

Variation and Path Coefficient Analysis of Important Agronomic Traits in Two- and Six-rowed Recombinant Inbred Lines of Barley (*Hordeum vulgare* L.)

F. SHAHINNIA, A. M. REZAI and B. E. SAYED TABATABAEI

College of Agriculture, Isfahan University of Technology, Isfahan, 84154-83111 Iran,
e-mail: fshahinnia@ag.iut.ac.ir, am.rezai@cc.iut.ac.ir, sayedt@cc.iut.ac.ir

Abstract: The variation and relations of fifteen agronomic and morphologic traits (e.g., yield and its components, days to heading and test weight) were evaluated in 42 two-rowed and 57 six-rowed recombinant inbred lines of barley derived from Azumamugi × Kanto Nakate Gold cross. Significant differences were detected among lines for all traits studied. Kernel number/spike, awn length, kernel weight/spike, yield, harvest index and spike length had the highest coefficient of phenotypic variation (39.73%, 35.79%, 34.59%, 32.57%, 31.05% and 27.79%, respectively). Harvest index and biological yield explained more than 90% of total yield variation among two- and six-rowed lines. Discriminant analysis distinguished two- and six-rowed lines with spike length, awn length, kernel weight/spike, peduncle length and harvest index having the highest weight. Phenotypic and genotypic correlations showed a high positive relationship between yield and harvest index, biological yield, test weight, kernel weight/spike and kernel number/spike. Path analysis based on genotypic correlations for grain yield also showed the highest direct effect of kernel number/spike and indirect effect of kernel weight/spike via kernel number/spike.

Keywords: coefficient of determination; discriminant analysis; genotypic correlations; yield components

Cultivated barley, the fourth most produced cereal worldwide is a major feed grain crop (HAN *et al.* 2003). In barley, there is limited genetic variation for economically important traits because of domestication bottlenecks and intensive post-domestication selection. Breeding new barley varieties is based on creating new allele combinations and subsequent testing and selection of the desirable phenotypes in the selfing generations until an acceptable level of homozygosity in advanced generations is reached (PURI *et al.* 1982). Barley breeders sometimes make two row/six row crosses in an attempt to transfer desirable genes such as disease resistance, straw stiffness or high protein from one type to another (JUI *et al.* 1997). Accordingly, crosses between the two germplasm groups could be expected to produce positive transgressive segregants for economically important phenotypes (MARQUEZ-CEDILLO *et al.* 2001).

The objectives of this study were to (1) study the variation of important agronomic traits and to determine the contributions of each trait to total variability among genotypes, and (2) determine the direct and indirect effect of yield components on yield in two and six-rowed recombinant inbred lines (RILs) of barley using multivariate statistical methods.

MATERIALS AND METHODS

The experimental materials were ninety-nine F_{13} recombinant inbred lines of barley consisting of 42 two-rowed and 57 six-rowed lines derived from a cross between the barley cultivars Azumamugi (AZ) × Kanto Nakate Gold (KNG). These lines have been developed by the single seed descent method from F_2 plants, in field conditions at the National Institute of Agrobiological Sciences, Tsukuba, Japan

1. Means, coefficient of variations (CV) and mean squares for 15 important agronomic traits in RILs

Trait	Mean	CV (%)	Mean square ^A	
			between lines	within lines
Days to heading (DH)	161.00	5.15	136.28	8.16
Plant Height (PH, cm)	58.63	18.58	237.49	47.59
Peduncle length (PeL, cm)	17.74	22.36	30.50	18.80
Leaf length (LL, cm)	10.16	23.62	11.53	2.46
Leaf width (LW, cm)	1.18	18.80	0.09	0.03
Spike number (Sno./m ²)	657.00	19.44	32 632.02	19 217.22
Kernel number/spike (Kno.)	24.36	39.73	188.10	101.48
Kernel weight/spike (KW, g)	1.11	34.59	29.57	0.07
Awn length (AL, cm)	9.33	35.79	22.33	0.66
Spike length (SL, cm)	5.44	27.79	4.58	0.65
Biological yield (BY, kg/m ²)	1 243.04	20.64	131 664.61	62 828.50
Grain yield (GY, kg/m ²)	390.38	32.57	32 344.72	17 138.79
Harvest index (HI, %)	29.00	31.05	0.01	0.0072
1000 kernel weight (TKW, g)	32.57	14.11	42.33	10.23
Test weight (TW, g/250 cc)	158.19	6.58	217.12	142.42

