29.379	32.338
<i>H. bulbosum</i>	Diyarbakır-3	34.93 b	0.146	0.084	34.564	35.290
<i>H. bulbosum</i>	Diyarbakır-2	35.37 ab	0.528	0.305	34.061	36.686
<i>H. murinum</i>	Diyarbakır-2	31.31 c	0.400	0.231	30.316	32.303
<i>H. murinum</i> ssp. <i>leporinum</i>	Batman	31.65 c	0.116	0.067	31.360	31.935
<i>H. spontaneum</i>	Batman	36.20 a	0.495	0.286	34.974	37.433
<i>H. spontaneum</i>	Diyarbakır-3	34.81 b	0.108	0.062	34.539	35.075
<i>H. spontaneum</i>	Diyarbakır-1	19.30 d	0.165	0.095	18.887	19.705
Significance	**					
Coefficient variance (%)	1.17					

Table 3. Comparisons of species according to one-way ANOVA using Tukey HSD test (continued)

Species	Location	Mean	Std Dev	Std Err Mean	Lower 95%	Upper 95%
			NDF			
<i>H. bulbosum</i>	Karacadag	55.31 d	0.987	0.370	54.525	56.093
<i>H. bulbosum</i>	Diyarbakır-3	45.99 e	0.508	0.370	45.202	46.771
<i>H. bulbosum</i>	Diyarbakır-2	59.51 c	0.881	0.370	58.729	60.298
<i>H. murinum</i>	Diyarbakır-2	59.17 c	0.581	0.370	58.388	59.957
<i>H. murinum</i> ssp. <i>leporinum</i>	Batman	62.39 b	0.399	0.370	61.602	63.171
<i>H. spontaneum</i>	Batman	71.86 a	0.705	0.370	71.072	72.641
<i>H. spontaneum</i>	Diyarbakır-3	63.61 b	0.306	0.370	62.822	64.391
<i>H. spontaneum</i>	Diyarbakır-1	26.15 f	0.437	0.370	25.369	26.938
Significance	**					
Coefficient variance (%)	1.15					

**; $P \leq 0.01$, *; $P \leq 0.05$ significant levels. Levels not connected by the same letter are significantly different.

3.1. Crude protein

In the research, the CP rate in different *Hordeum* ecotypes varied between 8.15-23.38%. The highest CP ratio (23.38%) was at *H. spontaneum* in Diyarbakır-1. The lowest CP value (8.15%) was at *H. bulbosum* in Diyarbakır-3 (Table 3). High CP value is an important characteristic of the quality roughage. The findings regarding the CP ratio in this research were within the limits specified by Sirat and Bahar (2020) (12.73-15.6%), but were found to be higher than the results of Sarı and Alatürk (2023) (6.30-15.89%).

3.2. DM

DM ratios in different *Hordeum* ecotypes varied between 92.09-93.35%. The highest DM ratio (93.35%) among the ecotypes was at *H. murinum* in Diyarbakır-3. The lowest DM value (92.09%) was obtained from *H. bulbosum* collected from the same region (Table 3). The findings regarding dry matter in the study were within the limits of the findings (65.80-95.72%) of Sarı and Alatürk (2023).

3.3. ADF

The ADF value in roughage refers to the amount of cellulose, lignin, and insoluble protein in the structure of the plant cell wall (Kutlu, 2008), and is desired to be as low as possible (Schroeder, 1994; Başbağ et al., 2020). The ADF values of different *Hordeum* ecotypes were between 19.30-36.20%. The highest ADF value (36.20%) was detected in *H. spontaneum* collected from the Batman region. The lowest ADF value (19.30%) was obtained from *H. spontaneum* at Diyarbakır-1 (Table 3). Unlike this research, some studies have reported the ADF values between 1.43-8.65% (Alijosius et al., 2016; Erbaş Köse and Mut, 2019; Sirat and Bahar, 2020).

