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Allelic variations of glutenin subunits and their association with quality traits in bread wheat genotypes

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Abstract: The present study was conducted to evaluate the genotype × environment interaction of the yield and quality traits for five bread wheat varieties commonly grown in the Southeastern Anatolia Region of Turkey and 20 advanced lines developed within the framework of the International Winter Wheat Improvement Project. We also determined the allelic pattern of the *Glu-1* and *Glu-3* loci of these genotypes and examined whether these loci had an effect on the quality traits. There was a significant variation among the genotypes and environments in terms of grain yield, protein content, sedimentation volume (SV), and the extensograph dough energy value (EDEV). The results of the study indicated that genotypic effect was more influential on SV and EDEV than environmental effect; thus, both traits could be used in breeding programs to develop elite cultivars with better quality. Twelve different high-molecular-weight (HMW) glutenin alleles were identified at the *Glu-A1*, *Glu-B1*, and *Glu-D1* loci, resulting in 14 allelic combinations, and 17 different alleles were observed in 19 combinations for low-molecular-weight (LMW) subunits. Furthermore, among all the genotypes, 13 + 16 and 13 + 19 alleles at *Glu-B1* and 5 + 12 at *Glu-D1* were observed to have the lowest frequency. Our study indicated that the combinations of HMW glutenin alleles with 2* at *Glu-A1*, 17 + 18 and 13 + 16 at *Glu-B1*, and 5 + 10 at *Glu-D1*, as well as the combinations of LMW alleles with subunits c and d at *Glu-A3*; subunits d, b, c, and g at *Glu-B3*; and subunits a and b at *Glu-D3* had positive effects on the quality traits.

Key words: Seed storage protein, bread wheat, quality, high molecular weight

1. Introduction

Turkey is a major producer of wheat, with approximately 9 × 10^6 ha sown annually leading to an annual production of 20 × 10^6 t (Baloch et al., 2016). In Turkey, a national wheat breeding program was initiated in 1967, the National Wheat Release and Training Project, with the collaboration of international organizations such as the International Maize and Wheat Improvement Center (CIMMYT) and International Center for Agricultural Research in the Dry Areas (ICARDA). This program started the process of a national green revolution that resulted in an increase in wheat yield and the development of more than 100 wheat cultivars, most significantly contributing to the economy of Turkey. Nearly all the wheat cultivars in Turkey were direct or indirect input from CIMMYT and ICARDA bread wheat breeding programs that focused on increasing the quality and quantity of wheat for the developing world (Alsaleh et al., 2015, 2016). However, the Turkish flour industry has continued to import high-quality wheat at a high cost due to the lack of cultivars with good quality traits. Therefore, one of the main objectives of the Turkish bread wheat breeding program is to develop wheat varieties with higher grain yield (GY) and better bread-making qualities in terms of protein and gluten content (Baloch et al., 2017). In this process, it is important to consider and evaluate genotypes, environments, and their interaction as the main factors affecting wheat quality traits.

The quality of bread-making is mainly related to the composition of endosperm storage proteins, namely gliadins and glutenins (Dessalegn et al., 2011). Gliadins are monomeric proteins synthesized by the *Gli-1* and *Gli-2* loci, which are located on the short arms of homoeologous chromosomes (He et al., 2005). Gliadin and glutenin represent 80% of total seed storage proteins (Perron et al., 1998). Gliadins have effects on dough viscosity and glutenins contribute to dough elasticity (Ikeda et al., 2006). There are two types of glutenin, low-molecular-weight (LMW) and high-molecular-weight (HMW). LMW glutenin subunits are encoded by the *Glu-3* loci located on the short arms of chromosomes 1A, 1B, and 1D while HMW subunits are encoded by the *Glu-1* loci on chromosomes 1A, 1B, and 1D (Békés et al., 2006).

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Although HMW subunits constitute only 10% of storage proteins compared to the 40% LMW content, the former have a larger effect on bread-making quality (He et al., 2005).

