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Half sib selection for resistance to powdery mildew (*Erysiphe graminis* DC. f.sp. *secalis* Marchal) in rye populations

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Summary. Three rye populations originating from different cultivars were subjected to half sib progeny selection for three cycles to improve their resistance to powdery mildew (*Erysiphe graminis* DC. f.sp. *secalis* Marchal). Selection was carried out in the greenhouse (measurement of spore production at the three-leaf stage), in the field (scoring of mildew infections at ear emergence) or at both locations. All selections resulted in significant responses, the level of which varied depending on the genetic base of mildew resistance in the populations. Greenhouse selection was most effective when the frequency of dominant genes could be increased, whereas field selection was less effective. Estimates for heritability in the narrow sense ranged from 0.42–0.94 indicating that mildew resistance can be increased effectively. Inbred lines should be selected in base populations with a high level of quantitative resistance and/or a high frequency of dominant genes. For an effective resistance strategy in hybrid breeding such dominant genes are most valuable, and can be selected in young growth stages. In 'Strain CG' the increase of dominant resistance genes by early selection resulted in a significant improvement of kernel yield.

Key words: *Secale cereale* L. – *Erysiphe graminis secalis* – Half sib selection – Heritability – Resistance breeding

Introduction

The study of rye (*Secale cereale* L.) populations revealed a great variability for resistance to powdery mildew (*Erysiphe graminis* DC. f.sp. *secalis* Marchal). Frequency distributions showed that the majority of plants have a quantitative level of resistance, but low reesistance occurs much more frequently than high resistance, which in many populations is completely absent (Lind and Wenzel 1984). Recurrent selection increased the proportion of resistant genotypes and thus the resistance level of populations (Lind and Züchner 1985). The selection experiments provided information about the genetic base of resistance and the method to be used for population improvement.

From population studies (Lind and Züchner 1985) it was concluded that mildew resistance is controlled by a few too many genes. Kast (1983) used inbred lines and identified monogenic as well as polygenic resistance. Information about the genetics of mildew resistance is of increasing importance in rye breeding, since hybrid cultivars are grown. The low genetic variability of these cultivar types, compared with the populations, demands preventive action against attack by different pathogens. Hybrids make it possible to establish planned breeding for disease resistance by using isolated genes.

In the present study, populations from different breeding programs selected for high kernel yield under German agricultural conditions are used to demonstrate the effect of different selection methods on mildew resistance and its influence on yield potential. Selection aims at the production of populations applicable as pools for dominant resistance genes that can be used for hybrid breeding.

Materials and methods

Original populations

The selection experiments were performed with rye populations from three genetic sources.

(1) A population originating from 'Halo' × 'Danko' (provided by Dr. Wagner, Saatzucht Dr. h.c. R. Carsten oHG, FRG). The cross population was selected for high kernel yield

without considering mildew resistance. Both parents are commercial cultivars. The first parent contributed a low level of quantitative resistance in the young growth stages but had a high level of adult plant resistance (Lind et al. 1986). The second parent was characterized by a high level of quantitative resistance in all growth stages and had dominant genes conditioning complete resistance.

(2) Population 'Strain CG' (provided by Saatzucht P. H. Petersen, FRG), a selection from the German cv 'Carogold'. It had a medium level of resistance in the very early growth stages but was less resistant than 'Halo' at the adult stage. It also carried a few dominant resistance genes.

(3) Population 'Strain CG' x 'Halo' combined the resistances of both parents described above.

Selection for resistance

Three different selection methods were applied.

(1) Selection in the greenhouse (early selection) after artificial inoculation with a mildew population propagated on the cv 'Halo'. For the inoculation of trays, each containing 100 plants, a spore settling tower was used in which 200 spores/cm^2 were applied on the leaves. Resistance was evaluated by spore production measurements of seedlings (Lind 1983) at the three-leaf stage that corresponds to stage 13 in the decimal code system for growth stages (Zadoks et al. 1974).