3.4. NDF

In forages, NDF refers to the amount of hemicellulose, cellulose, lignin, cutin, and insoluble protein found in the structure of the plant cell wall (Aşçı and Acar, 2018). For this reason, low NDF value is desired in roughage (Mut et al., 2017; Başbağ et al., 2020). The NDF value in different *Hordeum* ecotypes was between 26.15-71.86%. The highest NDF value (71.86%) was detected in *H. spontaneum* collected from the Batman region. The lowest NDF value (26.15%) was obtained from *H. spontaneum* in the Diyarbakır-1 location (Table 3). Fife et al. (2008) reported NDF contents in barley between 19.9% and 24.5%. Also, Sirat and Bahar (2020) reported NDF values between 20.42 and 25.03%. It is stated by different researchers that NDF values may vary depending on genotype/varieties (Barteczko et al., 2009; Can and Ayan, 2017).

Table 4. Comparisons of species according to one-way ANOVA using Tukey HSD test

ADP						
Species	Location	Mean	Std Dev	Std Err Mean	Lower 95%	Upper 95%
<i>H. bulbosum</i>	Karacadag	0.17 f	0.074	0.043	-0.015	0.351
<i>H. bulbosum</i>	Diyarbakır-3	0.71 a	0.020	0.011	0.660	0.757
<i>H. bulbosum</i>	Diyarbakır-2	0.46 c	0.012	0.007	0.435	0.492
<i>H. murinum</i>	Diyarbakır-2	0.58 b	0.033	0.019	0.498	0.660
<i>H. murinum ssp. leporinum</i>	Batman	0.45 cd	0.015	0.008	0.411	0.484
<i>H. spontaneum</i>	Batman	0.37 d	0.000	0.000	0.370	0.370
<i>H. spontaneum</i>	Diyarbakır-3	0.43 cd	0.014	0.008	0.392	0.460
<i>H. spontaneum</i>	Diyarbakır-1	0.26 e	0.010	0.006	0.237	0.285
Significance	**					
Coefficient variance (%)	7.14					
DDM						
<i>H. bulbosum</i>	Karacadag	64.86 b	0.464	0.268	63.709	66.014
<i>H. bulbosum</i>	Diyarbakır-3	61.69 c	0.114	0.066	61.409	61.975
<i>H. bulbosum</i>	Diyarbakır-2	61.34 cd	0.412	0.238	60.322	62.367
<i>H. murinum</i>	Diyarbakır-2	64.51 b	0.312	0.180	63.736	65.284
<i>H. murinum ssp. leporinum</i>	Batman	64.25 b	0.090	0.052	64.023	64.470
<i>H. spontaneum</i>	Batman	60.70 d	0.386	0.223	59.740	61.656
<i>H. spontaneum</i>	Diyarbakır-3	61.79 c	0.084	0.049	61.577	61.994
<i>H. spontaneum</i>	Diyarbakır-1	73.87 a	0.128	0.074	73.550	74.187
Significance	**					
Coefficient variance (%)	0.46					
DMC						
<i>H. bulbosum</i>	Karacadag	2.17 c	0.039	0.022	2.074	2.266
<i>H. bulbosum</i>	Diyarbakır-3	2.61 b	0.029	0.017	2.538	2.681
<i>H. bulbosum</i>	Diyarbakır-2	2.02 de	0.030	0.017	1.942	2.091
<i>H. murinum</i>	Diyarbakır-2	2.03 d	0.020	0.012	1.979	2.078
<i>H. murinum ssp. leporinum</i>	Batman	1.92 ef	0.012	0.007	1.893	1.954
<i>H. spontaneum</i>	Batman	1.67 g	0.016	0.009	1.629	1.711
<i>H. spontaneum</i>	Diyarbakır-3	1.89 f	0.009	0.005	1.864	1.909
<i>H. spontaneum</i>	Diyarbakır-1	4.59 a	0.077	0.044	4.399	4.780
Significance	**					
Coefficient variance (%)	1.27					
RFV						
<i>H. bulbosum</i>	Karacadag	109.12 c	2.728	1.575	102.350	115.900
<i>H. bulbosum</i>	Diyarbakır-3	124.80 b	1.609	0.929	120.810	128.800
<i>H. bulbosum</i>	Diyarbakır-2	95.91 de	2.070	1.195	90.760	101.050
<i>H. murinum</i>	Diyarbakır-2	101.42 d	1.486	0.858	97.730	105.110
<i>H. murinum ssp. leporinum</i>	Batman	95.80 de	0.746	0.431	93.950	97.650
<i>H. spontaneum</i>	Batman	78.59 f	1.270	0.733	75.430	81.740
<i>H. spontaneum</i>	Diyarbakır-3	90.36 e	0.536	0.310	89.030	91.690
<i>H. spontaneum</i>	Diyarbakır-1	262.79 a	4.846	2.798	250.750	274.830
Significance	**					
Coefficient variance (%)	1.91					
Phosphorous						
<i>H. bulbosum</i>	Karacadag	0.36 c	0.007	0.004	0.346	0.379
<i>H. bulbosum</i>	Diyarbakır-3	0.30 e	0.002	0.001	0.297	0.307
<i>H. bulbosum</i>	Diyarbakır-2	0.40 b	0.001	0.001	0.400	0.400
<i>H. murinum</i>	Diyarbakır-2	0.41 b	0.004	0.003	0.395	0.417
<i>H. murinum ssp. leporinum</i>	Batman	0.35 d	0.001	0.001	0.346	0.350
<i>H. spontaneum</i>	Batman	0.35 d	0.006	0.003	0.332	0.361
<i>H. spontaneum</i>	Diyarbakır-3	0.36 c	0.002	0.001	0.360	0.370
<i>H. spontaneum</i>	Diyarbakır-1	0.42 a	0.002	0.001	0.417	0.428
Significance	**					
Coefficient variance (%)	0.83					