It has been reported that subunits designated as *GluA1*-1, *GluA1*-2*, *GluB1*-7 + 9, *GluB1*-17 + 18, and *GluD1*-5 + 10 are associated with high technological quality, whereas their allelic variants such as *GluA1* null, *GluB1*-6 + 8, and *GluD1*-2 + 12 are related to lower baking quality (Horvat et al., 2006). Similarly, environmental conditions and soil fertility have high positive and negative effects on wheat quality. The only factor that is not influenced by environmental conditions is the coding of HMW and LMW glutenin subunits by different alleles. Therefore, glutenin subunits could serve as useful markers in wheat breeding programs to improve end-use quality. Although the efforts of wheat breeders to produce elite cultivars with high yield and improved quality have produced positive results, breeding for higher quality needs further investigation to minimize the effect of the environment on quality traits.

The aims of the present study were to identify HMW and LMW subunits of 25 bread wheat genotypes and investigate the correlation between the quality scores of HMW subunits and quality traits, to determine whether HMW and LMW glutenin subunits can be used as markers to improve genotypes with high quality in wheat breeding programs, and to investigate the relationship of GY and quality traits with genotype, environment, and genotype × environment interaction (GEI) in three provinces of Turkey.

2. Materials and methods

2.1. Plant culture

The plant materials of this study were 20 advanced winter wheat genotypes that originated from the International Winter Wheat Improvement Project and five cultivars commonly grown in Southeast Turkey. Wheat genotypes were planted in November for all locations during the 2011–2012 cropping season according to a completely randomized block design with three replications at three different experimental stations (Diyarbakır, Adıyaman, and Mardin provinces) of the GAP International Agricultural Research and Training Center, Turkey. All the plots were treated in the same manner by following standard local agricultural practices. Plots had 6 rows, which were 5 m long and spaced at 20 cm. Plots were seeded at 200 kg ha^{-1} and fertilized with 60 kg ha^{-1} urea and 120 kg ha^{-1} DAP fertilizers. The four middle rows of each plot were harvested to calculate GY (t ha^{-1}) according to Pask et al. (2012).

2.2. Soil and climatic conditions

The soil properties of the three locations were clay-silt with pH ranging from 7.5 to 7.9. In the 2011–2012 growing season, precipitation was 570 mm, 382 mm, and 295 mm in Diyarbakır, Adıyaman, and Mardin provinces, respectively.

2.3. Protein extraction and SDS-PAGE

Protein extraction was performed following the method described by Singh et al. (1991) using three or more individual seeds of a single spike of each genotype. The SDS-PAGE method (Payne et al., 1987) was used with 11.5% polyacrylamide with minor modifications. Allelic variations of HMW and LMW glutenin subunits at the *Glu-1* loci were noted by labeling the bands at each subunit as *Glu-Al*, *Glu-B1*, and *Glu-DI*. The allelic variations of LMW glutenin subunits at the *Glu-3* loci were recorded using the labels *Glu-A3*, *Glu-B3*, and *Glu-D3*. The quality scores of HMW subunits were calculated according to the method described by Payne et al. (1987) by adding together the score of individual subunits.

2.4. Flour milling and quality traits

The wheat grains of each genotype were stored at 16% moisture and milled using a Brabender Junior mill according to AACC Method No. 26-50 (AACC, 1995). Protein analysis was performed according to AACC Method No. 39-10 (AACC, 1990) using a Near Infrared Model 6500 spectrophotometer. The Zeleny sedimentation value (SV) was determined according to ICC Standard Method No. 115/1 (ICC, 1982b) and the wet gluten (WG) value was calculated according to ICC Standard Method No. 155/1 (ICC, 1994) using a Glutomatic 2200 gluten washer. The extensograph dough energy value (EDEV) was determined according to ICC Standard Method No. 114/1 (ICC, 1982a). The test weight (TW) of each genotype was calculated as kg/hL based on AACC Method No. 44-15A (AACC, 2000). Thousand-kernel weight (TKW) was recorded in g/1000 kernels of cleaned wheat and GY was recorded as t ha^{-1}.

2.5. Data analysis

Variance and correlation analyses were performed using Statistica software version 7, and the means were calculated and compared with the LSD test ($P < 0.05$).

