



Article

Phenotypic Variability, Heritability and Associations of Agronomic and Quality Traits in Cultivated Ethiopian Durum Wheat (*Triticum turgidum* L. ssp. *Durum*, Desf.)

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Abstract: Quality is an important aspect of durum wheat in the processing sector. Thus, recognizing the variability of quality and agronomic traits and their association is fundamental in designing plant breeding programs. This study aimed to assess the variability, heritability, genetic advance, and correlation of some agronomic and quality traits among 420 Ethiopian durum wheat genotypes and to identify the promising genotypes with distinct processing quality attributes to produce superior quality pasta. The field experiment was conducted at two locations (Sinana and Chefe Donsa) using an alpha lattice design with two replications. Analysis of variance, chi-square test, and Shannon–Weaver diversity index revealed the existence of highly significant ($p < 0.001$) variation among genotypes for all studied traits. The broad-sense heritability values were ranging from 46.2% (days to maturity) to 81% (thousand kernel weight) with the genetic advance as a percent of the mean ranging from 1.1% (days to maturity) to 21.2% (grain yield). The phenotypic correlation coefficients for all possible pairs of quantitative traits showed a significant ($p < 0.05$) association among most paired traits. The gluten content (GC) and grain protein content (GPC) were negatively correlated with grain yield and yield-related traits and positively associated with phenological traits, while yield and phenological traits correlated negatively. The frequency distributions of amber-colored and vitreous kernels, which are preferable characters of durum wheat in processing, were highly dominant in Ethiopian durum wheat genotypes. The identified top 5% genotypes, which have amber color and vitreous kernel with high GC and GPC content as well as sufficient grain yield, could be directly used by the processing sector and/or as donors of alleles in durum wheat breeding programs.



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1. Introduction

Durum wheat (*Triticum turgidum* L. ssp. *Durum*, Desf.) is one of the most commonly cultivated cereal crop globally with a production of 33.8 million tons in 2019/20, which is 5% of the global wheat production [1]. Ethiopia is considered a center of durum wheat diversity [2–5] where the crop has been cultivated in small-to-medium-scale farming systems in diverse agro-ecological and climatic conditions [6,7]. Although durum wheat has a lower grain yield potential than bread wheat [8], it continues to be the crop of choice for smallholder farmers. Major factors contributing to its preference include its wide adaptation to diverse climatic conditions [9,10], a higher price than bread wheat, millet,

maize, and sorghum [11], and high straw yield for livestock feed [12]. Additionally, the expanding value chain of pasta on the global market has contributed to its preference for production [13].

The major significance of durum wheat lies in its characteristics of producing premium quality pasta, macaroni, and related products. The characteristics that made durum wheat preferable for making superior quality pasta include its grain hardness, yellow pigmentation, vitreousness, dough elasticity, dough stability, and dough strength, as well as the pasta's firmness and taste [14,15]. The grain texture and protein content have a significant impact on milling, processing, and end-use quality [16]. Among proteins, the gluten content and quality are crucial in processing or pasta making because gluten confers elasticity and viscosity to the dough [17].

Phenotypic diversity analysis is useful for determining the genetic diversity of crop genetic resources, a prerequisite for successful crossbreeding [18,19], and these results may assist in the design of efficient breeding programs [20]. Crops' genetic diversity provides an opportunity to develop cultivars well adapted to a specific environment [21] as well as cultivars with broad adaptation. An efficient plant breeding strategy utilizes crop genetic diversity as a source of novel alleles for crop improvement. This is crucial in order to face the unpredictably changing climates, improve production, and meet new end-user demands [22,23]. In line with this, information on genetic variability, heritability, genetic advance, and correlation constitutes one of the most desirable parameters that are widely used as prerequisites for making a wise parental selection in designing breeding programs [24]. Heritability and genetic advance are parameters useful to predict the response of succeeding generations and estimate the magnitude of genetic gain through selection [25], and the correlation of different traits helps to make the decision of direct or indirect selection [26].

The Ethiopian Biodiversity Institute's (EBI) gene bank has ex situ conserved over 7000 durum wheat landrace accessions [27]. To uncover the genetic diversity of these landraces, a limited number of studies have been conducted previously and identified a high level of variability for phenotypic and quality traits [27–29]. However, the existing genetic variation in the Ethiopian gene pool has not been sufficiently utilized for the improvement of desirable quantitative and qualitative traits [30]. According to the pedigree records of modern durum wheat varieties released in Ethiopia, its breeding programs have primarily utilized genotypes originating from the Mediterranean region, Eastern Europe, and North America [31], while the Ethiopian germplasm has been underutilized. Recently, however, Ethiopian durum wheat landraces have been considered sources of desirable traits in international breeding programs [32].

Although Ethiopia has a large number of durum wheat cultivars and landraces under production and is currently producing 90% of durum wheat cultivated in sub-Saharan Africa [13], the grain yield and protein quality are unsatisfactory. Consequently, the country imports high-quality durum wheat grains, pasta, and other related products [8]. In addition, introduced exotic cultivars have frequently failed to adapt to the wide-ranging agro-ecological and climatic conditions in Ethiopia [30]. The identification of Ethiopian durum wheat genotypes that meet the processing quality guidelines or that can be used as sources of alleles for the development of suitable cultivars for the processing sector is crucial to satisfy domestic and international demands. Therefore, this study aimed to assess the extent of genetic variation among Ethiopian durum wheat genotypes, to estimate the magnitude of heritability, and genetic advance, as well as the correlation among agro-morphological and quality traits. It also aimed to identify promising durum wheat genotypes with favorable grain quality attributes for making superior pasta.

