



Genetics and Breeding

Correlations between grain yield and related traits in winter wheat under multi-environmental traits

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Abstract. *In a series of field trials, a database of quantitative traits associated with winter wheat grain yield has been collected. The aim of the present study is to determine the relationships between the winter wheat (*Triticum aestivum* L.) traits of productivity in environments causing the maximum possible variation of each of the traits. In order to determine the correlations between the quantitative characters studied, all possible statistical methods have been applied (regression analysis, PCA, Multiple Correspondence analysis), which complement each other. It was found that the nature of the correlations between traits depends to a large extent on the methods for their evaluation. There are high and significant correlations between grain yields and the grain number per spike (weight of grain per spike and number of grains per m²) even under strong genotype by environment interaction of all the traits in trials. The established results are related to possible options for increasing winter wheat grain yield by breeding.*

Keywords: environment, evaluation, grain yield, interaction, productivity, *Triticum aestivum* L.

Introduction

Breeding of wheat has two main directions: grain yield and quality (Tsenov et al., 2009, 2010). The main objective of each breeding program is grain yield, regardless of the enormous variety of conditions (Valvo et al., 2018; Senapati et al., 2019). Grain yield is a quantitative trait that is formed by the level and variability of a number of traits of the plant the effect of which changes it (Mitchell and Sheehy, 2018). The interrelationships between these traits are an important element in the selection of grain yield (Tsenov et al., 2009; Hristov et al., 2011). In direct dependence on environmental conditions, correlations between the characters affecting the yield change substantially (Hristov et al., 2011; Terzić et al., 2018). Therefore, a similar grain yield is obtained as a result of various “combinations” of traits, directly dependent on the particular environments (Tsenov et al., 2012; Terzić et al., 2018). Each distinct trait changes with the conditions (Djuric et al., 2018), which is why the correlations between the traits are important for targeted breeding in a given region (Valvo et al., 2018). It is generally known that increasing the level of one trait is directly related to lowering the level of another for biological

reasons. For example, the negative correlation between grain size and the number of grains per spike (Mandea et al., 2019; Tsenov and Gubatov, 2020). In order to have a constant success in efforts to increase the productive potential of the wheat plant, it must meet certain morphological and biological characteristics, depending on the changing climate conditions and the current level of cultivation technology (Valvo et al., 2018). In order to make progress, it is necessary to build a proper plant biotype. The present high level of productivity as a result of long-term breeding in the country requires even more precise knowledge of the interrelationships between the traits to achieve a combination rather than an antagonism between them (Flohr et al., 2018; Quintero et al., 2018). In winter common wheat, the enormous genetic abundance of varieties and the contrasting conditions for their cultivation cause different correlations between these traits (Hristov et al., 2008; Raykov et al., 2016). This is the reason why a number of researchers are constantly studying the complex relationships between characters through various selection indices (Tsenov et al., 2017; García et al., 2019). The latter are mainly used to mitigate the negative correlations between essential traits directly affecting grain yields. Their application

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should be the result of lengthy and in-depth studies of the relationships between the components of productivity (Alonso et al., 2018). In order to derive maximum objective information, long-term Multi-Environmental field Trials (MET) should be organized, the conditions of which provoke to a maximum degree the variation of each of the traits to be investigated (Mandea et al., 2019). Any change to a trait will affect each of the other traits as well as the grain yield itself. For this purpose, MET are extremely useful (Gubatov et al., 2016; Öztürk et al., 2019).

The aim of the study is to estimate the nature of relationships between winter wheat (*Triticum aestivum* L.) traits known as components of productivity.

