Frequency of transgression and gene distribution in barley doubled haploid populations from first and second cycle hybrids

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Abstract. Barley doubled haploids (DH) derived from first and second cycle hybrids were investigated in field experiments. Parental lines designed for the second cycle hybrids were three doubled haploids from the first cycle hybrids, which were observed to have the highest grain yield. Yield structure characters, crude protein content and protein fractions were analysed. Phenotypic and genetic variability and the frequency of transgression in the studied populations were calculated. For the studied traits additive, [d], and epistatic (additive × additive), [i], effects as well as coefficient of gene dispersion were estimated. It was found that the phenotypic and genetic variability of DH populations derived from second cycle hybrids was higher than that of the original population for all the studied traits except grain yield. A greater proportion of transgressive lines than in the original population was also observed in populations from second cycle hybrids. Only one DH line exceeding the high yielding parent was found among 141 lines under study. The relationship between the frequency of transgressions and gene dispersion was recorded: the greatest number of transgressive lines occurred in those traits for which the dispersion was observed.

Key words: barley, doubled haploids, gene dispersion, recurrent selection, transgression.

Introduction

In barley, production of doubled haploids (DH) is an integral part of many breeding programmes because the use of doubled haploid techniques allows to shorten the time required for obtaining completely homozygous breeding lines (FRIEDT et al. 1986, PICKERING, DEVAUX 1992, ADAMSKI 1993, SNAPE

1997). DH lines are usually derived from F_1 hybrids (by androgenesis or by chromosome elimination in *Hordeum vulgare* × *H. bulbosum* crosses (see, e.g., PICKERING, DEVAUX 1992). The frequency of transgressive lines with suitable agronomic characters in DH populations derived from F_1 hybrids is rather low because haploid segregants are the result of only one round of recombination between parental genomes (FRIEDT, FROUGHI-WEHR 1983, CHOO et al. 1986, SURMA 1996). For increasing the frequency of desirable lines, the recurrent selection methods can be used (CHOO et al. 1979, PATEL et al. 1985, GALLAIS 1988, 1993, GOLDRINGER et al. 1996, 1997). This paper presents experimental results for comparison of the original DH population derived from the first cycle hybrids with populations of DH lines derived from the second cycle hybrids, i.e., from hybrids between the high yielding DHs selected from the original DH population.

Material and methods

Material for the studies covered four populations of doubled haploids: the original population (FCH) derived from the F_1 hybrids between cvs. Roland and Krystal (first cycle hybrids), and three populations: SCH1, SCH2 and SCH3, obtained from the F_1 hybrids between lines selected from the original population (second cycle hybrids). Parental lines designed for the second cycle hybrids were three doubled haploids, RK22, RK58 and RK63.1, which were observed to have the highest grain yield in breeding trials (ADAMSKI et al. 1992). The SCH1 population was derived from the F_1 hybrids RK22 × RK63.1, SCH2 – from RK63.1 × RK58 and SCH3 – from RK22 × RK58.

Doubled haploids were produced by "bulbosum" technique (KASHA, KAO 1970, DEVAUX 1986, ADAMSKI 1993).

Thirty FCH, 40 SCH1, 43 SCH2 and 28 SCH3 lines along with their parents and grand parents (Roland, Krystal) were investigated in four field experiments in 1996 at Cerekwica Station near Poznań. Each experiment was conducted in a complete randomized block design with three replications. The materials were planted in 4 m^2 plots with 10 × 3 cm spacing between plants. In each experiment the time of planting and soil type were the same. After harvesting the following agronomical traits were observed: grain yield per plot, grain weight per ear, 1000-grain weight, crude protein content (Kjeldahl N × 6.25) and albumin, globulin and hordein content (KAPAŁA 1994).

Statistical analyses

The analysis of variance for complete randomized block design was done for each experiment. It was used for comparison of phenotypic variances of

populations from first and second cycle hybrids and for partitioning the phenotypic variances into genetic (additive) and environmental components.