2. Materials and Methods

2.1. Plant Materials and Experimental Design

A total of 420 durum wheat "genotypes" were used in this study (Table S1). Of these, 386 are landrace accessions obtained from the Ethiopian Biodiversity Institute (EBI) that

were originally collected from different geographic regions in Ethiopia (Figure 1) whereas the remaining 34 are cultivars obtained from the Debrezeit agricultural research center (DzARC) and Sinana Agricultural Research Center (SARC), both of which are part of the Ethiopian Institute of Agricultural Research (EIAR) (Table S1). Landraces collected from nearby geographical zones were considered as an individual population to proportionate the number of samples per population.

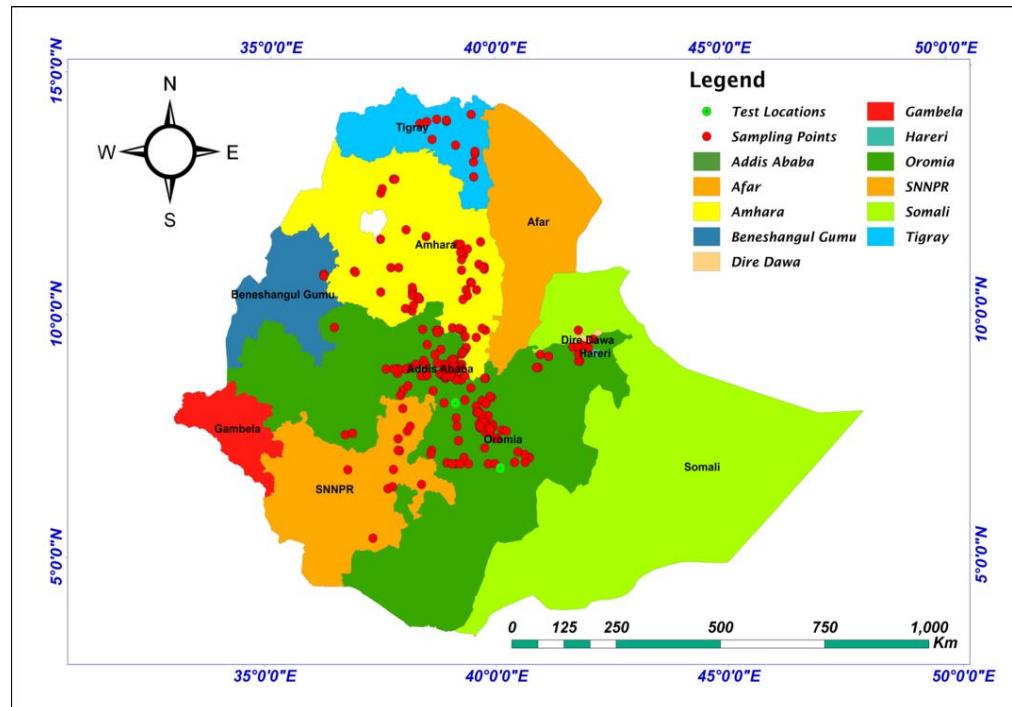


Figure 1. Map of Ethiopia showing the sample collection sites of landrace accessions and field trial sites.

The field experiment was conducted at two sites in Ethiopia, Sinana ($07^{\circ}07'N$ and $40^{\circ}10'E$, 2400 m.a.s.l) and Chefe-Donsa ($08^{\circ}44'N$ and $39^{\circ}09'E$, 2450 m.a.s.l), during the 2019/2020 main crop growing season using an alpha lattice design with two replications. The soil types at both Sinana and Chefe Donsa research centers were vertisols with minor light soils. The genotypes were randomly assigned to plots within each block. The area of each plot was 0.4 m^2 (2 rows with 1 m length and 0.4 m width) and the spacing between entry, plants, blocks, and replications was 0.4 m, 0.1 m, 1 m, and 2 m, respectively. Fertilizer was applied at a rate of 100 Kg/ha of DAP and 50 Kg/ha of urea at the planting and jointing stages. All other management practices recommended for wheat were also applied to all experimental plots.

2.2. Data Collection

Data were collected for the following phenotypic traits: days to heading (DH), days to maturity (DM), number of spikelets per spike (SPS), spike length (SPL), plant height (PLH), thousand kernel weight (TKW), grain yield (GY), lodging (LDG), spike density (SPD), glume hairiness (GLH), glume color (GLC), kernel color (KCL), vitreousness (VTR), grain moisture content (GMC), gluten content (GC) and grain protein content (GPC). The SPS, PLH, LDG, SPD, GLH, GLC, KCL, and VTR traits data were collected using the descriptor list for wheat [33] whereas DH, DM, and SPL traits data were collected following the description provided in Hailu et al. [34]. TKW (the weight of 1000 kernels in grams) was calculated by multiplying the weight of 250 kernels by four, and GY was estimated based on the weight (in grams) of the total seed harvest per plot. GMC, GC, and GPC were

measured using Near-Infrared Spectroscopy (NIRS) as described in AOAC [35] method 997.06 (32.2.03A). The values of GY and TKW were standardized to 12% moisture content.