^Aall of traits mean squares were significant at $p < 0.01$

(KOMATSUDA unpublished data). AZ is a six-rowed Japanese, winter barley of *uzu* or semibrachytic type and KNG is a two-rowed Japanese, spring of non-*uzu* type. A field experiment in randomized complete block design with two replications was conducted in 2003–2004 at Isfahan University of Technology Research Station, Isfahan, Iran (32°32' N, 51°32' E). Plots consisted of four rows 2 m long, 0.2 m apart with interplant spacing of 4 cm. Two hundred kg/ha ammonium phosphate before planting and 250 kg/ha at tillering stage as fertilizer were used.

Fifteen important agronomic traits including: days to heading (DH), plant height (PH, cm), peduncle length (PeL, cm), leaf length (LL, cm), leaf width (LW, cm), spike number/m² (Sno./m²), kernel number (K no.), kernel weight (KW, g), awn length (AL, cm), spike length (SL, cm), biological yield (BY, kg/m²), grain yield (GY, kg/m²), harvest index (HI, %), test weight (TW, g/250cc) and thousand kernel weight (TKW, g) were recorded for each plot or the mean of 10 randomly selected plants in the center rows of each plot. The omission of outer rows eliminated potential border effects.

Data were analyzed using SAS Software, Version 6 (SAS Institute Inc. 1993). The genotypic correlation coefficients were computed by the method

described by FISHER (1963). The path coefficient analysis was performed using genotypic correlations to assess direct and indirect effect of yield components on grain yield (DOFING & KNIGHT 1992).

RESULTS AND DISCUSSION

Analysis of variance (Table 1) revealed highly significant differences among lines for all of the traits. Kernel no./spike, awn length, kernel weight/spike, yield, harvest index and spike length had the highest coefficient of phenotypic variation (Table 1). The results revealed that there was considerable variation among the lines for yield, yield components, and related important agronomic traits. The contribution of each trait to variation in yield, based on stepwise regression analysis is shown in Table 2. Harvest index and biological yield explained most (96.8%) of the total yield variation. A high degree of variability for harvest index and biomass was found in winter wheat, oat and barley (SINGH & STOSKOPF 1971; TAKEDA & FREY 1985; BOUKERROU & RASMUSSEN 1990). Breeding for high biomass has been proposed as a way to enhance yield in small grain. Selection for larger plants or vegetative biomass, results in photosynthetic area

Table 2. Stepwise regression analysis for grain yield based on agronomic traits

Step	Variable entered to model ^A	Regression coefficient (%)						R2 (%)
		b0	b1	b2	b3	b4	b5	
1	HI	35.70	1126.13**					53.85
2	BY	-377.88	1156.77**	0.32**				96.80
3	AL	-386.21	1168.28**	0.31**	1.28*			96.89
4	KW	-389.44	1104.33**	0.31**	1.85*	20.81*		97.07
5	LL	-383.25	1105.99**	0.31**	3.12*	24.66*	-2.31*	97.16

^Avariables are defined in Table 1; **highly significant at $p < 0.01$; *significant at $p < 0.05$

to produce more carbohydrate or grater source as well as larger spikes with more and larger kernels or larger sink (BOUKERROU & RASMUSSEN 1990).