3. Results

3.1. Results of ANOVA testing

According to combined ANOVA, significant differences ($P < 0.05$) were observed between genotypes, environments, and GEI in terms of GY and quality traits (Table 1). In addition, the environmental variation explained a major portion of the total variation for GY (89.5%), TKW (57.8%), TW (74.9%), and protein content (PC) (71%) and a smaller proportion of the total variation for WG content (51.3%), SV (45.2%), and EDEV (26.2%) (Table 1).

Table 1. ANOVA and G, E, and GEI variance for grain yield and quality traits across environments.

	Mean square				Variation (%)			Locations mean for traits		
	G	E	GEI	Rep & random	G	E	GEI	DYB	ADY	MRD
Df	24	2	48	6						
GY	1.16**	530.33**	1.30**	0.21 ns	2.4	89.5	5.3	8.20 a	5.10 b	2.96 c
TKW	32.68**	1510.99**	25.12**	2.2 ns	15.1	57.8	23.1	42.33 a	36.71 b	33.46 c
TW	14.71**	894.72**	4.09**	0.33 ns	14.8	74.9	8.2	82.52 a	80.03 b	75.70 c
PC	5.59**	210.08**	0.14 ns	0.29 ns	23	71	1.2	11.73 c	12.30 b	14.87 a
WG	30.15**	579.34**	2.61*	1.1 ns	32.1	51.3	5.5	25.88 c	27.81 b	31.36 a
SV	196.70**	2153.90**	3.29 ns	2.1 ns	49.6	45.2	1.7	23.24 c	27.17 b	33.84 a
EDEV	9014.52**	40,109.30**	138.59**	41.9 ns	70.5	26.2	2.2	79.55 c	97.73 b	125.5 a

GY: Grain yield (t ha^{-1}), TKW: 1000-kernel weight (g), TW: test weight (kg/hL), PC: grain protein content (%), WG: wet gluten content (%), SV: sedimentation volume (mL), EDEV: extensograph dough energy value (cm^2), **: significant at level 0.01, *: significant at level 0.05, ns: nonsignificant, G: genotype, E: environment, GEI: genotype × environment interaction, Df: degrees of freedom, DYB: Diyarbakır location, ADY: Adıyaman location, MRD: Mardin location. Values in a row followed by different letters are significantly different.

The mean values for the locations of the examined traits are given in Table 1. There was a high diversity in the mean values for GY (2.96 to 8.20 t ha^{-1}), PC (11.73% to 14.87%), SV (23.24 to 33.84 mL), and WG (25.88% to 31.36%). Furthermore, TKW ranged from 33.46 to 42.33 g, TW from 75.70 to 82.52 kg/hL, and EDEV from 79.55 to 125.5 cm^2.

3.2. Quality scores of allelic combinations of HMW and LMW glutenin subunits and the correlation between the examined traits of wheat genotypes

The combinations of HMW and LMW glutenin subunit alleles with their frequencies and quality scores are given in Tables 2 and 3. Twelve different *Glu-1* alleles were observed in 15 combinations of HMW glutenin subunits: three at *Glu-A1*, six at *Glu-B1*, and three at *Glu-D1*. The allelic variations at the *Glu-A1* locus (null, 1 and 2*) found in our germplasm were in agreement with those reported by Békés and Wrigley (2003). At the *Glu-A1* locus, the subunits with the 2* allele had the highest frequency (72%) while the null allele represented the lowest percentage (4%). At the *Glu-B1* locus, six different combinations of HMW glutenin subunits were observed (17 + 18, 13 + 16, 7 + 9, 7 + 8, 7, and 13 + 19), and at the *Glu-D1* locus, three different combinations of subunits (5 + 10, 2 + 12, and 5 + 12) were determined. The allelic combinations of HMW glutenin subunits were observed at the following frequencies: *Glu-A1*-1 (24%), *Glu-A1*-2* (72%), *Glu-A1*-null (4%), *Glu-B1*-7 + 9 (12%), *Glu-B1*-7 + 8 (44%), *Glu-B1*-17 + 18 (20%), *Glu-B1*-13 + 19 (4%), *Glu-B1*-13 + 16 (12%), *Glu-B1*-7 (12%), *Glu-D1*-5 + 10 (56%), *Glu-D1*-2 + 12 (40%), and *Glu-D1*-5 + 12 (4%). The HMW glutenin subunit combination *Glu-B1*-13 + 19 was only found in G13 while the *Glu-B1*-13 + 16 combination was present in three genotypes (G15, G16, and G19). The overall quality score of individual genotypes ranged from 6 to 10 (Table 2). According to the results, genotypes with high quality scores had a high SV and EDEV. Genotypes G7, G8, G9, and G25 with *Glu-A1*-2* (at *Glu-A1*), *Glu-B1*-17 + 18, *Glu-B1*-13 + 16, *Glu-B1*-7 + 8, and *Glu-D1*-5 + 10 had a high performance in terms of SV and EDEV.

