This is a postprint of:

Shtaya, M.J.V., J.C. Sillero, K. Flath, R. Pickering & D. Rubiales, 2007. The resistance to leaf rust and powdery mildew of recombinant lines of barley (*Hordeum vulgare* L.) derived from *H. vulgare* x *H. bulbosum* crosses. **Plant Breeding** 126: 259-267.

doi: 10.1111/j.1439-0523.2007.01328.x

The final printed version can be visited at: http://onlinelibrary.wiley.com/doi/10.1111/j.1439-0523.2007.01328.x/epdf

The resistance to leaf rust and powdery mildew of recombinant lines of barley (*Hordeum vulgare* L.) derived from *H. vulgare* x *H. bulbosum* crosses

M. J. Y. Shtaya^{1*}, J. C. Sillero², K. Flath³, R. Pickering⁴ & D. Rubiales¹

¹ Institute of Sustainable Agriculture, CSIC, Apdo. 4084, 14080 Córdoba, Spain

² CIFA, Alameda Del Obispo, IFAPA-CICE, Apdo. 3092, 14080 Córdoba, Spain

³ Federal Biological Research Centre for Agriculture and Forestry, Institute for Plant Protection of Field Crops and Grassland, D-14532 Kleinmachnow, Germany

⁴ New Zealand Institute for Crop & Food Research, Private Bag 4704, Christchurch, New Zealand.

^{*} Current Address: Faculty of Agriculture, An-Najah National University, P.O.Box 707, Nablus, Palestinian Territories.

Abstract

A set of 23 recombinant lines (RLs) of barley (*Hordeum vulgare* L.) derived from *H. vulgare* x *H. bulbosum* L. crosses was inoculated with barley leaf rust (*Puccinia hordei*) and powdery mildew (*Blumeria graminis* f.sp. *hordei*) at the seedling stage to identify their levels and mechanisms of resistance. Eight RLs were studied further in glasshouse and field tests. All three barley parents were highly susceptible to powdery mildew and leaf rust isolates. Several RLs showed partial resistance expressed as high relative latency periods (RLPs) and low relative infection frequencies (RIFs) against leaf rust. 182Q20 Golden Promise RL showed a higher RLP and a lower RIF than Golden Promise and had a similar response to Vada with all leaf rust isolates. This high level of partial resistance was due to a very high level of early aborted colonies without host cell necrosis. Several RLs showed hypersensitive resistance to some or all isolates. The resistance of 102C2/14, 169P15 and 38P18 was due to a

high percentage of early aborted colonies associated with host cell necrosis. For powdery mildew, 81882 Vada RL was completely resistant to the CC1 isolate and had a hypersensitive resistance to the CO-02 isolate. Three Emir RLs (216U3, 219W4 and 177L20) were completely resistant to both powdery mildew isolates. The resistant RLs generally showed high percentages of early aborted colonies not associated with host cell necrosis. 219W4 and 81882 showed a higher percentage of early aborted colonies associated with host cell necrosis with isolates CC1 and CO-02, respectively. Three of the eight RLs tested in the field had higher levels of partial resistance than their parents. Our results indicate that *H. bulbosum* contains major and minor gene(s) for resistance to leaf rust and powdery mildew that can be transferred to cultivated barley.

Keywords Barley, *Hordeum bulbosum*, *Blumeria graminis* f.sp. *hordei*, *Puccinia hordei*, hypersensitive resistance, partial resistance.

Introduction

Leaf rust (*Puccinia hordei*) and powdery mildew (*Blumeria graminis* f. sp. *hordei*), are two of the most important foliar diseases on barley and cause significant economic losses. Wild barley relatives such as *Hordeum vulgare* L. ssp. *spontaneum* have been widely used in barley breeding programmes for disease resistance. *H. vulgare* ssp. *spontaneum* has broad resistance to leaf rust and powdery mildew, and many genes have been identified and transferred to cultivated barley (Jahoor and Fischbeck 1993; Kintzios et al. 1995; Backes et al. 2003). However, most sources of powdery mildew and leaf rust resistance have been overcome by corresponding virulence within the pathogen. New sources of resistance should, therefore, be identified for breeding programmes.

The wild barley species *Hordeum bulbosum* L., the only species in the secondary genepool of barley, is interesting to plant breeders for two reasons. Firstly, its chromosomes

are eliminated in crosses with barley to produce doubled haploids. Secondly, it has some desirable agronomic characters such as disease resistance (Thomas and Pickering 1983; Xu and Snape 1989; Walther et al. 2000) and has shown resistance for many years to many powdery mildew and leaf rust isolates. This resistance can be transferred to cultivated barley. Xu and Snape (1989) reported resistance to powdery mildew and rusts in *H. vulgare* x *H. bulbosum* hybrids. Xu and Kasha (1992) and Pickering et al. (1995) reported the transfer of powdery mildew resistance gene(s) from *H. bulbosum* to *H. vulgare*. Pickering (1992) identified chromosome substitution lines developed from *H. vulgare* x *H. bulbosum* hybrids that were more resistant to powdery mildew and other foliar diseases than their *H. vulgare* parents. Singh et al. (2003) reported the transfer of a dominant gene for scald resistance from *H. bulbosum* to barley.