2.3. Statistical Analysis

Analysis of variance and variance components: prior to statistical analysis, the collected quantitative data were checked for normal distribution using the Shapiro–Wilk test through the shapiro.test function of the Stats package [36] in R software. The effects of genotype, replication, block, location, and genotype by location interaction were tested via analysis of variance (ANOVA) using the aov function of the Stats package [36] based on the alpha lattice design linear additive model.

$$P_{ijk} = \mu + \tau_i + \pi_j + b_k + \xi_{ijk} \quad (1)$$

where P_{ijk} is the phenotypic value of i th genotype under j th replication and k th block, μ is the grand mean, τ_i is the effect of i th genotypes, π_j is the effect of j th replication, b_k is the effect of k th block and ξ_{ijk} is residual error.

The phenotypic, genotypic, and genotype by location interaction variances were computed as described by Singh and Chaudhary [37]:

$$\text{Genotypic variance} = \sigma_g^2 = \frac{MS_g - MS_{g \times 1}}{rl} \quad (2)$$

$$\text{Genotype} \times \text{Location interaction variance} = \sigma_{g \times 1}^2 = \frac{MS_{g \times 1} - MS_e}{r} \quad (3)$$

$$\text{Phenotypic variance} = \sigma_p^2 = \sigma_g^2 + \frac{\sigma_{g \times 1}^2}{l} + \frac{\sigma_e^2}{rl} \quad (4)$$

where MS_g is the mean square of genotype, $MS_{g \times 1}$ is the mean square due to genotype by location interaction, MS_e is the error mean square, r is the number of replications and l is the number of locations.

The genotypic and phenotypic coefficient of variation was calculated as described in Johnson et al. [38]:

$$\text{Genotypic coefficient of variation} = GCV = 100 \times \sigma_g / \mu \quad (5)$$

$$\text{Phenotypic coefficient of variation} = PCV = 100 \times \sigma_p / \mu \quad (6)$$

where σ_g and σ_p are genotypic and phenotypic standard deviations, respectively, and μ is the grand mean. The GCV and PCV values were categorized as high (>20%), medium (10–20%) and low (<10%) as classified by Burton and DeVane [39].

Heritability and genetic advance: broad-sense heritability (H_b) was calculated using the formula presented in Allard [40]:

$$\text{Broad sense heritability} = H_b = \left(\sigma_g^2 / \sigma_p^2 \right) \times 100 \quad (7)$$

where σ_g^2 and σ_p^2 are genotypic and phenotypic variance, respectively. The values of broad-sense heritability were classified as low ($H_b < 25\%$), moderate ($25\% < H_b < 50\%$), and high ($H_b > 50\%$).

The expected genetic advance and genetic advance as a percent of the mean were estimated as described in Johnson et al. [38]:

$$\text{Genetic advance (GA)} = k \times H_b \times \sigma_p \quad (8)$$

$$\text{GA as percent of mean (GAM)} = (GA / \mu) \times 100 \quad (9)$$

where σ_g and σ_p are genotypic and phenotypic standard deviations, respectively, k is a constant value at a selection intensity of 5% ($k = 2.06$); H_b is broad-sense heritability, GA is genetic advance and μ is the grand mean.

The GAM is considered low, moderate, and high when its values are below 10%, 10–20%, and above 20%, respectively, as described in Falconer and Mackay [41].

Correlation between traits: The Pearson correlation coefficient (r) between quantitative phenotypic traits was computed using the chart.Correlation function of the PerformanceAnalytics package [42] in R.

Frequency distribution, chi-square test and Shannon diversity index of qualitative data: the qualitative data were transformed into frequency data and subjected to the chi-square test using prob.table and chisq.test function of the Stats package [36]. The standardized Shannon–Weaver diversity index (“ H' ”) was calculated from the frequency data of qualitative traits as described in Eticha et al. [43]:

$$H' = \left(\sum_{i=1}^R p_i \ln(p_i) \right) / \ln(n) \quad (10)$$

where n is the number of phenotypic classes of the trait and p_i is the proportion of i th phenotypic class. H' was estimated for all qualitative traits and all genotypes.

The Shannon diversity index was regarded as high ($H' \geq 0.60$), intermediate ($0.40 \leq H' < 0.60$) and low ($H' < 0.40$) as described in Eticha et al. [43].

3. Results

3.1. Phenotypic Variation among the Genotypes

A wide range of variation was observed for most of the quantitative phenotypic traits (Table 1). The mean GPC and GC values of the genotypes were 13.4% and 25.7%, respectively (Table 1). The optimal GC for pasta and related products is 28% and above, according to Sissons [15]. Interestingly, 97 landraces of the 386 landraces (25.1%) had a GC value 28% and above, with genotypes 364, 150, 191, 287, 354, 263, 282, 285, 294, 67, 197, and 276 having GC values above 32%. On the other hand, all cultivars had a gluten content of below 28%. Likewise, 285 genotypes had a grain protein content (GPC) of 13%, which is the lower limit for pasta production [44], with genotypes 208, 190, 386, 191, 364, 282, 373, 354, 285, 247, 224, 110, 263, 287, 205, 25, 272, and 115 having a GPC of above 15%. In total, 208 genotypes (49.5%) produced grain yields exceeding the mean value of 336.4 g of seeds per plot (gm/pt), of which 48 genotypes (11.4%) produced grain yields exceeding 400 g per plot (Table 1). One hundred and seventy-four genotypes (41.4%) had a TKW above the mean value of 42.4 g, of which 34 genotypes had a TKW above 50 g (Table 1).