According to the results of the SDS-PAGE analysis for LMW glutenin subunits, 17 different *Glu-3* alleles were determined in 19 combinations: five at *Glu-A3*, eight at *Glu-B3*, and four at *Glu-D3*. The frequencies of LMW glutenin subunits at *Glu-A3* ranged from 4% to 60%, being 4% for *Glu-A3*-a, 8% for *Glu-A3*-b, 60% for *Glu-A3*-c, 24% for *Glu-A3*-d, and 4% for *Glu-A3*-e (Tables 2 and 3). The highest and the lowest frequencies of *Glu-B3* belonged to *Glu-B3*-c (4%) and *Glu-B3*-g (4%), while the frequency of *Glu-B3*-b was 36%. The combinations with the highest and lowest frequencies of alleles at the *Glu-D3* loci were *Glu-D3*-c (48%) and *Glu-D3*-d (4%), respectively.

The mean values for the examined traits are given in Table 3 by alleles. The genotypes with *Glu-A1*-2* had the highest mean value for TKW (42.5 g), TW (82.5 kg/hL), and EDEV (86.4 cm^2), whereas the genotypes with null subunits had the lowest scores for all traits except grain PC and TKW. The genotypes with subunits *Glu-A1*-2* and *Glu-B1*-1 showed similar results for SV (23.7 and 23.2 mL, respectively) and GY (8.22 and 8.83 t ha^{-1}, respectively). The variation in *Glu-B1* alleles was higher compared to *Glu-A1* and *Glu-D1*. The genotypes carrying the subunit combination *Glu-B1*-17 + 18 had the highest mean score for SV (36.3 mL) and EDEV (143 cm^2). Similarly, subunits

Table 2. Combination of *Glu-1 and Glu-3* alleles and quality scores of genotypes.