The objectives of the present study were: 1) to record the level of resistance and characterise the mechanisms of resistance to powdery mildew and leaf rust in recombinant lines (RLs) derived from *H. vulgare* x *H. bulbosum* crosses; 2) to identify race-specific resistance genes in the RLs and to determine their novelty by comparing their infection types (ITs) with the ITs on a differential set; 3) to evaluate the partial resistance of several of the RLs in the field by comparing mean disease severities (MDS) with their parents. Disease resistant RLs were selected in New Zealand and subsequent tests carried out in Spain and Germany.

Materials and methods

Plant material

Twenty-three recombinant lines (RLs) were used in the Spanish experiments, eight of which were also tested in Germany for powdery mildew (Table 1). The RLs contain introgressed DNA from *H. bulbosum* and were derived from hybrids between *H. vulgare* x *H.*

bulbosum (Pickering et al. 1998, 2000; Pickering and Johnston 2005). These plants, together with the three recurrent barley parents Vada, Emir and Golden Promise, were studied for resistance. Three resistance alleles in two RLs have already been assigned gene symbols: 38P18 with resistance to leaf rust (*Rph18.ag*) and 81882 with resistance to powdery mildew (*Mlhb1.a*) and leaf rust (*Rph17.af*) (Pickering et al. 1995, 1998, 2000).

The 'Pallas' isolines differential set for powdery mildew of barley (Kølster et al. 1986) and a differential set for leaf rust (Steffenson et al. 1993) were used to determine the virulence spectrum of all isolates used.

Inoculum

In Spain, plants were inoculated with five isolates of barley leaf rust, representing a wide virulence range, and two isolates of barley powdery mildew. In Germany, inoculations were carried out with 24 isolates of powdery mildew on eight RLs and the Pallas isolines (Kølster et al. 1986). The virulence/avirulence factors and the origin of the isolates used in the experiment are shown in Table 2.

Inoculation

Leaf rust

Three to four seeds per RL were sown in $35 \times 20 \times 8$ cm trays in three replicates. Each tray contained eight accessions. The susceptible line L94 and the partially resistant cv. Vada were added to each box as references. Eleven days after sowing, when the primary leaf was fully expanded and the second leaf was emerging, first leaves were placed in a horizontal position, adaxial surface up, with the help of metal staples, and inoculated with *P. hordei* in a settling tower by dusting a mixture of freshly collected spores with talcum powder (1:10, v/v). Each box was inoculated with 3 mg of spores of the appropriate isolate (Niks and Rubiales

1994). The inoculated plants were kept in an inoculation chamber in darkness for about 11 hours at 20°C with a relative humidity of about 100%. Plants were then transferred to a growth chamber at 20°C and white fluorescent light (12 h light / 12 h dark). To reduce the risk of cross-contamination of the isolates, inoculation with each isolate was carried out on different days.

Powdery mildew

Seedlings of all RLs were grown under mildew-free conditions at 16°C and 10,000 lx continuous light. Eleven days after sowing when the primary leaf was fully expanded, 50 mm (Spain) or 30 mm (Germany) of a central leaf segment was excised from each seedling and placed adaxial surface up in a square petri dish filled with 0.6% agar and 125 ppm (Spain) or 30 ppm (Germany) Benzimidazole. In each petri dish, two to four leaf segments per line were randomly fixed in three replicates. One day before inoculum was required, heavily infected plants were shaken to remove ageing conidia, to ensure a supply of vigorous young spores. Inoculation was carried out by blowing spores from the infected plants over the leaf segments using a settling tower. A glass slide was placed in the settling tower to monitor inoculum density, which was adjusted to give approximately 20 conidia mm⁻² (Spain) or 2-4 conidia mm⁻² (Germany) (Haugaard et al. 2002). After inoculation, petri dishes were transferred to a growth chamber at 18-20°C (Spain) or 16°C (Germany) and incubated in darkness for 12 h. They were then transferred to a growth chamber with fluorescent lighting (12 h light / 12 h dark – Spain, or continuous light - Germany) with temperatures as before (Edwards 1993).

Field tests

RLs were grown in the field at 2003, Germany, with resistant and susceptible controls in a randomised block with two replicates (Moll et al. 2000). Strips of mildew-susceptible cultivars were grown between each block, and these were artificially inoculated with a mixture of 11 isolates at growth stage Zadoks 21-23 (Zadoks et al. 1974).