**; P≤0.01, *; P≤0.05 significant levels. Levels not connected by the same letter are significantly different.

3.5. ADP

The ADP ratios of different *Hordeum* ecotypes were between 0.171-0.71%. The highest ADP ratio (0.71%) was at *H. bulbosum* in Diyarbakır-3 location. The lowest ADP value (0.17%) was obtained from *H. bulbosum* from Karacadağ region (Table 4). Low ADP values are desirable as they reflect the amount of indigestible protein in roughage (Aşçı and Acar, 2018).

3.6. DDM

The DDM values in *Hordeum* ecotypes varied between 60.70 and 73.87%. The highest DDM value (73.87%) was detected at *H. spontaneum* in Diyarbakır-1 location. The lowest DDM value

(60.70%) was obtained from *H. spontaneum* from the Batman region (Table 4). The DDM value varies depending on the species, and different results have been reported by different researchers (Kaplan, 2021; Tutar and Kökten, 2022; Arıkan et al., 2023) regarding the DDM value.

3.7. DMI

The DMI values in *Hordeum* ecotypes varied between 1.67 and 4.59%. The highest DMI value (4.59%) among the ecotypes was determined at *H. spontaneum* in the Diyarbakır-1 location. The lowest DMI value was obtained from *H. spontaneum* (1.67%) in the Batman region (Table 4). Arıkan et al., (2023) reported the DMI value in barley as 2.02%.

3.8. RFV

RFV values in *Hordeum* ecotypes varied between 78.59 and 262.79. The highest RFV value (262.79) was determined at *H. spontaneum* in the Diyarbakır-1 location. The lowest RFV value (78.59) was obtained from *H. spontaneum* in the Batman region (Table 4). Canbolat (2012) and Arıkan et al., (2023) reported higher RFV value in wheat compared to barley.