Genotypes	Pedigree/genotypes	HMW-GS				LMW-GS		
		Glu-A1	*Glu-B1*	*Glu-D1*	Quality scores for HMW-GS (QS)	*Glu-A3*	*Glu-B3*	*Glu-D3*
G1	BEZOSTAYA	2*	7 + 9	5 + 10	9	c	b	c
G2	KINACI97	2*	7 + 9	2 + 12	7	c	e	b
G3	KATIA	1	7 + 8	2 + 12	8	c	e	c
G4	KONYA	2*	7 + 8	2 + 12	8	a	c	c
G5	CEMRE	2*	17 + 18	5 + 12	6	c	c	a
G6	PYN/PARUS/3/VPM/MOS83-11-4-8//PEW/4/BLUEGIL	1	7 + 8	2 + 12	8	d	i	b
G7	SHARK-1/3/AGRI/BJY//VEE/4/SHARK/F4105W2.1	2*	17 + 18	5 + 10	10	c	c	a
G8	SHARK-1/3/AGRI/BJY//VEE/4/SHARK/F4105W2.1	2*	17 + 18	5 + 10	10	c	c	a
G9	RSK/CA8055//CHAM6/4/NWT/3/TAST/SPRW//TAW12399.75	2*	17 + 18	5 + 10	10	c	c	c
G10	PYN/PARUS/3/VPM/MOS83-11-4-8//PEW/4/BLUEGIL	1	7	2 + 12	6	d	h	b
G11	TAM200/KAUZ//YU MAI30	2*	7 + 9	2 + 12	7	d	e	a
G12	NWAU15/ATTILA//SHARK/F4105W2.1	2*	7 + 8	5 + 10	10	c	d	a
G13	BLUEGIL-2/BUCUR//SIRENA	1	13 + 19	2 + 12	8	e	i	c
G14	F6038W12.1/ERYT25221//F6038W12.1	2*	7 + 8	5 + 10	10	c	c	c
G15	YMH/HYS//HYS/TUR3055/3/DGA/4/VPM/MOS/5/5/STEPOWI	2*	13 + 16	5 + 10	10	d	i	a
G16	AIZAO781/6/LOV11/SON64/4/PJ/GB55//093//7/RSK/CA8055//CHAM6	2*	13 + 16	2 + 12	8	c	i	a
G17	4WON-IR 257/5/YMH/HYS//HYS/TUR3055/3/DGA/4/VPM/MOS	2*	7	5 + 10	7	d	d	c
G18	4WON-IR-257/5/YMH/HYS//HYS/TUR3055/3/DGA/4/VPM/MOS	2*	7	5 + 10	7	d	g	c
G19	I.TIJ/KS82142	2*	13 + 16	5 + 10	10	c	j	c
G20	NS46.11/3/SDY/TI.RESE1//KTA1/4/55.1744/MEX671//NO57/3/ATTILA	1	7 + 8	5 + 10	10	b	c	b
G21	BSP01/18 (DUZI)	2*	17 + 18	2 + 12	8	b	i	d
G22	PAMYAT	1	7 + 8	5 + 10	10	c	c	c
G23	KM75.4552/ZH93.51736	0	7 + 8	2 + 12	6	c	j	c
G24	OWL*2/SHIROODI	2*	7 + 8	5 + 10	10	c	h	b
G25	CH111.14422	2*	7 + 8	5 + 10	10	c	c	c

Glu-B1-7 + 8 and *Glu-B1*-13 + 19 had the highest mean score for PC (15.4%). In terms of EDEV, subunits *Glu-B1*-7 + 8, *GluB1*-7 + 9, *Glu-B1*-13 + 16, and *Glu-B1*-13 + 19 gave similar results while subunit *Glu-B1*-7 had the lowest score. Subunit *Glu-D1*-5 + 10 gave the highest mean scores for grain PC (12.4%), SV (28.4 mL), WG (28%), TW, and EDEV (112.4 cm^2), while subunits *Glu-D1*-2 + 12 had the lowest scores for all traits except grain PC and WG. Subunit *Glu-B1*-5 + 12 showed the highest performance for GY (6.24 t ha^{-1}).

LMW glutenin alleles at *Glu-A3* had a similar effect on GY and TW. The highest values were obtained from *Glu-A3-b* for PC (13.8%) and WG (30.8%), *Glu-A3-a* for SV (29.1 mL), and *Glu-A3*-d and *Glu-A3*-e for EDEV, indicating that these alleles had a positive effect on the mentioned traits. The *Glu-B3*-c allele had higher mean scores for grain PC (14%), SV (32.5 mL), and WG (31%), and subunits *Glu-B3*-b (110 cm^2), *Glu-B3*-c (114 cm^2), and *Glu-B3*-d (115.4 cm^2) had a higher mean for EDEV. *Glu-D3*-a and *Glu-D3*-b had the highest scores for SV (30.3 and 28.9 mL, respectively) and EDEV (130 and 100 cm^2, respectively). G19 and G23 (8%) harboring the 1B/1R translocation had lower scores for EDEV (59 cm^2), WG (27.2%), SV (20.8 mL), grain PC (12.5%), and GY (4.94 t ha^{-1}) but higher scores for TKW (39.8 g) and TW (81 kg/hL) compared to the means of quality traits of non-1B/1R genotypes.