Table 1. Descriptive statistics (minimum, maximum and mean values) for 11 phenotypic traits investigated over combined locations and their corresponding estimates of genotypic variance (σ_g^2), environmental variance (σ_e^2), phenotypic variance (σ_p^2), genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV), broad-sense heritability (H_b), genetic advance (GA), and mean genetic advance (GAM).

Traits	Mean	Range		σ_e^2	σ_{g+1}^2	σ_g^2	σ_p^2	GCV	PCV	H_b (%)	GA	GAM (%)
		Min	Max									
DH	71.4	61.8	78.0	6.0	0.8	6.2	8.1	3.5	4.0	76.5	4.5	6.3
DM	137.8	134.0	145.2	5.1	0.6	1.1	2.7	0.8	1.2	42.6	1.4	1.1
SPS	19.5	16.5	22.3	1.8	0.3	0.5	1.1	3.5	5.3	44.2	0.9	4.8
SPL	8.6	5.3	11.5	1.2	0.3	0.9	1.4	11.2	13.5	68.5	1.6	19.1
PLH	108.7	75.8	138.5	72.0	5.5	23.3	44.0	4.5	6.1	52.9	7.2	6.7

Table 1. Cont.

Traits	Mean	Range		σ_e^2	σ_{g*1}^2	σ_g^2	σ_p^2	GCV	PCV	H_b (%)	GA	GAM (%)
		Min	Max									
TKW	42.4	32.0	55.4	7.4	3.4	15.0	18.6	9.2	10.2	81.0	7.2	17.0
GY	336.4	171.2	505.6	3723.0	1465.0	2127.3	3790.0	13.7	18.3	56.1	71.2	21.2
GC	25.7	17.5	35.4	10.0	3.0	5.0	9.0	8.7	11.6	55.6	3.4	13.4
GPC	13.4	11.5	16.2	1.0	0.3	0.3	0.7	4.3	6.4	44.8	0.8	5.9

DH = days to heading; DM = days to maturity; SPS = the number of spikelets per spike; SPL = spike length; PLH = plant height; TKW = thousand kernel weight; GY = grain yield; GC = gluten content and GPC = grain protein content.

3.2. Analysis of Variance (ANOVA)

The ANOVA has revealed significant ($p < 0.05$) effects of most phenotypic traits on Ethiopian durum wheat genotypes with R^2 values ranging from 0.67 (DM) to 0.89 (DH) (Table 2). Highly significant ($p < 0.001$) differences among genotypes were observed for all phenotypic traits. The test locations (environments), as well as the genotype by location interaction (genotype by environment interaction; G × E), also had significant ($p < 0.05$) effects on all traits (Table 2).

Table 2. Combined analysis of variance for 11 traits of 420 durum wheat genotypes tested at two locations (Sinana and Chefe Donsa) during 2019/20.

Trait	Model	GEN	LOC	GEN: LOC	REP	BLK	Error	CV (%)	LSD	R ²
df	880	419	1	419	1	40				
DH	44.9 ***	31.0 ***	19,632 ***	8.0 *	73 ***	89.0 ***	6.0	3.3	3.4	0.89
DM	9.1 ***	9.6 ***	298.4 ***	6.1 *	24.29 *	26.9 ***	5.1	1.2	2.7	0.67
SPS	6.9 ***	3.7 ***	3159.8 ***	2.4 ***	31.1 ***	9.4 ***	1.8	6.7	1.4	0.81
SPL	5.4 ***	4.9 ***	1358.8 ***	1.7 ***	173 ***	11.9 ***	1.2	11.7	1.4	0.83
PLH	282.8 ***	165 ***	116,913 ***	83.0 *	3635 ***	612.0 ***	72.0	7.4	9.3	0.81
TKW	45.8 ***	67.5 ***	172.7 ***	14.1 ***	387 ***	138.3 ***	7.4	6.1	4.7	0.87
GY	9376 ***	12,232 ***	17,319 *	6653.0 ***	48 ns	7947 ***	3723.0	16.7	72.0	0.74
GC	41.6 ***	30.0 ***	14,050 ***	16.0 ***	68 *	85.0 ***	10.0	10.8	3.8	0.82
GPC	5.6 ***	2.3 ***	2774.2 ***	1.6 ***	19.6 ***	12.3 ***	1.0	6.6	1.1	0.86

DH = days to heading; DM = days to maturity; SPS = the number of spikelets per spike; SPL = spike length; PLH = plant height; TKW = thousand kernel weight; GY = grain yield; GC = gluten content and GPC = grain protein content. df = degree freedom; GEN = genotype; LOC = location; REP = replication; BLK = block; LSD = the least significant difference and CV = coefficient of variation. “***” = significant at $p < 0.001$; “**” = significant at $p < 0.05$; “ns” = non-significant at $p < 0.05$.

3.3. Estimation of Components of Variability

Medium GCV and PCV values (10–20%) were recorded for SPL (11.2% and 13.5%, respectively) and GY (13.7% and 18.3%, respectively) (Table 1). However, the remaining traits had low GCV and PCV values (<10%) except PCV for TKW (10.2%) and GC (11.6%) (Table 1).

3.4. Broad-Sense Heritability and Genetic Advance

The values of broad-sense heritability were high to intermediate for all traits, which ranged from 42.6% for DM to 81% for TKW (Table 1). Within this range, high heritability values ($H_b > 50\%$) were observed for TKW (81%), DH (76.5%), SPL (68.5%), GC (55.6%), and PLH (52.9%), whereas moderate heritability values (25% $< H_b < 50\%$) were recorded for the remaining traits. GA is a useful selection parameter in addition to heritability for crop improvement. The GAM values for most of the traits were low except for GY (21.2%), SPL (19.1%), and GC (13.4%), which can be regarded as moderate (Table 1).