Material and methods

Field experiments

For the purpose of the study, two ecological trial databases were used. These include data from field trials conducted in a total of ten locations in Bulgaria over a period of eight years. The first group of experiments includes data from a study of 30 winter wheat varieties at five locations (Selanovtsi, Pordim, Brashlen, Burgas and Radnevo) and four consecutive seasons (2007-2010). The second group includes a group of 24 winter wheat varieties in five locations (Dobrich, Russe, Veliko Tarnovo, Yambol and Plovdiv) and four seasons (2009-2012). Some of those data on the studied traits, with a major emphasis on the environmental conditions on the plasticity and the stability of the varieties, have already been published (Tsenov and Gubatov, 2015; Gubatov et al., 2016).

Table 1. Summary descriptive statistics for the winter wheat varieties traits (N=1080)

Trait	Mean	SD*	CV**	Min	Max	Range	Skewness	Kurtosis
GY	6.64	2.025	30.5	2.17	13.24	11.07	.374	.265
EED	130	5.2	4.0	116	141	25	-.296	-.345
GFP	44.4	9.35	21.0	19	69	50	.080	1.028
VP	228	14.5	6.3	185	248	63	-1.037	.992
NPT	675	166.8	24.7	330	1480	1150	1.313	2.487
NGS	25.1	7.08	28.2	9	50	41.00	.126	-.432
TGW	40.4	5.34	13.2	25.6	57.7	32.1	-.267	-.004
WGS	1.01	0.307	30.3	0.33	1.92	1.59	-.173	-.713
NGm	16497	5035.9	30.5	6950	34673	27723	1.062	1.224

Legend: *Standard deviation, **Coefficient of variation, GY- grain yield, t/ha, EED- ear emergence date, as number of days after 1st of January, GFP- grain filling period, VP- vegetation period, NPT- number of productive tillers, NGS- number of grains per spike, TGW- 1000 grain weight, WGS- weight of grain per spike) and NGm- number of grains per m².

Components of productivity

In the two groups of field trials, the same characteristics of the winter wheat varieties were studied as follows: grain yield (GY, t/ha), ear emergence date (EED), as number of days after 1st of January, grain filling period (GFP), vegetation period (VP), number of productive tillers (NPT), number of grains per spike (NGS), 1000 grain weight (TGW), grain weight per spike (WGS) and number of grains per m² (NGm). Basic statistics on traits are shown in Table 1. Each of the signs has been investigated on the basis of each repetition of the three of the field experiments. All significant details of the experiments conducted can be found in the previous publications of Tsenov et al. (2014) and Gubatov et al. (2016)

Statistical analyses

Correlations between all tested traits have been studied with the main focus being on grain yield as a trait of their interdependencies. All possible types of correlation analyses that can be applied are used. To confirm the results, the capacities of Principal Component Analysis (PCA), a regression analysis and Multiple Correspondence analysis are also used. All of these are applied using the statistical programs IBM

SPSS 19, Statgraphics XVI and Statistica 10. Before being analyzed, the numerical values of each trait are ordered and standardized.

Results

The data from the field experiments show a strong interaction of the traits with the environment conditions (Table 2). The effects of each of the factors studied (season, location and variety) are reliable at the highest level. In general, traits are strongly influenced by the interaction of the variety*year, with the exception of only one NPT. It is clear that the variation of each of the traits is the result of the action and the interplay between the factors of the environment. The degree of change in the traits expressed by the coefficient of variation is also very high (Table 1), especially in the grain yield and the integral traits - WGS, NGm. This is a good enough prerequisite for studying the links between them.

There are positive correlations between grain yield and other traits (Table 3). The bold correlations are quite high at 95% probability. However, the values of R² (over the diagonal) show that there are only real interrelations between several characters.