Table 4 presents the results of the correlation analysis between the examined traits. According to these results, there was a significant and high correlation between grain PC and WG (r = 0.92), SV and EDEV (r = 0.82), TKW and TW (r = 0.53), and TW and quality score of HMW subunits. Another significant finding was the correlation between the quality score of HMW subunits and EDEV.

Table 3. Allele numbers (#) and frequencies (F) and statistical analysis of the effects of HMW-GS and LMW-GS on grain yield and quality traits.

	Alleles	(#)	F (%)	GY	PC	SV	WG	TKW	TW	EDEV
Glu-A1	Null	1	4	6.83	11.8	15	24.7	42.3	81.3	30
	2*	18	72	8.22	11.6	23.7	25.7	42.5	82.9	86.4
	1	6	24	8.83	12.1	23.2	26.7	41.7	81.5	67.2
Glu-B1	17 + 18	5	20	2.98	14.8	36.3	31.3	32.8	76.2	143
	7 + 8	11	44	3.03	15.4	34.6	32.7	32.8	75.9	120
	7 + 9	3	12	3.1	15	31.4	32.3	32.8	74.7	126
	7	3	12	2.92	14	31.6	28.9	35.5	75.7	108
	13 + 16	2	8	2.42	14.1	31.8	29.3	35.5	76.4	130
	13 + 19	1	4	3.55	15.4	34	28.5	33.2	73	131
Glu-D1	5 + 10	14	56	5.1	12.4	28.4	28	36.9	80.4	112.4
	5 + 12	1	4	6.24	11.1	27.8	26.7	38.8	80	98
	2 + 12	10	40	5.0	12.3	25.3	27.7	36.3	79.5	77.3
Glu-A3	a	1	4	5.42	12.9	29.1	28.6	36	78.3	64
	b	2	8	5.27	13.8	26.3	30.8	37.2	79.3	82
	c	15	60	5.4	13	28.3	28.6	37.8	79.9	106
	d	6	24	5.47	12.4	27.9	26.9	37.6	78.8	99.6
	e	1	4	5.77	13.5	28.7	28.2	35.3	77.7	103
Glu-B3	b	9	36	5.55	13.3	29.9	29.4	37.5	79.4	110
	c	1	4	4.77	14	32.5	31	35.2	79.6	114
	d	2	8	5.35	12.1	28	26	39.1	80.1	115.4
	e	3	12	5.52	12.7	26.1	27.7	37.3	78.9	95.4
	g	1	4	5.81	12	28.2	25.8	40.7	81.1	102
	h	2	8	5.21	13.3	27.3	28.3	36.4	78.9	95
	j	2	8	4.94	12.5	20.8	27.2	39.8	81	59
	i	5	20	5.48	13	28.4	28.2	36.4	78.6	98
Glu-D3	a	7	28	5.49	12.6	30.3	27.2	37.9	79.1	130
	b	5	20	5.41	13.3	28.9	29.3	35.8	78.2	100
	c	12	48	5.41	13	27	28.5	37.8	80	89
	d	1	4	5.2	13.8	21.5	30.5	39.4	80.4	46
1B/1R	+	2	8	4.94	12.5	20.8	27.2	39.8	81	59
	-	23	92	5.46	13	28.7	28.5	37.3	79.3	105
LSD (P < 0.05)				1.85	1.25	5.97	1.91	2.78	1.9	22.7

4. Discussion

In this study, the genotypes explained a higher proportion of total variation for SV and EDEV compared to environment (Table 1). These results indicate that the genotype effect is greater than the environment effect. Atlı and Koçak (2004) reported that SV has a high heritability value; thus, it can be used as a selection criterion in early generations of wheat breeding programs. Dough properties such as dough energy value and extensibility also have high heritability values, but they are used in advanced generations in breeding programs. In addition, a dough analysis method requires more labor and seed material (Labuschagne et al., 1996). For all examined traits except SV and EDEV, the effect of environment was greater than that of genotype and GEI, which means that these traits were largely affected by the environment. Kaya and Akcura (2014) found similar results indicating that the influence of the environment on GY, PC, WG, and TKW

Table 4. Correlation coefficients among the examined traits.