3.5. Correlation Analysis among Quantitative Traits

The phenotypic correlation among most pairs of quantitative traits was significant ($p < 0.05$) (Figure 2). GC demonstrated a highly significant ($p < 0.001$) positive correlation

with GPC ($r = 0.71$), SPL ($r = 0.19$), PLH ($r = 0.23$) and significant positive correlation ($p < 0.05$) with DM ($r = 0.10$). On the other hand, it was negatively correlated with SPS ($r = -0.22$), and GY ($r = -0.25$) ($p < 0.001$). GPC demonstrated highly significant positive correlations ($p < 0.001$) with DM ($r = 0.18$), SPL ($r = 0.44$), PLH ($r = 0.17$) and GC ($r = 0.71$). It also showed significant positive correlation with DH ($r = 0.14$; $p < 0.01$). In contrast, GPC was negatively correlated with GY (-0.22 ; $p < 0.001$), and TKW (-0.11 ; $p < 0.05$).

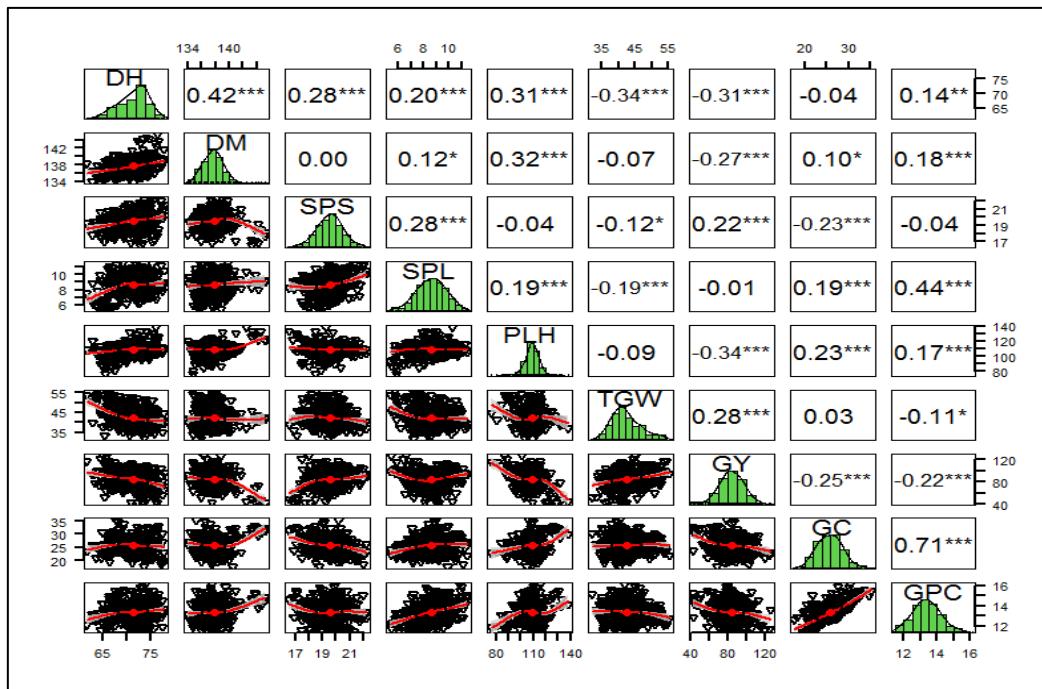


Figure 2. Phenotypic (above diagonal) correlation coefficients, distribution (diagonal), and scatterplot with regression curve (below diagonal) of 11 quantitative traits for the 420 durum wheat genotypes. Critical values for Pearson's correlation coefficient are 0.1002 at 0.05%, 0.13 at 0.01% and 0.17 at 0.001. *** = significant at $p < 0.001$; ** = significant at $p < 0.01$; * = significant at $p < 0.05$.

GY showed highly significant ($p < 0.001$) positive correlations with TKW ($r = 0.28$) and SPS ($r = 0.20$), and highly significant ($p < 0.001$) negative correlations with DH ($r = -0.31$), DM ($r = -0.27$), PLH ($r = -0.34$), GC ($r = -0.25$) and GPC ($r = -0.22$) (Figure 2). TKW showed significant ($p < 0.01$) negative correlations with DH ($r = -0.34$), SPS ($r = -0.13$), SPL ($r = -0.19$) and GPC ($r = -0.11$; $p < 0.05$). TKW only showed a significant positive correlation with GY ($r = 0.28$; $p < 0.001$).

3.6. Frequency Distribution and Diversity of Qualitative Traits

For data analysis, five of the six qualitative traits (SPD, GLC, GLH, KCL, and VTR) were classified into three phenotypic classes, while LDG was classified into four classes (Table 3). In terms of lodging, 47.8% of the genotypes were lodging types with medium to complete lodging. The vast majority (92.3%) of the genotypes had hairless glumes. Interestingly, about 70% and 76.4% of the genotypes had amber kernel color and vitreous kernel, respectively, which are among the most important processing quality traits. The chi-square test revealed highly significant ($p < 0.05$) variation among the genotypes in terms of all qualitative traits with Shannon diversity index (H') ranging from 0.27 (GLH) to 0.96 (SPD) (Table 3). Hence, the diversity of each trait is high ($H' > 0.60$) for all qualitative traits except for GLH ($H' = 0.27$). Test locations (environments) had a significant effect ($p < 0.05$) on LDG and GLC but not on the other qualitative traits (Table 3).