Discriminant analysis revealed that two and six-rowed lines could be separated by the first two functions. According to Table 3, spike length, awn length, kernel weight/spike and peduncle length had considerable loads in linear discriminant function. The high value of harvest index in the function again emphasized the important contribution of this trait in explaining the differences between two- and six-rowed lines. SINGH and STOSKOPF

(1971) compared harvest index in cereals. They found considerable variation in harvest index of spring and winter barley (51 and 45% respectively). Compared to direct selection for grain yield, the use of harvest index as indirect selection criteria gave 43% relative selection efficiencies (BOUKERROU & RASMUSSEN 1990).

Highly significant positive phenotypic correlations existed between grain yield and harvest index (0.73**), biological yield (0.63**), test weight (0.73**), kernel weight/spike (0.57**) and kernel no./spike (0.45**). Also, highly significant posi-

Table 3. Contribution of each trait to linear discriminant function for two and six rowed lines

Trait ^A	Function	
	six rowed lines	two rowed lines
DH	0.49	0.75
PH	0.58	-0.83
PeL	10.25	10.05
LL	2.06	1.78
LW	-0.65	0.41
Sno./m ²	0.05	0.05
Kno.	1.45	0.64
KW	-6.81	-3.99
AL	5.37	-4.79
SL	9.78	10.19
BY	0.14	0.16
GY	0.74	0.77
HI	889.05	930.73
TKW	3.74	5.04
TW	1.45	10.16
Constant value	-1337.00	-1341.00

^Atraits are defined in Table 1

Table 4. Path coefficient analysis for yield and yield components based on genotypic correlations

Trait ^A	Direct effect	Indirect effect via			Trait correlation with grain yield
		KW	Sno./m ²	Kno.	
Kw	-2.21	-	-0.09	2.84	0.54**
Sno./m ²	0.72	0.28	-	-0.60	0.41**
Kno.	2.99	-2.11	-0.14	-	0.75**

^Atraits are defined in Table 1; ^Bresidual factor = -0.35; **highly significant at $p < 0.01$

tive genotypic correlations were revealed between these traits (0.78**, 0.63**, 0.58**, 0.54**, and 0.75**, respectively). Highly significant association between yield, harvest index, biomass and test weight also was present in other studies (PURI *et al.* 1982; BOUKERROU & RASMUSSEN 1990; JUI *et al.* 1997). A highly significant negative correlation (-0.91**) was observed between kernel weight/spike and kernel no./spike. This negative correlation was mentioned by DOFING and KNIGHT (1992). Characters, which are phenotypically correlated but not genotypically correlated, will not produce repeatable estimates of inter-character associations and any selection based on the relationship is likely to be unreliable (ARIYO *et al.* 1987).

Correlation between yield and its components simply measures mutual relationships without presumption of causation (PURI *et al.* 1982) but the result of path coefficient analysis (Table 4) for grain yield and yield components can describe genotypic correlations to direct and indirect effects. The highest and positive direct effect on yield was comes from kernel no./spike. The direct effect of kernel weight/spike was negative but it had the highest positive indirect effect via kernel no./spike. We also observed highly significant negative correlation between kernel weight and kernel number. The negative and positive direct effects emphasize that yield components may compensate for each other under competitive conditions and that the maximum expression of each yield component is determined sequentially in the order of development (DOFING & KNIGHT 1992). The results of path analysis in this study are parallel to others such as PURI *et al.* (1982); RASMUSSEN (1987) and DOFING and KNIGHT (1992). They suggested kernel weight and kernel number as two easily measured characters that would be valuable in selecting for yield improvement in barley.

The RILs used in this study were produced to identify the QTLs for shoot differentiation in bar-

ley (KOMATSUDA *et al.* 1989). The parents, AZ and KNG, also show variation for morphological and physiological traits (MANO *et al.* 2001). The results of this study confirmed the presence of variation in important agronomic traits in this population that could be utilized in QTL mapping for agronomic traits.

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