The frequency of positive and negative transgressions in the studied populations was calculated by comparing each line with both parents. The differences were tested by F-statistic at the 5% significance level. The genetic parameters connected with additive, [d], and epistatic (additive × additive), [i], effects were also estimated and tested (SURMA et al. 1984). Additive gene effects were calculated by two methods, one based on extreme DH lines, $[d]_L$, (SURMA et al. 1984), and the second based on parental forms, $[d]_P$, (MATHER, JINKS 1982). From the $[d]_P/[d]_L$ ratio the coefficient of gene distribution (r) in the FCH lines being parents for SCH hybrids, was estimated (JINKS, PERKINS 1969). Two hypotheses concerning gene distribution, H_0: r = 0 and H_0: r = 1, were tested as contrasts by F-statistic. For testing the H_0: r = 0 hypothesis, the contrast was formulated as a half of the difference between parental lines $(P_1 - P_2)$ and for H_0: r = 1 – as comparison of the differences between extreme and parental lines: $\frac{1}{2}(SCH_{max} - SCH_{min} - P_1 + P_2)$.

Results

Mean values of the studied traits for all the DH lines (DH-mean) and extreme lines (DH-max, DH-min) in the original population RK-FCH and in populations from second cycle hybrids, RK-SCH1, RK-SCH2 and RK-SCH3, are given in Table 1. The average grain yield per plot of DH lines derived from second cycle hybrids was higher than in the original population. It is visible both in values of DH-means and in values of extreme DH lines. In the case of grain weight per ear, DH-means of the original and SCH populations were similar, whereas maximal values were higher in populations of DHs from second cycle hybrids. Interesting results were obtained for 1000-grain weight. The SCH1 population derived from the cross between FCH lines of similar 1000-grain weight (RK22 – 50.08 g, RK63.1 – 50.30 g) distinguished by a high average value of this trait (DH-mean – 52.12 g) and a very high 1000-grain weight for maximal line – 57.12 g. Similarly, in the SCH2 population the maximal line appeared to be transgressive in 1000-grain weight (54.55 g versus 51.63 g for the better parent). For protein content no important differences were observed between the original population and populations from second cycle hybrids. However, significant differences between the studied populations were found for protein fractions: the minimal albumin and globulin content was lower, and the maximal hordein content was higher in populations from second cycle hybrids than in the original population.

Table 1. Mean values for studied traits of barley DH populations derived from first (FCH) and second (SCH) cycle hybrids

Generation	Grain weight per ear (g)	1000-grain weight (g)	Grain yield per plot (kg)	Protein content (%)	Albumin + globulin (%)	Hordein (%)
FCH						
Roland (R)	1.15	51.07	2.44	12.63	3.17	5.57
Krystal (K)	1.09	48.14	1.97	11.50	3.57	4.47
DH lines (RK)						
DH max.	1.20	52.40	2.58	13.77	3.77	5.93
DH min.	0.99	46.42	1.91	11.00	3.07	3.93
DH-mean	1.12	49.78	2.28	12.25	3.37	4.95
S.E.	0.065	1.273	0.134	0.443	0.093	0.094
SCH1 (RK22 × RK63.1)						
Roland	1.15	50.41	2.34	12.58	3.18	5.56
Krystal	1.13	47.73	2.05	11.70	3.52	4.45
RK22	1.08	50.08	2.33	11.20	3.57	4.47
RK63.1	1.12	50.30	2.57	11.90	3.30	5.63
DH lines						
DH max.	1.35	57.12	2.79	13.30	3.77	6.26
DH min.	0.98	44.98	2.04	10.70	2.70	4.00
DH-mean	1.14	52.12	2.49	11.96	3.27	5.09
S.E.	0.044	1.173	0.144	0.494	0.074	0.098
SCH2 (RK63.1 × RK58)						
Roland	1.12	49.23	2.40	12.60	3.16	5.59
Krystal	1.06	48.07	1.97	11.62	3.60	4.48
RK63.1	1.14	49.99	2.55	11.90	3.32	5.60
RK58	1.16	51.63	2.41	11.87	3.19	5.47
DH lines						
DH max.	1.33	54.55	2.71	13.77	3.73	6.13
DH min.	0.99	46.35	2.01	11.10	2.77	4.10
DH-mean	1.14	49.86	2.38	12.08	3.32	4.93
S.E.	0.081	1.156	0.147	0.466	0.064	0.099
SCH3 (RK22 × RK58)						
Roland	1.16	51.30	2.41	12.53	3.20	5.55
Krystal	1.07	49.57	1.88	11.80	3.60	4.47
RK22	1.13	49.17	2.34	11.53	3.55	4.46
RK58	1.22	51.49	2.35	11.87	3.15	5.49
DH lines						
DH max.	1.29	50.53	2.66	12.97	3.70	5.60
DH min.	0.97	44.80	1.95	11.20	2.90	4.13
DH-mean	1.14	48.74	2.30	12.00	3.33	4.84
S.E.	0.064	1.380	0.118	0.459	0.073	0.093