Table 3. Summary of chi-square (χ^2) test and Shannon diversity index (H') for six qualitative traits analyzed based on data collected from 420 durum wheat genotypes grown at two locations.

Trait	Phenotypic Class		Prop. (%)	χ^2		H'
	Name	Size		GEN	LOC	
LDG	No	4	17			
	Low		35.4			
	Medium		42	1638 ***	32.9 ***	
	Complete		5.6			0.87
SPD	Lax	3	32.9			
	Intermediate		45.8	2059 ***	3.24 ns	
	Dense		21.3			0.96
GLC	White	3	61.8			
	Red-brown		34.1	1974 ***	26.5 ***	
	Black		4.1			0.73
GLH	Hairless	3	92.3			
	Less		1.1	1722 ***	5.1 ns	
	High		6.6			0.27
KCL	Amber	3	70.3			
	Brown		12.4	2205 ***	0.05 ns	
	Purple		17.3			0.74
VTR	Non-vitreous	3	5.8			
	Partly vitreous		17.8	1744 ***	0.02 ns	
	Vitreous		76.4			0.62

LDG = lodging; SPD = spike density; GLC = glume color; GLH = glume hairiness, KCL = kernel color, VTR = vitreousness, SN = Sinana; CDR = Chefe Donsa, GEN = genotype; REP = replication; LOC = location. *** = significant at $p < 0.001$; "ns" = non-significant at $p < 0.05$.

3.7. The Performance of Selected Genotype

First, genotypes having vitreous grains with amber kernel color were selected (Table S2), and then the top 5% of the total genotypes analyzed were selected based on their GC and GPC content by also taking TKW and grain yield into consideration. Because of the highly negative correlation between GC and GPC with grain yield and grain yield-related traits, genotypes with better TKW and GY were prioritized. The difference between the grand mean and the mean of the selected genotypes was significant ($p < 0.05$) for GC and GPC, but not for the other quantitative traits (Table 4). The GC mean value for the top 5% genotypes (30.5%) was greater by 4.8% than the grand mean of all genotypes (25.7%) (Table 4), thus meeting the highest grain quality grade as per the description of Sissons [15]. Interestingly, the mean GPC for the top 5% genotypes was 14.4%, exceeding the minimum threshold (13%) required for high processing quality [44]. It should be noted that none of the released cultivars are part of these top 5% genotypes.

Table 4. Comparisons of the mean values of the top 5% genotypes (\bar{x}) with the mean value of the 420 genotypes (μ) for the nine quantitative traits.

Trait	M	\bar{x}	$\mu - \bar{x}$	$\%(\mu - \bar{x})$	t-Test
DH (day)	71.4	70.0	1.4	2	-0.81 ns
DM (day)	137.8	137.6	0.2	0.2	-0.23 ns
SPS	19.5	19.4	0.1	0.6	-0.21 ns
SPL (cm)	8.6	9.2	0.6	6	0.90 ns
PLH (cm)	108.7	108	0.7	0.7	-0.25 ns

Table 4. Cont.

Trait	M	\bar{x}	$\mu - \bar{x}$	$\%(\mu - \bar{x})$	t-Test
TKW (g)	42.4	42.2	0.2	0.5	-0.07 ns
GY (gm/pt)	336.4	354	17.6	5	0.59 ns
GC (%)	25.7	30.5	4.8	15.7	3.78 ***
GPC (%)	13.4	14.4	1.0	6.9	2.43 **

DH = days to heading; DM = days to maturity; SPS = the number of spikelets per spike; SPL = spike length; PLH = plant height; TKW = thousand kernel weight; GY = grain yield; GC = gluten content and GPC = grain protein content. t-value (0.05) ($df = 41$) = 1.68. “***” = significant at $p < 0.001$; “**” = significant at $p < 0.01$; “ns” = non-significant at $p < 0.05$.

4. Discussion

4.1. Variations of Quantitative Traits among Genotypes

A thorough understanding of genetic variation within a crop gene pool, as well as genetic relationships between germplasm of different origins, will facilitate the development of new cultivars with desirable traits. Our understanding of the heritability and correlations of their traits is also imperative. These can be gained through phenotypic studies based on agro-morphological and quality traits. In the present study, nine quantitative and six qualitative traits of durum wheat were analyzed, revealing a wide range of phenotypic variation among the genotypes in terms of most of the traits studied. This suggests the presence of high genetic variation within the Ethiopian durum wheat gene pool. Previous studies on durum wheat also reported a wide phenotypic variation for GPC, TKW, and GY [30,45–47]. Among the 420 genotypes studied, 285 of them meet the minimum threshold value of 13% GPC content necessary for making high-quality pasta and related products [44]. Interestingly, 68 of the 285 genotypes performed well at both experimental sites and were therefore stable across environments, suggesting their abilities to adapt to various agro-ecological conditions.