(2) Selection in the field at ear emergence (late selection) after natural inoculation, which was secured by the adjacent growth of highly susceptible genotypes. Resistance was determined on the two leaves below the flag leaf by the use of a scale from 1–9, in which 1 indicated complete resistance and 9 high susceptibility. The mean of both scores, which were taken at ear emergence (stage 55), was used for selection.

(3) Selection in two successive steps by the combination of early and late selection (combined selection) within the same population.

Table 1 shows experiment surveys that were conducted with each population. Depending on the genetic base of mildew resistance, not all selection methods were used in each population. All populations that originated from a cross with 'Halo' were subjected to early or combined selection, as this cultivar lacked resistance at the young stages; whereas the 'Carogold' population was used in all selection methods.

Table 1. Selection methods applied to different rye populations at the three-leaf stage and at ear emergence

Populations	Selection experiment	Stage of selection	
		three leaves (13)[a]	ear emergence (55)[a]
Halo x Danko (original)			
Halo x Danko	1	+	+
Strain CG (original)			
Strain CG	2	+	
Strain CG	3		+
Strain CG	4	+	+
Strain CG x Halo (original)			
Strain CG x Halo	5	+	

[a] 13, 55 = selection at the 3-leaf stage and at ear emergence, respectively

Population improvement

Single plant and progeny selection were combined to produce populations with improved mildew resistance. As starting material, 3,000 kernels of the original populations, partitioned into two replications, were sown in the greenhouse for each of the five selection experiments (Table 1). They had to be sown once for 'Halo' x 'Danko' and 'Strain CG' x 'Halo' and three times for 'Strain CG'. At this first step, selection was based on single plants. In the case of early selection at the three-leaf stage, 12% of plants (360 genotypes) from these populations were transplanted to an isolated field plot. This portion included the majority of genotypes with the lowest spore production. If late selection was applied all the kernels were sown directly in the isolated field plot where selection was performed for low mildew attack at ear emergence. Twelve percent of these plants were allowed to pollinate each other randomly. For combined selection in a first step, 24% of the plants were selected in the greenhouse, whereas for standardization of selection intensity the reduction to 12% was done by selection in the isolated field plot. As in separate early and late selection, low spore production and low mildew scores were used as selection criteria. In all five populations the plants were harvested individually.

In the two following selection steps the size of selected populations was kept constant (12% of 3,000 plants). Selection, however, was carried out on a family base, i.e. the most resistant plants from the families with the highest mean resistance were used to compose the next population. Finally, after the second step of family selection, the most resistant plants were bulked to form the final populations that were compared for their resistance and yield properties.

Comparison of populations

The yield comparisons between the original and final populations were performed in a randomized complete block design with four replications at four environments (Bad Schönborn, Niendorf, Grünbach 1986 and 1987). The trials were sown in 10 m^2 plots without fungizide treatment.

Comparison of resistance was performed with field and greenhouse data. The field data were obtained from the two Grünbach yield trials (two environments) and were used to determine adult plant resistance. For this purpose 100 plants per plot were scored for mildew attack. The greenhouse data were obtained from measurements of spore production at the three-leaf stage in two succeeding test sets, where the original and final populations were arranged in the same experimental design as in the yield comparison. Per greenhouse plot, 150 plants were measured. In all tests the cvs 'Halo' and 'Danko' were included. Comparisons of mildew resistance were performed by using: (a) the mean spore production and mean mildew score of whole populations; and (b) the 10% fraction of populations that include the most resistant genotypes. The observed selection response of populations was expressed as a percentage decrease of mildew scores and spore production in the final populations compared to the original populations. A t-test at the 0.05 level of significance was used to compare the population means.

The yield and the resistance data from populations were subjected to analyses of variance, where replications (R), environments or test sets respectively (S), families (F), interactions (SF) and error (E) were regarded as sources of variation. The error term was used for comparisons between population means based on least significant differences (Snedecor and Cochran 1973). In addition, the coefficients of variation (C.V.) averaged over replications and locations were calculated as $C.V. = \sigma/x$, where σ and x are the standard deviation and the mean of a population, respectively.