Table 2. F-statistic values for comparisons of phenotypic variances of barley DH populations derived from first (FCH) and second (SCH) cycle hybrids

Trait	F-statistic value					
	SCH1 v. FCH	SCH2 v. FCH	SCH3 v. FCH	SCH3 v. SCH1	SCH1 v. SCH2	SCH3 v. SCH2
Grain weight per ear	1.82	1.97	1.84	1.01	1.10	1.08
1000-grain weight	2.17	1.49	1.32	2.86	1.46	1.96
Grain yield per plot	1.43	1.56	1.17	1.22	1.08	1.31
Protein content	1.04	1.25	1.86	1.78	1.19	1.49
Albumins + globulins	3.01	2.71	1.78	1.69	1.11	1.52
Hordeins	1.01	1.22	1.60	1.62	1.23	1.31

$F_{0.05} = 1.79$
$F_{0.01} = 2.29$

Comparison of the phenotypic variability of the studied populations showed that SCH populations had significantly higher variances than the original population for grain weight per ear and for albumin and globulin content, and SCH1 population, additionally, for 1000-grain weight (Table 2). Genetic variances and estimates of heritability coefficients of these traits were also relatively higher in SCHs than in the original population (Table 3). Only for grain yield and protein content the genetic variance in the population from the second cycle hybrids was lower than in populations from the first cycle hybrids. It may be noticed that epistatic effects [i] for all the traits, with the exception of hordein content in SCH2, were not significant (Table 4). This indicates that the genetic

Table 3. Estimates of genetic variance (V_G) and heritability coefficient (H, %) for studied traits in barley DH populations derived from first (FCH) and second (SCH) cycle hybrids

Trait	FCH		SCH1		SCH2		SCH3	
	V_G	H	V_G	H	V_G	H	V_G	H
Grain weight per ear	0.002	18.53	0.004	63.03	0.003	24.12	0.004	38.96
1000-grain weight	2.139	39.64	6.187	74.98	3.229	46.87	1.714	37.43
Grain yield per plot	0.033	54.79	0.021	39.68	0.017	34.84	0.030	58.81
Protein content	0.259	46.75	0.214	36.94	0.164	23.19	0.085	21.26
Albumins + globulins	0.019	58.95	0.061	88.24	0.054	88.25	0.037	82.30
Hordeins	0.306	96.94	0.308	95.43	0.257	94.56	0.207	94.04

Table 4. Estimates and F-statistic values for additive × additive effects [i] in barley DH populations derived from first (FCH) and second (SCH) cycle hybrids

Trait	FCH		SCH1		SCH2		SCH3	
	[i]	F	[i]	F	[i]	F	[i]	F
Grain weight per ear	−0.025	0.61	0.028	1.70	0.017	0.20	−0.005	1.26
1000-grain weight	0.492	0.48	−0.776	1.84	0.591	0.60	−0.805	0.94
Grain yield per plot	−0.029	0.21	−0.065	0.87	−0.018	0.07	−0.005	0.23
Protein content	0.134	0.39	0.034	0.02	0.353	2.40	0.075	1.16
Albumins + globulins	0.047	1.12	−0.040	1.24	−0.066	3.83	0.130	2.98
Hordeins	−0.015	0.15	0.038	0.63	0.135	15.30	0.055	1.57

$F_{0.05}$ = 4.00
$F_{0.01}$ = 7.08

variances for these traits were close to additive variance, thus, the calculated heritability coefficients may be treated as heritability in a narrow sense.