	GY	PC	SV	WG	TKW	TW	EDEV	QS
GY	1							
PC	–0.21	1						
SV	0.34	0.22	1					
WG	–0.07	0.92**	0.24	1				
TKW	–0.11	–0.13	–0.23	–0.25	1			
TW	–0.09	–0.16	–0.21	–0.15	0.53**	1		
EDEV	0.27	0.03	0.82**	–0.02	0.02	–0.05	1	
QS	0.03	0.43*	0.45*	0.47*	0.04	0.30	0.51**	1

GY: Grain yield, PC: grain protein content, SV: sedimentation volume, WG: wet gluten, TKW: 1000-kernel weight, TW: test weight, EDEV: extensograph dough energy value, QS: quality score of HMW glutenin subunits.

was greater than the genotypic effect. The proportions of the total explained variations for GY and TKW were higher through GEI as compared to the genotypic effect.

A correlation between different traits is generally due to the presence of linked genes and the epistatic effect of different genes. The environment plays an important role in such relationships. In some cases, the environment may affect traits either in the same direction or in different directions (Baloch et al., 2014). The results of the correlation analysis indicated that SV and quality scores of HMW glutenin subunits could be used to determine superior genotypes with desirable traits for high end-use quality such as increased dough energy value. This was also reported in several other studies (Horvat et al., 2006; Naghavi et al., 2009; Zhang et al., 2011). The significant and high correlation that was found between the quality score of HMW glutenin subunits and EDEV in the present study also indicates that these subunits are useful markers to improve the quality of genotypes to achieve higher end-use quality. Similarly, Bekes and Wrigley (2003) reported that HMW glutenin subunits have a considerable effect on bread-making quality traits; thus, they can be used as quality markers in breeding programs.

The effects of HMW and LMW glutenin subunits on GY and quality traits are shown in Table 3. These results indicate that the *Glu-A1* glutenin subunits had a significant effect on GY, SV, and EDEV. The genotypes possessing *Glu-A1*-2* and *Glu-A1*-1 displayed a high value for GY, SV, and EDEV while those with *Glu-A1*-null showed lower values for these traits (Table 3). Previous studies suggested that quality traits are correlated with *Glu-A1*-2* and *Glu-A1*-1 subunits and genotypes with null usually have poor quality scores (Martinez-Cruz et al., 2011). Similarly, Rasheed et al. (2014) reported that the presence of HMW glutenin subunits *Glu-A1*-1 and *Glu-A1*-2* combined with *Glu-D1*-15 + 10 had a positive effect on quality traits. Similarly, in the present study, we determined that PC, SV, and EDEV were influenced by *Glu-B1* alleles with subunit *Glu-B1*-17 + 18 having considerably higher scores for PC, SV, and EDEV. An interesting finding of this study was that the genotypes with subunits *Glu-B1*-13 + 16 and *Glu-B1*-13 + 19 had high scores for SV and EDEV. In the literature, there are limited data concerning the relationship between quality traits and *Glu-B1*-13 + 16 and *Glu-B1*-13 + 19 subunits; therefore, further studies would be indispensable for the detailed investigation of this topic and to confirm that these subunits can be used as markers in marker-assisted breeding programs. Yasmeen et al. (2015) reported that subunits *Glu-B1*-13 + 16 and *Glu-B1*-13 + 19 were observed in small numbers of Pakistani landraces. Earlier studies demonstrated that the frequency of *Glu-B1*-13 + 16 and *Glu-B1*-13 + 19 subunits is negligible in the global wheat collection and both these subunits are strongly related to good bread-making quality among the *Glu-B1* alleles (Ikeda and Takata, 2013). In contrast, Dessalegn et al. (2011) reported that *Glu-B1*-17 + 18 had a positive effect on dough properties and was more valuable than other subunits at *Glu-B1*. However, regarding the *Glu-B1*-13 + 16 and *Glu-B1*-13 + 19 subunits, there are many reports in the literature. Our results indicated both 13 + 16 and 17 + 18 have a positive effect on quality traits. Despite having the 13 + 19 and 13 + 16 alleles at *Glu-B1* and the 2 + 12 allele at *Glu-D1*, which are considered to be responsible for poor wheat quality, the bread wheat genotypes G13 and G16 exhibited the highest performance for EDEV, indicating that the 13 + 16 allele has a positive effect on the rheological traits of wheat flour. Further research is necessary to investigate alleles such as 13 + 16 and 13 + 19 that have low frequencies in bread wheat cultivars. Previous studies on HMW glutenin subunits reported that subunit *Glu-B1*-17 + 18 is more valuable than other