Heritability

The selection system included two selection steps in which progenies of individual plants were tested. These progenies are half-sib families (F) and can be used to estimate the additive genetic variance σ_A^2 (Wricke and Weber 1986). If inbreeding and epistatic effects are neglected, the heritability in the narrow sense is given by $h^2 = \sigma_A^2/\sigma_P^2$, where $\sigma_P^2 = (\sigma_E^2 + R\sigma_{SF}^2 + SR\sigma_F^2)/SR$. For experiments 1, 2, 4 and 5 (Table 1), heritability was estimated in the greenhouse by testing the spore production of progenies after early or combined selection. For experiment 3 only data from field tests were available.

Results

The response to selection for mildew resistance is presented in Tables 2 and 3. Both tables indicate the significance between the original and respective selected populations. For comparisons between populations of different genetic origin the least significant differences (LSD) are presented. Table 2 shows the response of spore production after greenhouse selection at the three-leaf stage. In the 'Halo' x 'Danko' population a significant reduction of spore production could be attained by combined early and late selection. In 'Strain CG', early selection alone was most successful and surpassed both late and combined selections. The latter two selection types do not differ significantly from each other. In 'Strain CG' x 'Halo', however, spore production was not reduced by early selection. The improvement of populations is not always correlated with an improvement of the 10% fraction of plants that comprises the most resistant genotypes. With the exception of 'Strain CG' there was no reduction of spore production in this selected portion. In 'Strain CG' the improvement of the resistant fraction was equally successful, irrespective of procedure at selection. Compared to the reference cvs 'Halo' and 'Danko', all selected populations had lower sporulation values than 'Halo', and the best of them were even more resistant than 'Danko'.

Comparing the coefficient of variation for populations, it can be seen that the respective original population had a lower value, i.e. that selection always resulted in an increase of variation. Such a trend was not obtained for the 10% fraction where changes in different directions occurred.

Table 2. Spore production of rye populations selected for improved mildew resistance at different growth stages. Means of populations with identical genetic origin followed by the same letter are not significantly different at $P = 0.05$ as determined by LSD

Populations[a]	Population		10% fraction	
	Mean ($\times 10^3$)	C.V.[b]	Mean ($\times 10^3$)	C.V.[b]
Halo x Danko	58.6 a	4.29	14.1 a	6.89
Halo x Danko (13+55)	31.6 b	5.32	8.0 a	6.35
Strain CG	99.1 a	2.80	42.2 a	7.07
Strain CG (13)	30.2 c	8.12	3.1 b	6.66
Strain CG (55)	70.0 b	3.45	18.1 b	11.00
Strain CG (13+55)	52.8 bc	4.67	10.2 b	5.75
Strain CG x Halo	58.3 a	5.61	9.9 a	8.53
Strain CG x Halo (13)	50.2 a	6.70	9.3 a	10.55
Halo	73.2	3.58	22.4	6.50
Danko	51.9	4.85	10.5	9.83
LSD (0.05)	24.0		18.9	

[a] 13, 55 = selection at the 3-leaf stage and at ear emergence, respectively

[b] C.V. = coefficient of variation

Table 3. Mildew scores from field trials of rye populations selected for improved mildew resistance at different growth stages. Means of populations with identical genetic origin followed by the same letter are not significantly different at $P = 0.05$ as determined by LSD. Mildew scores: 1.0 completely resistant, 9.0 highly susceptible

Populations[a]	Population		10% fraction	
	Mean	C.V.[b]	Mean	C.V.[b]
Halo x Danko	4.8 a	0.57	3.8 a	0.46
Halo x Danko (13+55)	4.1 b	0.70	3.3 a	1.80
Strain CG	5.2 a	0.53	4.0 a	1.56
Strain CG (13)	3.6 c	0.88	3.0 b	1.81
Strain CG (55)	4.9 a	0.58	3.7 ab	1.54
Strain CG (13+55)	4.5 b	0.70	3.4 b	2.00
Strain CG x Halo	4.9 a	0.50	4.1 a	1.43
Strain CG x Halo (13)	4.4 b	0.71	3.7 a	0.83
Halo	4.5	0.63	4.1	1.15
Danko	4.5	0.58	3.5	1.57
LSD (0.05)	0.4		0.5	