Estimation of differences between each DH line and the parental forms in each population has allowed to determine the transgression effects. The frequency of DH lines that significantly (at the level α = 0.05) exceeded the mean of the better or lower parent is presented in Table 5. It can be noticed that in the original population occurred only few transgressive lines in respect of protein content and protein fractions. SCH populations produced a greater proportion of transgressive lines as compared with the original (FCH) population for every character, especially for grain weight per ear, 1000-grain weight

Table 5. Proportion (%) of barley DH lines exceeding better parent ($> P_1$) and lower parent ($< P_2$) in populations derived from first (FCH) and second (SCH) cycle hybrids

DH population		Grain weight per ear	1000-grain weight	Grain yield per plot	Protein content	Albumins + globulins	Hordeins
FCH	$> P_1$	0	0	0	3.4	3.4	6.9
	$< P_2$	0	0	0	0	0	6.9
SCH1	$> P_1$	22.5	42.5	0	5.0	5.0	10.0
	$< P_2$	2.5	5.0	2.5	0	17.5	5.0
SCH2	$> P_1$	2.3	0	0	11.6	27.9	7.0
	$< P_2$	0	7.0	7.0	0	9.3	67.9
SCH3	$> P_1$	0	0	3.6	3.6	0	0
	$< P_2$	7.1	7.1	10.7	0	14.3	10.7

Table 6. Estimates and F-statistic values for additive effects [d] in barley DH populations derived from second cycle hybrids (SCH)

Trait	$[d]_L$ [1]		$[d]_P$ [2]	
	estimate	F	estimate	F
SCH1				
Grain weight per ear	0.184	68.8	0.020	0.86
1000-grain weight	6.370	117.94	0.111	0.04
Grain yield per plot	0.375	26.97	0.123	2.92
Protein content	1.300	27.73	0.350	2.01
Albumins + globulins	0.533	209.54	0.133	13.10
Hordeins	1.133	523.18	0.583	138.60
SCH2				
Grain weight per ear	0.167	17.05	0.012	0.10
1000-grain weight	4.100	27.56	0.820	1.10
Grain yield per plot	0.350	22.79	0.065	0.83
Protein content	1.333	32.72	0.016	0.01
Albumins + globulins	0.483	194.38	0.067	3.70
Hordeins	1.016	419.74	0.067	1.80
SCH3				
Grain weight per ear	0.159	24.46	0.040	1.57
1000-grain weight	2.863	17.17	1.163	2.83
Grain yield per plot	0.355	36.41	0.005	0.01
Protein content	0.883	14.80	0.167	0.53
Albumins + globulins	0.400	121.30	0.200	30.32
Hordeins	0.617	174.35	0.517	122.39

[1] estimate based on DH lines
[2] estimate based on parents
$F_{0.05} = 5.29$, $F_{0.01} = 8.49$

and content of albumins and globulins, where both plus and minus transgressions were observed. It should be noted that for grain yield only one recombinant line considerably exceeding the better parent was found among the studied populations.

In Table 6 estimates of parameter [d] obtained on the basis of extreme lines ($[d]_L$) and parental forms ($[d]_P$) for SCH populations are given. Estimates of parameter $[d]_L$ differed significantly from zero for all the traits, whereas $[d]_P$ only for protein fractions in SCH1 and SCH3 populations. Basing on parameters $[d]_L$ and $[d]_P$, the coefficient of gene dispersion (r) was calculated. Table 7 presents estimates of r and results of testing of the hypotheses H_0: r = 0 and H_0: r = 1. It can be noticed that in all the SCH populations for grain

Table 7. Estimates of dispersion coefficient (r) for studied traits in populations of barley DH lines derived from second cycle hybrids (SCH) and results of testing of hypotheses H_0: r = 0 and H_0: r = 1