The grain gluten content of durum wheat is also crucial in the processing for making pasta and related products because gluten confers elasticity and viscosity to the dough [17]. Ninety-seven genotypes had grain gluten content of above 28% with the highest values recorded in genotypes 364, 150, 191, 287, 354, 263, 282, 285, 294, 67, 197, and 276, which were also characterized by having high grain protein content, delayed days to maturity, high plant height, long spike, high thousand kernel weight, and intermediate grain yield. All cultivars evaluated in this study had less than 28% grain gluten. This is mainly because this trait has not been seriously taken into consideration during the breeding process, which focused mainly on grain yield improvements. Hence, crossbreeding that involves genotypes with high gluten content, on the one hand, and high-yielding genotypes, on the other hand, is crucial in order to develop high-yielding cultivars with desirable levels of grain gluten content. Although the genotype by location interaction ($G \times E$) was significant for GC, 24 landraces scored GC above 28% at both locations, suggesting a wide adaptation of these genotypes. Therefore, they should be considered in breeding programs for durum wheat as sources of desirable agro-morphological and quality traits.

The highly significant ($p < 0.01$) phenotypic variation among genotypes for most of the studied traits indicates the presence of an adequate amount of genetic variation among characterized genotypes. In line with the results of the present study, Mengistu and Pe [27] found significant differences among 49 Ethiopian durum wheat genotypes (comprising 45 landraces and 4 cultivars) in DM, PLH, TKW, and GY. Similarly, Wolde et al. [47] and Mengistu et al. [48] reported a significant variation among 68 and 289 Ethiopian durum wheat genotypes, respectively, in DM, PLH, TKW, and GY. Studies on durum wheat genetic resources of non-Ethiopian origins have also demonstrated significant variation among genotypes in GC, GPC, GY, TKW, and PLH [46,49,50]. These indicate that durum wheat has the potential for better adaptation to different agro-ecological conditions.

The highly significant ($p < 0.01$) $G \times E$ interaction for all quantitative traits obtained in this study and similar results of previous reports for GPC, TKW, and GY [50,51] indicate

that genotypes differ in their adaptability to different agro-ecological conditions, with some genotypes having excellent adaptation to specific environments.

For all traits, except GY, relatively small differences were observed between genotypic and phenotypic coefficients of variation. This indicates that genotypes have a more profound influence than environmental factors on the phenotypic expressions of these traits in durum wheat. Similarly, relatively smaller differences between GCV and PCV values were reported by Mohammed et al. [52] for TKW and GPC, and Wolde et al. [47] and Alemu et al. [53] for DH, DM, PLH, and TKW. These suggest a lower effect of environmental factors on phenological and quality traits. On the other hand, the relatively large differences between GCV and PCV values for GY obtained in the present study and that of Alemu et al. [53] indicate that grain yield is highly influenced by environmental factors.

4.2. Selection Efficiency of Quantitative Traits in Durum Wheat

The determination of the heritability and genetic advance of traits of interest in crops allows the selection of suitable parents for crossbreeding [24], prediction of the performance of progenies in successive generations, and estimation of the degree of genetic gains obtained from the selection [25]. Assuming 5% selection intensity, high heritability combined with high to moderate genetic advance indicates the presence of additive gene effects [54,55] while high heritability coupled with low genetic advance suggests the presence of non-additive gene effects for a given trait [55,56]. The results of the present study indicate the presence of strong additive gene effects for GC, TKW, and GY, which suggests the importance of undertaking effective phenotype-based selection at the early generation of breeding during cultivar development. The additive gene effect was also previously reported for TKW [46,47,51,52]. The influence of the additive gene effect on GY identified in this study was also reported by Wolde et al. [47], and hence early generation selection could be effective. In contrast, Alemu et al. [53], as well as Mengistu and Pe [27] reported a non-additive gene effect on GY, which could be attributed to the small numbers of genotypes (64 and 49 durum wheat landraces, respectively) they used.

In the present study, plant height was negatively correlated with grain yield, which may suggest a trade-off between the two traits in durum wheat. However, PLH was positively correlated with both grain gluten and protein contents (Figure 2). It may be difficult, therefore, to select genotypes using PLH as a selection criterion for crossbreeding with the aim of producing high-yielding cultivars with desirable levels of grain gluten and protein content. Hence, introducing dwarfing genes through modern breeding approaches could improve the grain yield of genotypes along with desirable levels of grain gluten and protein content [57].

The significant influence of the environment on the expression of protein traits lowered the heritability of both GC (55.6%) and GPC (44.8%) (Table 1). Based on the moderate heritability and low genetic advance obtained for GPC in this study, it appears that there is a non-additive gene effect, which is in accordance with previous studies [52,53], so it should be noted that selection for GPC may be complicated both in the early and advanced generations. The high heritability with a moderate GAM value of GC indicates the presence of an additive gene effect on the trait. This suggests the possibility of undertaking effective genotype selection through phenotypic expression at early generation and enables the development of effective breeding strategies for improving GC in durum wheat. Previously, there were limited numbers of researches conducted to determine the heritability value of GC in durum wheat. For instance, Brankovic et al. [45] and Mohammed et al. [52] reported high and moderate, respectively, heritability and moderate GA for wet gluten content.

4.3. Associations of Quantitative Traits in Durum Wheat

An understanding of the associations between different traits is a crucial step in crop breeding for both improving target traits simultaneously and indirectly selecting other desirable traits. The significant negative correlation between GPC and GY obtained in this study is in agreement with that of Alemu et al. [53], Sourour et al. [49], and Bilgin et al. [58].