Table 2. Analysis of variance for the winter wheat varieties traits - Type III Sums of Squares

Source	Df	MS		<i>p-value</i>		MS		<i>p-value</i>	
		GY		EED		GFP			
A:Year	3	643.960	0.000	4723.34	0.000	10860.9	0.000		
B:Location	4	223.340	0.000	1116.94	0.000	1315.8	0.000		
C:Variety	53	1.100	0.000	22.0372	0.000	13.1972	0.000		
A*B	12	102.362	0.000	507.313	0.000	4339.55	0.000		
A*C	159	0.625	0.000	7.81022	0.000	8.78262	0.000		
B*C	212	0.275	0.155	2.14739	0.092	3.12029	0.024		
Residual	636	0.246		1.85793		2.5168			
		VP		NPT		NGS			
A:Year	3	5846.94	0.000	878423	0.000	8817.52	0.000		
B:Location	4	22559.4	0.000	2.44E+06	0.000	929.097	0.000		
C:Variety	53	10.8069	0.000	8055.92	0.005	48.7538	0.000		
A*B	12	9489.29	0.000	983315	0.000	720.563	0.000		
A*C	159	4.70922	0.000	5168.49	0.383	19.3669	0.000		
B*C	212	3.15921	0.038	6583.25	0.005	12.1062	0.221		
Residual	636	2.60513		4996.76		11.1358			
		TGW		WGS		NGm			
A:Year	3	2558.67	0.000	21.53	0.000	2.39E+09	0.000		
B:Location	4	1737.29	0.000	0.1568	0.000	2.19E+09	0.000		
C:Variety	53	66.0236	0.000	0.0348	0.000	2.43E+07	0.000		
A*B	12	389.563	0.000	1.309	0.000	5.77E+08	0.000		
A*C	159	17.0346	0.000	0.0281	0.000	6.46E+06	0.000		
B*C	212	6.20382	0.538	0.0217	0.000	2.98E+06	0.021		
Residual	636	6.28435		0.0154		2.39E+06			

Legend: GY- grain yield, t/ha, EED-ear emergence date, as number of days after 1st of January, GFP- grain filling period, VP-vegetation period, NPT- number of productive tillers, NGS- number of grains per spike, TGW- 1000 grain weight, WGS- weight of grain per spike) and NGm- number of grains per m².

Table 3. Pearson correlation matrix between grain yield and related traits

Trait	GY	EED	GFP	VP	NPT	NGS	TGW	WGS	NGm
GY		0.33	0.01	0.19	0.10	0.40	0.11	0.48	0.83
EED	0.57		0.07	0.02	0.01	0.21	0.02	0.20	0.32
GFP	0.10	-0.27		0.15	0.06	0.00	0.00	0.00	0.01
VP	0.43	0.14	0.38		0.24	0.00	0.00	0.00	0.17
NPT	0.32	0.11	0.25	0.49		0.15	0.01	0.16	0.15
NGS	0.63	0.46	-0.04	-0.01	-0.38		0.00	0.82	0.44
TGW	0.34	0.14	-0.05	0.07	-0.09	0.03		0.19	0.00
WGS	0.69	0.44	-0.07	0.04	-0.41	0.91	0.44		0.30
NGm	0.91	0.57	0.12	0.41	0.38	0.66	-0.06	0.55	

Legend: Correlation coefficients below and coefficients of determinations above the diagonal; Values in bold are different from 0 with a significance level alpha=0.05, GY-grain yield, t/ha, EED-ear emergence date, as number of days after 1st of January, GFP-grain filling period, VP-vegetation period, NPT- number of productive tillers, NGS-number of grains per spike, TGW-1000 grain weight, WGS- weight of grain per spike and NGm-number of grains per m².

To consider that there is a relationship, we assume that at least 40% ($R^2=0.40$) of the values of the cases correspond to the model applied. According to the R^2 values, we can divide the correlations into two groups: traits with strong GY-NGm, WGS-NGS; traits with a good relationship - WGS-NGS, GY-EED. Similar are the correlations between pairs of traits - NGm-NGS, NGm-EED. Despite high values, all the other correlations are questionable and should not be considered as existing. A number of authors do not report that putative values of (r) are associated with low R^2 values and accept correlations for existing without being fully significant (Desheva, 2016; Phillip et al., 2018).