3.9. Phosphorous (P)

P values in *Hordeum* ecotypes varied between 0.30-0.42%. The highest phosphorus value (0.42%) among the species was determined at *H. spontaneum* in Diyarbakır-1. The lowest P value (0.30%) was at *H. bulbosum* in Diyarbakır-2, (Table 4). Sirat and Bahar (2020) reported the P rates between 0.42-0.44%. In another study, P values varied between 0.363 and 0.408% (Mut and Erbaş Köse, 2018). Some researchers reported that P values vary according to genotypes (Poutanen, 2012; Jakobsone et al., 2015).

Table 5. Comparisons of species according to one-way ANOVA using Tukey HSD test

Species	Location	Potassium				
		Mean	Std Dev	Std Err Mean	Lower 95%	Upper 95%
<i>H. bulbosum</i>	Karacadag	1.73 d	0.092	0.053	1.497	1.953
<i>H. bulbosum</i>	Diyarbakır-3	2.56 b	0.033	0.019	2.480	2.645
<i>H. bulbosum</i>	Diyarbakır-2	2.50 b	0.074	0.043	2.314	2.680
<i>H. murinum</i>	Diyarbakır-2	2.84 a	0.020	0.011	2.796	2.893
<i>H. murinum ssp. leporinum</i>	Batman	2.25 c	0.088	0.051	2.035	2.472
<i>H. spontaneum</i>	Batman	1.79 d	0.075	0.043	1.600	1.973
<i>H. spontaneum</i>	Diyarbakır-3	2.32 c	0.021	0.012	2.264	2.368
<i>H. spontaneum</i>	Diyarbakır-1	2.59 b	0.053	0.031	2.460	2.723
Significance	**					
Coefficient variance (%)	2.58					
		Calcium				
<i>H. bulbosum</i>	Karacadag	0.12 f	0.062	0.036	-0.034	0.274
<i>H. bulbosum</i>	Diyarbakır-3	1.12 b	0.009	0.005	1.099	1.146
<i>H. bulbosum</i>	Diyarbakır-2	0.54 c	0.023	0.013	0.486	0.601
<i>H. murinum</i>	Diyarbakır-2	0.41 d	0.024	0.014	0.345	0.466
<i>H. murinum ssp. leporinum</i>	Batman	0.58 c	0.004	0.002	0.572	0.592
<i>H. spontaneum</i>	Batman	0.31 e	0.010	0.006	0.285	0.335
<i>H. spontaneum</i>	Diyarbakır-3	0.32 e	0.001	0.000	0.318	0.321
<i>H. spontaneum</i>	Diyarbakır-1	1.62 a	0.004	0.003	1.613	1.635
Significance	**					
Coefficient variance (%)	3.22					
		Magnesium				
<i>H. bulbosum</i>	Karacadag	0.25 c	0.004	0.002	0.237	0.257
<i>H. bulbosum</i>	Diyarbakır-3	0.30 a	0.002	0.001	0.301	0.309
<i>H. bulbosum</i>	Diyarbakır-2	0.17 f	0.000	0.000	0.170	0.170
<i>H. murinum</i>	Diyarbakır-2	0.24 c	0.003	0.002	0.234	0.250
<i>H. murinum ssp. leporinum</i>	Batman	0.23 d	0.001	0.001	0.231	0.235
<i>H. spontaneum</i>	Batman	0.22 e	0.000	0.000	0.220	0.220
<i>H. spontaneum</i>	Diyarbakır-3	0.17 f	0.003	0.002	0.162	0.179
<i>H. spontaneum</i>	Diyarbakır-1	0.29 b	0.004	0.003	0.282	0.304
Significance	**					
Coefficient variance (%)	0.86					

Table 5. Comparisons of species according to one-way ANOVA using Tukey HSD test (continued)