[a] 13, 55 = selection at the 3-leaf stage and at ear emergence, respectively

[b] C.V. = coefficient of variation

The field trials (Table 3) demonstrated a significant selection effect in 'Halo' x 'Danko' and 'Strain CG' x 'Halo'. On the other hand, late selection in the field did not improve the resistance of 'Strain CG' compared with the original population. After early selection, however, it was significantly more resistant than the reference cvs 'Halo' and 'Danko'. As in sporulation measurements, there was an increase of the coefficients of variation after selection in the total populations. As before, these responses were not the same in the resistant 10% fractions.

Table 4 presents additive variances and heritability estimates. Both parameter were changed by selection in about the same direction. In 'Halo' x 'Danko' they de-

Table 4. Heritability and additive variance σ^2_A of spore production (experiments 1, 2, 4, 5) and mildew scores (experiment 3), and observed response to selection (%) of rye populations selected at different growth stages

Population[a]	Exp. no.	σ^2_A		Heritability		Observed response (%)	
		Cycle 2	Cycle 3	Cycle 2	Cycle 3	Spore production	Mildew scores
Halo × Danko (13+55)	1	983.8	352.2	0.72	0.55	46.1*	11.1*
Strain CG (13)	2	774.6	1,424.1	0.45	0.94	69.5*	18.9*
Strain CG (55)	3	0.1	0.2	0.48	0.59	29.4*	2.7
Strain CG (13+55)	4	453.2	887.2	0.42	0.68	46.7*	10.8*
Strain CG × Halo (13)	5	2,005.5	2,027.1	0.59	0.58	13.9	8.3*

* Final populations significantly improved at $P=0.05$ as determined by the t-test

[a] 13, 55 = Selection at the 3-leaf stage and at ear emergence, respectively

Table 5. Response of kernel yield on different types of selection for mildew resistance in rye populations. Values followed by the same letter are not significantly different at $P=0.05$ as determined by LSD

Populations[a]	Kernel weight (kg/ha)
Halo × Danko	2,369 a
Halo × Danko (13+55)	2,474 a
Strain CG	2,266 b
Strain CG (13)	2,565 a
Strain CG (55)	2,432 a b
Strain CG (13+55)	2,506 a
Strain CG × Halo	2,375 a
Strain CG × Halo (13)	2,397 a
Halo	2,421
Danko	2,494
LSD (0.05)	187

[a] 13, 55 = selection at the 3-leaf stage and at ear emergence, respectively

creased from cycle 2 to cycle 3, whereas in 'Strain CG' they increased. Heritability in cycle 2 calculated from field data (experiment 3) ranged in about the same order as estimates from greenhouse experiments. In cycle 3, however, the increase of the estimate was especially striking after early selection (experiment 2). In 'Strain CG' × 'Halo', heritability and additive variance had about the same size in both cycles.

The observed response to selection is shown in the last two columns of Table 4. In general, responses were higher for spore production than for mildew scores and high heritability estimates were not always correlated with high observed responses or with effects on both resistance traits. The field selection (experiment 3) did not improve mildew resistance in the field but effected spore production alone. On the other hand, early selection in the greenhouse (experiment 5) did not significantly reduce spore production; rather, it improved mildew resistance in the field.

Table 5 shows the effects of selection for mildew resistance on kernel yield. Significant responses did not occur in the populations 'Halo' × 'Danko' and 'Strain CG' × 'Halo'. In 'Strain CG', however, all selection types caused significant yield increases that were most pronounced after early selection. The degree of yield increase suggests a correlation with the improvement of mildew resistance. Most of the selected populations reached or even surpassed the yield level of 'Halo' and 'Danko'.