The NGS can be considered a major trait for grain yield. This assertion results from its direct correlation with GY ($r=0.63$) and the impact it has on WGS ($r=0.91$) and NGm ($r=0.66$), which in turn have a strong relationship with GY.

Wheat-based trait (NPT) shows an insignificant relationship with grain yield as well as strong negative correlations with NGS and WGS. On the other hand, its role in NGm has been proven. However, this correlation to the trait is significantly lower than that of NGS. The role of NGS on GY has been described extensively in various publications (Tsenov et al., 2012; Desheva, 2016; Mirosavljević et al., 2018; Valvo et al., 2018).

Discussion

Additional models for calculating correlation between traits are also used to check the results obtained (Table 4). Well known correlation of Spearman and Kendall models can be formulated as special cases of correlations. Both models reflect some extent the non-linear relationships between the traits through their ranks. Data analysis shows that the relationship

between the individual traits is maintained in principle, although the correlation coefficient values change with those of the Pearson model. This applies to the highest degree of NGm, and less for WGS and NGS, which still retain the link to grain yield. The relationship between GY and the traits VP and NGS is confirmed but at low values of R². In this situation, it is difficult to draw proper conclusions for selection of components of productivity.

Table 4. Rank correlations between grain yield and yield related traits

Trait	1*	<i>p</i>	R ²	2*	<i>p</i>	R ²	3*	<i>p</i>	R ²
EED	0.45	< 0.0001	0.20	0.29	< 0.0001	0.09	0.08	< 0.0001	0.11
GFP	0.15	< 0.0001	0.02	0.10	< 0.0001	0.01	0.26	< 0.0001	0.15
VP	0.51	< 0.0001	0.26	0.35	< 0.0001	0.12	-0.10	< 0.0001	0.22
NPT	0.31	< 0.0001	0.10	0.20	< 0.0001	0.04	0.14	< 0.0001	0.14
NGS	0.63	< 0.0001	0.39	0.46	< 0.0001	0.21	0.62	< 0.0001	0.41
TGW	0.33	< 0.0001	0.11	0.22	< 0.0001	0.05	0.29	< 0.0001	0.35
WGS	0.69	< 0.0001	0.47	0.51	< 0.0001	0.26	0.66	< 0.0001	0.36
NGm	0.90	< 0.0001	0.82	0.74	< 0.0001	0.55	0.95	< 0.0001	0.65

Legend: *Correlations by: 1- Spearman, 2- Kendall, 3- Partial by multi linear regression, EED- ear emergence date, as number of days after 1st of January, GFP- grain filling period, VP- vegetation period, NPT- number of productive tillers, NGS- number of grains per spike, TGW- 1000 grain weight, WGS- weight of grain per spike and NGm- number of grains per m².

Finally, the Multiple Linear Regression (MLR) model has been applied, which makes it possible to trace the relationship of the traits to the grain yield, regardless of their interrelation. In addition, this model (Table 5) provides information on the multicollinearity between the traits. Standardized regression coefficient values show strong connections of GY with NGS (r=0.62), WGS (r=0.72), and NGm (r=0.92). Practically, the

magnitude of these correlations is quite similar to those of the Pearson model. The additional removal of the variation of the individual traits through the partial and semi-partial correlations makes the connection of each character with the research unique. Again, the same traits have a strong relationship with GY: NGS (r=0.52-0.62), WGS (r=0.59-0.66), and NGm (r=0.65-0.85).