Species	Location	Mean	Std Dev	Std Err Mean	Lower 95%	Upper 95%
Ca/P						
<i>H. bulbosum</i>	Karacadag	0.33 e	0.177	0.102	-0.107	0.773
<i>H. bulbosum</i>	Diyarbakır-3	3.72 a	0.056	0.032	3.576	3.854
<i>H. bulbosum</i>	Diyarbakır-2	1.36 c	0.058	0.033	1.215	1.502
<i>H. murinum</i>	Diyarbakır-2	1.00 d	0.049	0.028	0.878	1.122
<i>H. murinum</i> ssp. <i>leporinum</i>	Batman	1.67 b	0.007	0.004	1.656	1.690
<i>H. spontaneum</i>	Batman	0.89 d	0.017	0.010	0.851	0.937
<i>H. spontaneum</i>	Diyarbakır-3	0.88 d	0.006	0.004	0.861	0.892
<i>H. spontaneum</i>	Diyarbakır-1	3.84 a	0.033	0.019	3.764	3.925
Significance	**					
Coefficient variance (%)	4.09					
K/(Ca+Mg)						
<i>H. bulbosum</i>	Karacadag	4.77 a	0.618	0.357	3.235	6.307
<i>H. bulbosum</i>	Diyarbakır-3	1.80 d	0.010	0.005	1.772	1.819
<i>H. bulbosum</i>	Diyarbakır-2	3.50 b	0.025	0.014	3.439	3.562
<i>H. murinum</i>	Diyarbakır-2	4.40 a	0.217	0.125	3.858	4.934
<i>H. murinum</i> ssp. <i>leporinum</i>	Batman	2.77 c	0.125	0.072	2.456	3.075
<i>H. spontaneum</i>	Batman	3.37 bc	0.078	0.045	3.175	3.564
<i>H. spontaneum</i>	Diyarbakır-3	4.73 a	0.078	0.045	4.533	4.922
<i>H. spontaneum</i>	Diyarbakır-1	1.35 d	0.034	0.020	1.268	1.437
Significance	**					
Coefficient variance (%)	6.90					

**; $P \leq 0.01$, *; $P \leq 0.05$ significant levels. Levels not connected by the same letter are significantly different.

3.10. Potassium (K)

K, which has important roles in photosynthesis, enzyme activity, and regulation of the water content of plants, is very important for the sugar and protein contents of cereal grains (Güneş et al., 2000). K value in *Hordeum* ecotypes varied between 1.73-2.84%. The highest K value (2.84%) was at *H. murinum* in Diyarbakır-2 location. The lowest K values were at *H. bulbosum* in Karacadağ (1.73%) and from *H. spontaneum* (1.79%) from Batman region (Table 5). Sirat and Bahar (2020) reported a K value between 0.68-0.79%. The findings obtained in this study regarding the K value were higher than the findings of other researchers. This difference may be sourced from the performance of different genotypes under different ecological conditions. Similarly, Poutanen (2012) stated that K values vary according to varieties.

3.11. Calcium (Ca)

Due to Ca deficiency, rickets, osteomalacia, and urinary system stone disease may occur in animals. Ca, which is found in limited amounts, especially in the bones of newborn animals, is absolutely necessary for the development of bones (Anonymous, 2021). Ca value obtained in *Hordeum* ecotypes varied between 0.12-1.62%. The highest Ca value (1.62%) was determined at *H. spontaneum* in Diyarbakır-1 location. The lowest Ca value (0.12%) was obtained from *H. bulbosum* from the Karacadağ region (Table 5). Gül et al., (2022) determined Ca as 0.45% in their research.

3.12. Magnesium (Mg)

Mg acts as an enzyme activator and its deficiency may result in meadow tetany disease in animals (Underwood, 1981). Magnesium is also known as the "antistress mineral" as it helps reduce the hypersensitivity of the animal nervous system. It plays a role in activating enzymes and converting sugar into energy in the blood. Mg deficiency in sheep causes meadow tetany, in the form of contraction of the legs and lifting of the head backward (Ensminger et al., 1990).