However, Graumann et al. [59] found a significant positive correlation, which deviates from the commonly reported negative correlation between the two traits. The significant negative correlation between GPC and TKW obtained in the present study has also been reported previously [59–61]. In contrast, Mohammed et al. [52] found a highly significant positive correlation ($p < 0.01$) between these traits. Sourour et al. [49] reported a significant positive correlation ($p < 0.05$) of GC with PLH, SPL, SPS, and GPC, while negatively correlated with GY. Similarly, Bilgin et al. [58] found a significant positive correlation ($p < 0.05$) of GC with GPC and a significant negative correlation ($p < 0.05$) with TKW and GY. These results suggest that grain yield tends to correlate negatively with processing quality traits in durum wheat landraces. Hence, breeding for the improvement of processing quality traits may lead to a decrease in grain yield. Hence, exploring the diverse durum wheat gene pool to identify genotypes with high GPC and GC along with moderate to high TKW and GY that can be used in a breeding program is a crucial step for avoiding the trade-off between the two groups of traits.

4.4. Distribution and Variability of Qualitative Traits among Genotypes

The proportion of phenotypic classes of the studied qualitative traits was variable among Ethiopian durum wheat genotypes (Table 3). The existence of technologically important phenotypic characters in high frequency in Ethiopian durum wheat genotypes allows them to be valuable for the processing sector. Among these traits, amber kernel color and vitreous kernel classes are dominant in the studied genotypes. Similarly, Hailu et al. [34] reported the dominance of amber kernels among tetraploid wheat. However, Mengistu et al. [30] described the rarity of amber kernels in Ethiopian durum wheat landraces. The distinct genotypes with a purple kernel color observed in this study were also reported by Bechere et al. [28], which accounted for about 16% of their collections. In addition, Zeven [62] identified that genotypes with purple kernels are endemic to Ethiopia, and hence classified as *Triticum turgidum* sub. *aethiopicum* [43].

The GLH traits showed lower diversity than other qualitative traits with the dominance of the hairless subclass. Similar findings were reported in previous works [28,43,63], which indicate the majority of Ethiopian landraces have hairless glumes. In contrast, a high level of polymorphism of GLH in Ethiopian durum wheat genotypes was reported by Bekele [64]. Negassa [9] suggested that the hairy glume phenotypic class of durum wheat may be associated with resistance to powdery mildew. The observed higher frequencies of lax and intermediate spike subclasses than that of dense spike subclass for SPD (Table 3) were contrary to the results obtained by Bechere et al. [28] and Hailu et al. [34] who reported the dominance of a dense spike in the Ethiopian durum wheat collections. The highest diversity index of SPD creates opportunities for breeders to develop suitable varieties for various environmental conditions. For instance, the lax spike is associated with resistance to spike-related diseases [43,63]. In agreement with this study, Jain et al. [65] reported the rarity of black glume color in Ethiopian durum wheat genotypes which is associated with gluten quality [9].

The observed significant genotypic differences, based on the chi-square test, and high Shannon diversity index for all qualitative traits, except for GLH, suggest the existence of high variation within Ethiopian durum wheat genotypes for the traits targeted in this study (Table 3). The significant impact of test sites on LDG and GLC traits identify that the growing environment had significant effects on the performance of durum wheat genotypes for these traits, while location had no significant effect on the remaining qualitative traits, implying that these traits are more governed by genetic than environmental factors (Table 3).

4.5. Promising Landraces for Processing Quality and Future Breeding

In this study, all genotypes that were ranked in the top 5% were landraces (Table S2), and none of the cultivars made it to the top 5%, mainly due to their low GC and GPC. The results may imply a trade-off between grain yield and some quality traits, as the cultivars were bred primarily for grain yield and related traits. The results also

suggest the superiority of durum wheat landraces over cultivars with regard to processing quality traits. The mean value of the grand population and the selected genotypes showed significant differences for GC and GPC, as they are the criterion for the selection of genotypes (Table 4). However, grain yield and related traits, such as TKW, which correlated negatively with the prioritized quantitative quality traits (GC and GPC), appeared to have non-significant differences in the mean value of the grand population and the selected top 5% genotypes (Tables 4 and S2). These landraces with optimal GC and GPC along with sufficient grain yield could be used in wheat breeding programs to improve both groups of traits simultaneously.

5. Conclusions

This study revealed the existence of substantial genetic variation among Ethiopian durum wheat genotypes for agro-morphological and quality traits. Some genotypes performed better than others for several phenotypic traits across the experimental sites. Hence, further studies on top performers to validate their stability over multiple environments are required. The moderate to high heritability with moderate genetic advance for TKW, SPL, GY, and GC suggests that the selection of genotypes for these traits in early generations could be effective. Ethiopian durum wheat may add value to the processing sector due to its significant proportions of distinct phenotypic characters such as amber color and vitreous kernels. Protein-related traits showed a significant positive correlation ($p < 0.05$) with phenological traits and a significant negative correlation ($p < 0.05$) with yield-related traits. The results may suggest breeding for the improvement of protein-related traits might negatively affect GY in durum wheat. Therefore, the top 5% of genotypes that have vitreous kernels with amber color, high GC and GPC along with optimal TKW and GY are highly valuable resources that should be used as donors of desirable alleles in durum wheat breeding programs. Given that only two locations were used in this study, traits that showed significance ($p < 0.05$) for genotypes by environmental interaction should be studied further across environments to determine the consistency of their performance.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/agronomy12071714/s1>, Table S1: Description of Ethiopian durum wheat genotypes used in the phenotypic diversity study. Table S2: Genotypes ranked in the top 5% based on their values in gluten content, grain protein content, thousand-kernel weight and grain yield over combined locations.

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