Table 5. Statistics of multilinear Regression analysis by SPSS

Trait	Standardized Coefficients*	<i>t</i>	<i>p</i>	Correlations		Collinearity	
				Partial	SemiPart	Tolerance	VIF**
	(Constant)	-0.012	0.990				
EED	0.01	2.52	0.012	0.08	0.06	0.499	2.00
GFP	0.03	8.71	0.000	0.26	0.22	0.626	1.59
VP	-0.01	-3.24	0.001	-0.01	-0.01	0.480	2.08
NPT	0.23	4.63	0.000	0.14	0.11	0.123	8.10
NGS	0.62	-26.03	0.000	0.62	0.52	0.010	99.08
TGW	0.10	9.99	0.000	0.29	0.24	0.054	18.53
WGS	0.72	28.97	0.000	0.66	0.59	0.009	107.37
NGm	0.92	106.02	0.000	0.85	0.65	0.076	13.21

Legend: *Dependent Variable is GY; **Variance Inflation Factor;

Model Equation: GY= 0.01*EED+0.03*GFP-0.01*VP+0.03*NPT-0.62*NGS+0.10*TGW+0.72*WGS+0.92*NGm

EED-ear emergence date, as number of days after 1st of January, GFP-grain filling period, VP-vegetation period,

NPT- number of productive tillers, NGS-number of grains per spike, TGW-1000 grain weight, WGS- weight of grain per spike and NGm-number of grains per m².

However, these traits show a very high level of interdependence from others (collinearity by VIF). According to the statistical criteria, values of this parameter above 10 indicate their relationship to grain yield is influenced by the relationship with other traits. To predict the effects of multi collinearity, a principal component analysis (PCA) was applied (Figure 1). The data show three components

(eigenvalues) of the variation of the traits, which is an indication of a complex non-linear manifestation of each of them against the background of the others. We can, with a high degree of credibility, claim that the traits NGm, WGS, NGS, EED actually correlated to grain yield. The "cleaning" of the variance between the characters is represented as vectors and is an indication of a link between them.

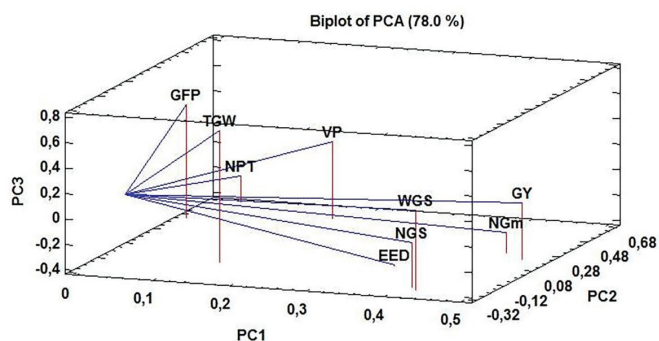


Figure 1. 3D Spatial representation of the trait vectors by PCA (GY- grain yield, t/ha, EED- ear emergence date, as number of days after 1st of January, GFP- grain filling period, VP-vegetation period, NPT- number of productive tillers, NGS- number of grains per spike, TGW- 1000 grain weight, WGS- weight of grain per spike and NGm- number of grains per m²).

The closer the vectors to each other (sharp angle), the greater the correlation between the traits, that express them. The information from the last applied analysis corresponds to a large extent with that of the correlation analyses before and can therefore be another confirmation of the links between the traits.

Conclusion

Finally, we can say that a detailed study of the relationship between grain yield of the tested 54 winter wheat varieties and some quantitative traits gives different information in direct dependence on the statistical methods that are applied. When looking for a link between traits that could potentially help the grain yield breeding, it is imperative that the data be analyzed by various statistical methods. The non-linear variation of each character complicates the determination of real connections between them. For this reason, caution should be taken not to make mistakes in the conclusions. On the other hand, it is clear that the WGS and NGm traits, which are practically indices, have the most tangible relationship to grain yield. The basis of both indices is the NGS trait, which according to the majority of research on the topic is largely determining grain yield in contrasting wheat environments. The NGS and WGS traits have the strongest relationship to grain yield, a strong negative correlation with NPT trait. The latter has a connection with GY, but it is weak and unreliable. On the other hand, its correlation with NGm is positive. In order to increase the number of grains per m², it is necessary to increase productive tillering because it is a component of this index ($NGm = NPT \cdot NGS$). The rise in NPT is associated with a decrease in WGS due to its negative correlation. The latter is important for improving lodging tolerance.

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