Mg value of studied *Hordeum* ecotypes varied between 0.17-0.30%. The highest Mg value (0.30%) was at *H. bulbosum* in Diyarbakır-3. The lowest Mg values were in Diyarbakır-2 both at *H. bulbosum* (0.17%) and at *H. spontaneum* (0.17%) (Table 5). Sirat and Bahar (2020) reported that the Mg values varied between 0.17-0.19%, which is similar to this study's Mg values.

3.13. Calcium/Phosphorus (Ca/P)

The Ca/P ratio in studied *Hordeum* ecotypes varied between 0.33-3.84. The highest Ca/P ratio (3.84) was at *H.spontaneum* Diyarbakır-1. The lowest Ca/P ratio value (0.33) was obtained from *H. bulbosum* collected from the Karacadag region (Table 5). Ayan et al., (2010) and Albu et al., (2012) reported that the Ca/P ratio in an ideal feed should be 1/1 or 2/1. In their research, Gül et al. (2022) found the Ca/P ratio in barley as 1.3. The findings we obtained regarding the Ca/P ratio were within the range of the findings reported by Gül et al. (2022).

3.14. Potassium/(Calcium+Magnesium) [K/(Ca+Mg)]

The K/(Ca+Mg) ratios in studied *Hordeum* ecotypes varied between 1.35 and 4.77. The highest K/(Ca+Mg) ratios were obtained from *H. bulbosum* in Karacadag (4.77), *H. murinum* in Diyarbakır-2 (4.39) and *H. spontaneum* in Diyarbakır-3 (4.73). The lowest K/(Ca+Mg) ratio value (1.35) was obtained from *H. spontaneum* collected from Diyarbakır-1 (Table 5). Gül et al. (2022) reported the K/(Ca+Mg) ratio in barley as 3.31. The findings obtained from this study were higher than the findings of Gül et al. (2022).

3.15. Interpretation of relationships between features with Scatterplot matrix and Biplot analysis

The relationships between features can be interpreted in the scatterplot matrix graphic obtained based on correlation coefficient values (Karaman, 2022). If the distribution representing the relationship between any two features does not show a regular accumulation on the regression curve, the relationship between these two features can be commented as weak or non-existent. However, if the distribution on the regression curve is regular, it can be concluded that there is a strong relationship between these two features. There were generally strong relationships between the examined traits in our research, especially between ADF and CP; DDM and CP; DMI and ADF; RFV and ADF; RFV and NDF; DMI and DDM; RFV and DDM; RFV and DMI; Ca/P and Ca, K values. The correlation coefficient between K/(Ca+Mg) and Mg and Ca/P values were close to ± 1 and the distribution was regular on the regression line which shows that there were very strong relationships between these characters (Figure 1).

The relationships between the examined features of *Hordeum* ecotypes were determined by using the scatterplot biplot technique obtained from the average data of the relevant examined features through polygons and sectors (Figure 2). In the biplot analysis, it was observed that the two-dimensional PCA score PC1 was 61.96%, PC2 was 21.36%, and the total variation (PC1+PC2) was 83.33%. In different studies, PC1, PC2, and PC1+PC2 scores were also found very diverse (Sayar et al., 2018; Başbağ et al., 2021).

In the Scatterplot biplot graph, the regression coefficient between some features, was close to ± 1 , which gave the same result in the biplot produced by using sectors, polygons, and mega environments. This showed that the mentioned features were strongly interrelated (Figure 2).

There appeared four sectors in the graph: K/(Ca+Mg) and DM in the first sector; P, DDM, CP, DMI, RFV, K, Mg, Ca, and Ca/P in the second sector; ADF and ADP in the third sector; and NDF in the fourth sector. The mentioned features represent the highest values for the ecotypes in the relevant sectors. If there were ecotypes and features in different sectors, it can be commented that there was no genotype standing out in terms of the relevant feature. Instead, if they are located in the same sector, this indicates a positive relationship (Figure 2).