subunits at the *Glu-B1* locus (17 + 18 > 7 + 8 > 7 + 9) and has a positive effect on dough traits (Naghavi et al., 2009). Subunit *Glu-D1*-5 + 10 positively affected quality traits such as SV, PC, and EDEV compared to the *Glu-D1*-12 + 12 and *Glu-D1*-5 + 12 alleles. Several researchers previously reported that the 5 + 10 allele has a positive effect on bread-making quality traits while *Glu-D1*-2 + 12 is an indicator of poor quality (Pflüger et al., 2001; Rasheed et al., 2012). The general frequency of the *Glu-D1*-5 + 12 allele in bread wheat genotypes is low and there is limited information concerning its effect on quality traits. Aktaş (2014) reported that genotypes with subunit 5 + 12 have a higher score than *Glu-D1*-2 + 12 and lower score than *Glu-D1*-5 + 10 for SV, dough stability time, and dough energy value. Similar results were reported in several other studies, indicating that subunit *Glu-D1*-2 + 12 has a negative effect while *Glu-D1*-5 + 10 has a positive effect on end-use quality traits (Ammar et al., 2000; Horvat et al., 2006).

Data regarding the effect of LMW glutenin subunits on wheat quality traits are limited as compared to HMW subunits (Luo et al., 2001). Our results demonstrated that subunits c, d, and e at *Glu-A3*; d, b, c, and g at *Glu-B3*; and a and b at *Glu-D3* had positive effects on SV, PC, and EDEV. Genotypes with subunit d at *Glu-D3* showed poor performance for glutenin quality traits. Subunit j at *Glu-B3* associated with 1B/1R translocation had low scores for EDEV, which means that genotypes with this subunit could not be used in breeding programs. He et al. (2005) reported that *Glu-B3*-d had a slightly higher positive effect on dough properties and gluten quality. Previous studies on LMW glutenin subunits suggested that *Glu-A3*-b, *Glu-A3*-d, *Glu-B3*-d, *Glu-B3*-g, and *Glu-D3*-b were correlated with superior dough properties (Zhang et al., 2011; Rasheed et al., 2014).

According to the results of the present study, the genotypes with 1B/1R translocation had high scores for TKW and TW while their EDEV and SV were very low. These results indicate that the 1B/1R translocation has a negative effect on quality traits, especially for EDEV. Several authors reported that genotypes with 1B/1R score higher in GY, TKW, TW, and SV; however, dough made from their flour has poor quality due to their low EDEV and farinograph stability time (Liu et al., 2005; Yediay et al., 2010; Rasheed et al., 2014; Yasmeen et al., 2015).

In conclusion, the presence of *Glu-A1*-2*, *Glu-B1*-17 + 18, and *Glu-D1*-5 + 10 in HMW glutenin subunits and *Glu-A3*-d, *Glu-B3*-d, and *Glu-D3*-b in LMW subunits positively contributes to quality traits, indicating that these alleles can be used as selection markers to improve the quality of genotypes in wheat breeding programs. Similarly, an interesting result of the study was that subunit 13 + 16 at *Glu-B1* presented as a useful allele for achieving high-quality traits and therefore its frequency in breeding materials should be increased. Furthermore, genotypes G5, G7, G8, G9, and G12 had a higher yield with better protein quality and content compared to wheat varieties in Turkey. Finally, the wheat lines investigated in this study have great potential for developing new varieties with higher yield and better quality if used as parents in breeding programs.

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