Discussion

In all populations selection resulted in an increased mildew resistance. The degree of improvement depended on the type of selection applied and on the effects of genes within each population. As was indicated by selection experiments (Lind et al. 1986), 'Danko' carries genes with dominant effects that can be easily recognized in young growth stages. Similar acting genes were identified in 'Carokurz', which is closely related to 'Carogold'. Both cultivars did not differ in the genetic base of mildew resistance but had a lower frequency of dominant genes than 'Danko'. In 'Halo' such genes could not be detected. Its resistance seems to be based polygenically.

In the population 'Halo' × 'Danko' a reduction of spore production was attained but there was no significant improvement of the 10% fraction of plants, which indicates that the original population had a high number of highly resistant genotypes.

The results obtained in 'Strain CG' × 'Halo' showed that dominant resistance genes could not be identified. As the original population of 'Strain CG' had a very low frequency of dominant genes, the randomly selected plants crossed with 'Halo' presumably did not carry such genes. On the other hand, greenhouse selection led to the appearance of genotypes with a significantly improved resistance level in the field. The success of selection may be related to the selection of minor genes that increase adult plant resistance. Lind and Züchner (1985) showed that mass selection in 'Halo' could slightly increase the frequency of highly resistant genotypes at the three-leaf stage, but not the population mean. The higher resistance of genotypes in this population was mainly the result of favourable minor gene combinations and not of dominant genes.

Selection was very effective in 'Strain CG'. As there were no significant differences between the means of 10% fractions of all selected populations, it may be assumed that each selection method was equally effective in identifying the most resistant genotypes. According to earlier investigations they demonstrate a preference for dominant genes (Lind et al. 1986). Even late selection increased the frequency of resistance genes; their effects, however, were low in the field but significant in the greenhouse. Differences between the population means refer to the effectiveness of selection methods in reducing the proportion of highly susceptible genotypes.

Heritability in the narrow sense was changed in all populations during selection and was related to additive variance estimates. In 'Halo' × 'Danko' they decreased with each cycle as a consequence of the elimination of susceptible genotypes. In 'Strain CG' × 'Halo' the variance remained about the same, indicating that selection could not induce a remarkable decrease of variability. The increase of the frequency of genotypes with dominant resistance genes in 'Strain CG' resulted in higher estimates for the additive variance which depended, however, on the selection method applied. With greenhouse selection, which was performed under largely controlled conditions, the differences in resistance could be detected with higher accuracy than in field selection, which is especially true for the effects of dominant genes conditioning complete or a high level of resistance. Field selection even disturbed the success of previous greenhouse selection as is indicated by the lower response of combined selection (experiment 4).

All populations, except 'Strain CG' × 'Halo', can be used as a source for dominant genes. The 10% fraction of populations usually provides sufficient information about the existence of dominant genes. Their effects can easily be recognized in greenhouse tests at the three-leaf stage. Additionally, existing minor genes with additive effects are of importance in the improvement of inbred lines, but are actually of less importance if resistance of hybrids is to be improved rapidly and effectively. In the long run, however, minor genes should also be used with more effort to broaden the genetic base of resistance and to impede the adaptation of pathogens that may be a consequence of the frequent use of very few dominant resistance genes. The relationship between gene frequencies and the stability of host/parasite-systems is discussed by Geiger et al. (1980).

It is shown that selection for mildew resistance has a low effect on the yield potential of populations, unless selection results in a high increase in the number of resistance genes, as in 'Strain CG'. The yield of all populations, which included genetic material from 'Halo', was not affected by the selection. Comparing the 'Halo'-types with 'Strain CG', it was conspicious that the former types were characterized by the senescence of leaves in the early ripening stages that may impair late infection by leaf pathogens. Adult plant resistance, therefore, was often overestimated in 'Halo'. Consequently, the resistance to leaf pathogens had a lower effect of 'Halo'-types than in genotypes, which were represented by all descendants of the cvs 'Carogold' and 'Carokurz' and had leaves remaining photosynthetically active for a longer time. In none of the populations did the increase in the number of resistance genes result in a reduction of yield capacity.

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