If all of the features are located in the same sector, it can be commented that these features show a complex interaction (Chinipardaz et al., 2016). The ecotypes located on the diagonals of the polygon and indicated by a number produced the best performance in terms of the feature in the relevant sector (Yan and Tinker 2006; Ahmadi et al., 2012). From this perspective, numbers 1 (*Hordeum bulbosum* in Karacadag), number 2 (*Hordeum bulbosum* in Diyarbakır-3), number 6 (*Hordeum spontaneum* in Batman), and number 8 (*Hordeum spontaneum* in Diyarbakır-1) represent the best averages for the observed features in the sectors.

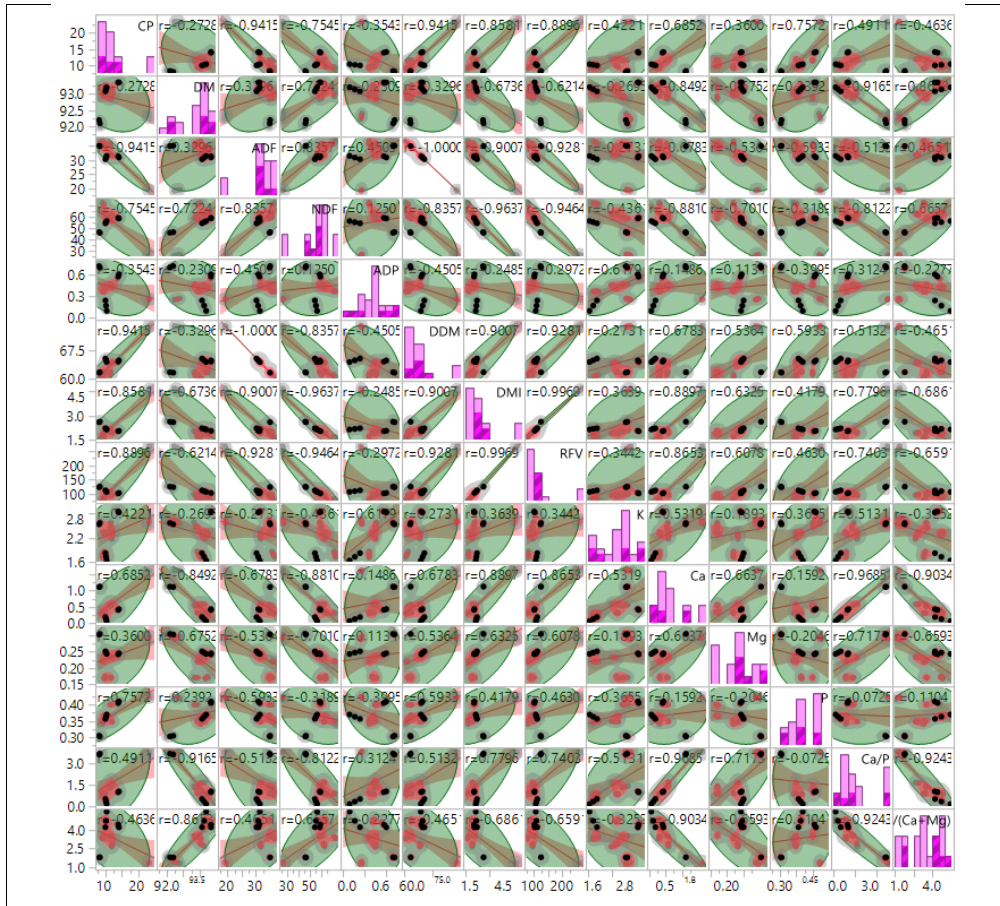


Figure 1. Representation of the relationships between features with a scatterplot matrix.