Trait	SCH1			SCH2			SCH3		
		F-statistic			F-statistic			F-statistic	
	r	H_0: r = 0	H_0: r = 1	r	H_0: r = 0	H_0: r = 1	r	H_0: r = 0	H_0: r = 1
Grain weight per ear	0.109	0.86	27.14	0.072	0.10	7.30	0.252	1.57	6.81
1000-grain weight	0.017	0.04	56.92	0.200	1.10	8.42	0.406	5.83	6.04
Grain yield per plot	0.328	2.92	6.07	0.186	0.83	7.47	0.014	0.01	17.72
Protein content	0.269	2.01	7.40	0.012	0.01	15.95	0.189	0.53	4.87
Albumins + globulins	0.249	13.1	58.93	0.139	3.70	72.23	0.500	30.32	15.16
Hordeins	0.514	138.6	61.61	0.066	1.80	183.25	0.838	122.39	2.29

$F_{0.05} = 5.29$
$F_{0.01} = 8.49$

weight per ear, grain yield, protein content and 1000-grain weight (with exception of SCH3) favourable alleles were dispersed in parental genomes (the hypothesis H_0: r = 1 was rejected and H_0: r = 0 was not rejected). In the case of 1000-grain weight in population SCH3 favourable alleles were partially associated (0 < r < 1). For albumin + globulin content alleles were found to be partially associated in the parents of populations SCH1 and SCH3 (both the tested hypotheses were rejected), and dispersed in SCH2 (only H_0: r = 1 was rejected). In the case of hordein content alleles were fully associated in the parents of population SCH3 (r = 1), partially associated in SCH1 (0 < r < 1) and dispersed in the parents of SCH2 (r = 0).

Discussion

The original population of DH lines derived from the first cycle hybrids was compared with populations of DH lines derived from hybrids between high yielding doubled haploids selected from the original population. DH lines from second cycle hybrids were found to have a shorter range of yield variability than the original population. It should be noted, however, that both phenotypic and genetic variances for grain yield in SCHs were considerable. This suggests that in high yielding lines selected from the original population the desirable

alleles were not associated. This was reflected in estimates of r coefficient, which were significantly lower than 1. It is of interest that only one DH line exceeding the high yielding parent was found among 141 studied DHs. On the other hand, seven lines exhibited a negative transgression in yielding. These results indicate a slight response to selection for grain yield. A low proportion of barley DH lines with a positive transgression in grain yield has been frequently observed. CHOO et al. (1986) among 398 doubled haploids found only seven superior lines, whereas the frequency of inferior lines was 14 times higher. Similarly, SURMA (1996) recorded two DH lines among 145 studied DHs that yielded higher than the better parent and 26 that yielded lower than the poorest parent. Predominance of a negative transgression for grain yield may be caused by the presence of the complementary additive × additive epistasis and association of genes in parental forms (CHOO et al. 1986) or by linkage disequilibrium (SURMA 1996). In this study no significant additive × additive epistasis was found for grain yield, and genes appeared to be dispersed between parents. This suggests that a linkage rather than epistasis is an important reason for the low proportion of superior lines in the studied materials.

In the present study the original population of DH lines as well populations derived from second cycle hybrids revealed a significant variability in all the traits. Greater phenotypic and genetic variances were recorded in populations from second cycle hybrids than in the original population for grain weight per ear, 1000-grain weight and albumin and globulin content. These results indicate that high yielding lines selected from the original population, although similar phenotypically in respect of yielding and yield components, differed in their genetic background.

In this study the relationship between the frequency of transgressions and gene dispersion in parents was observed; the greatest number of transgressive lines was recorded in those traits for which the r estimate did not differ significantly from zero. Such a relationship was also found between gene dispersion and heterosis in barley. POWELL and THOMAS (1992) showed that the level of heterosis observed in SCH hybrids was due to dispersion of favourable alleles between parents. A similar result was reported by SURMA (1996). This suggests that gene dispersion can be an important genetic factor conditioning the occurrence of heterosis and transgression in barley. When favourable alleles are dispersed between parents, their association in DH progenies may result in a situation where lines with associated (+) alleles will be superior and lines with associated (–) alleles will be inferior to their parents.

It is evident from the presented results that one cycle of recurrent selection increases the frequency of transgressive progenies. Thus, the recurrent selection applied along with the doubled-haploid method may increase the effectiveness of barley breeding.

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