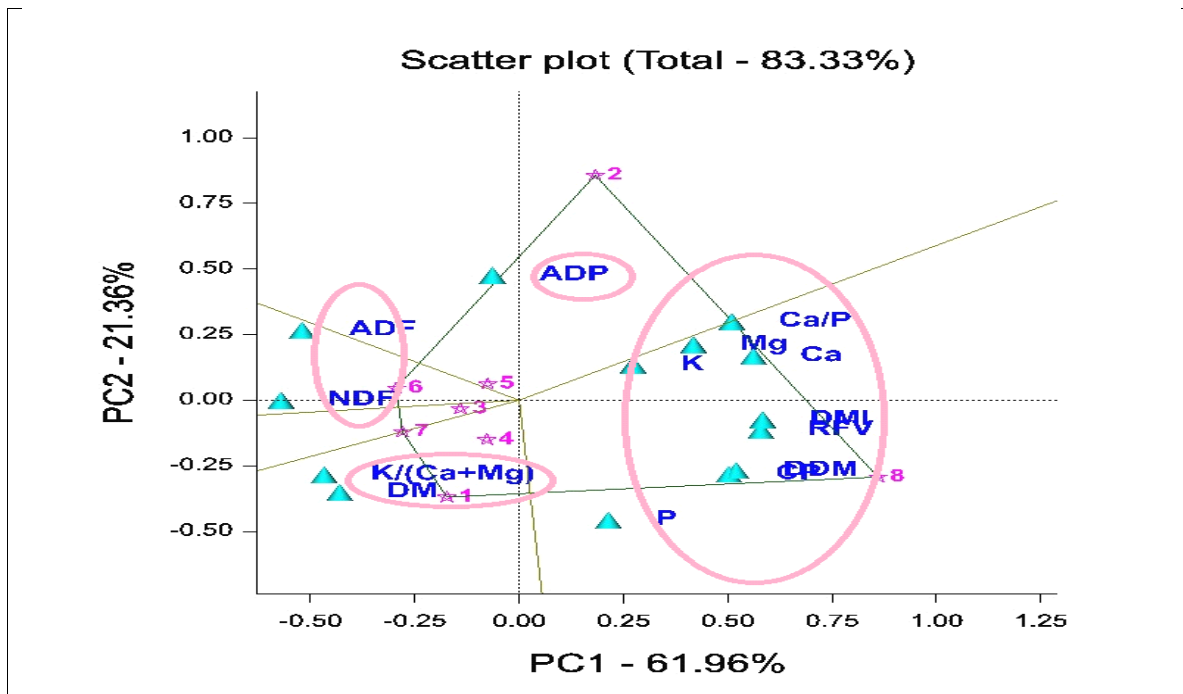


Figure 2. Representation of the relationship between features with scatterplot matrix. 1) *Hordeum bulbosum* in Karacadađ, 2) *Hordeum bulbosum* in Diyarbakır-3, 3) *Hordeum bulbosum* in Diyarbakır-2, 4) *Hordeum murinum* in Diyarbakır-3, 5) *H. murinum* ssp. *leporinum* in Batman, 6) *Hordeum spontaneum* in Batman, 7) *Hordeum spontaneum* in Diyarbakır-3, 8) *Hordeum spontaneum* in Diyarbakır-1.

Conclusion

The values were determined between 8.2-23.4% for CP; 92.1-93.4% for DM; 19.3-36.2% for ADF; 26.2-71.9% for NDF; 0.16-0.71% for ADP; 60.7-73.9% for DDM; 1.67-4.58% for DMI; 78.6-262.8 for RFV; 0.30-0.42% for P; 1.72-2.84% for K; 0.12-1.62% for Ca; 0.17-0.30% for Mg; 0.33-3.84 for Ca/P; 2.76-4.77 for K/(Ca+Mg). As a conclusion, the CP, ADF, NDF, DMI, RFV, and Ca/P values were found very variable in collected ecotypes and can be used for forage barley breeding purposes.

Ethical Statement

Ethical approval is not required for this study because ethics is not required for plants.

Conflict of Interest

All authors declare that there is no conflict of interest related to this article.

Author Contributions

The authors declare that they have contributed equally to the article. All authors declare that they have seen/read and approved the final version of the article ready for publication.

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