Preparation of leaves for microscopy

<u>Leaf rust</u>

Five days after inoculation a central leaf segment of nearly 2 cm² was collected from each plant in Spain. Leaves were fixed and cleared by boiling for 1.5 min in lactophenol/ethanol (1:2, v/v) and stored overnight in this mixture at room temperature. Segments were then washed once with 50% ethanol for 30 min, once with 0.05 M NaOH for 30 min, rinsed three times in water (10 min each), and soaked in 0.1 M Tris/HCl buffer (pH 8.5) for 30 min. They were then stained with 0.1% solution of Uvitex 2B in the same buffer. This was followed by rinsing four times with water before washing in a solution of 25% glycerol for 30 min. A few drops of lactophenol were added to the solution to prevent deterioration by fungi. Leaf segments were examined at 100x with Leica epifluorescence equipment (DM LB, 330 to 380 nm wavelength transmission).

Powdery mildew

Half of each previously inoculated leaf segment (about 25 mm) was excised 48 h after inoculation. These leaf segments were placed, with the adaxial (inoculated) surface up, on filter paper moistened with ethanol: acetic acid (3:1, v:v) for fixation. The fixative was changed every day until the leaves were free from chlorophyll. Leaves were then transferred onto filter paper moistened with water for 24 h, and finally stored on filter paper moistened with lacto glycerol (lactic acid : glycerol : water, 1:1:1 v/v) for microscopic observation (Rubiales and Carver 2000).

Macroscopic observation

<u>Leaf rust</u>

Latency period (LP) was determined daily by counting the number of uredia visible in a marked area (2-3 cm²) on each seedling, using a 6x lens. The LP was calculated as the time from the beginning of the inoculation to the time at which 50% of the uredia had appeared (Parlevliet 1975). The final number of uredia was used to determine the infection frequency (IF). The actual LP and IF were converted into relative latency period (RLP) and relative infection frequency (RIF), taking the LP and IF of L94 as 100%. The infection type was recorded 12 days after inoculation using the 0-9 scale of McNeal et al. (1971).

Powdery mildew

Infection type (IT) was recorded 5 days (Spain) or 12 days (Germany) after inoculation, following the 0-4 scale of Moseman (1965) where: 0 = no visible signs of infection; 1 = brown necrotic lesions with little or no mycelial development; 2 = some necrosis and chlorosis with slight to moderate mycelial development; 3 = chlorosis with moderate mycelial development; and 4 = abundant mycelial development with little or no necrosis or chlorosis. In addition to this in Spain, disease severity (DS) was estimated for each leaf segment as the percentage of the leaf surface covered by powdery mildew colonies. Infection frequency was calculated by counting the number of mildew colonies using a 6x lens and converting to colonies cm⁻². In Germany in the glasshouse ITs of 0-2 were considered resistant and 3-4 susceptible. In the field trials in Germany, disease development was assessed by recording the percentage leaf area infected on three dates and converting to mean disease severity – MDS (Moll et al. 2000).

Microscopic observations

Leaf rust

Accessions showing high levels of partial resistance or hypersensitive reaction, their recurrent parents and the two control lines were selected for microscopic observation. One hundred infection units were studied per leaf segment at 100x magnification, and classified according to their stage of development (Niks 1982). Early aborted colonies (EA) were defined as individuals that formed a primary infection hypha and no more than six haustorial mother cells. Those colonies that formed more than six haustorial mother cells were classified as established colonies (EST). Colony size (CS) was estimated by calculating the length (L) and the width (W) of 20 randomly chosen established colonies and CS calculated using the formula: $CS = \pi LW/4$.

Powdery mildew

Accessions showing resistance reactions (low IT) were selected for microscopic observation. To stain fungal structures and facilitate microscopy, a drop of Trypan blue in lactoglycerol (0.1%) was placed on a coverslip and a clear leaf segment was lowered onto the coverslip, so that the inoculated surface of the leaf segment contacted the stain. The coverslip was then inverted onto a microscope slide smeared with lactoglycerol to complete the mount (Rubiales and Carver 2000). Observations were made with Leica epifluorescence equipment (DM LB, 330 to 380 nm wavelength transmissions).

To determine the success of attempted plant epidermal cell penetration by fully developed germlings, 50-100 mature appressoria were examined on each leaf. If more than one fungal germ tube was in contact with a single epidermal cell, the germlings were disregarded, thus avoiding possible interactive effects between multiple attacks on the same cell. Some host epidermal cells survived attack, producing a papilla beneath the appressorium

of the fungus and resisting penetration (EA-); other epidermal cells died in response to attack and whole-cell autofluorescence was evident (EA+). Other cells that survived attack were penetrated by the fungus that formed a haustorium within the epidermal cells (EST-) and subsequent mycelial ramification.

Data analysis

Analysis of variance (ANOVA) was calculated by using PROC GLM in the SAS programme (SAS Institute 1988) or with SAS-Application RESI (Moll et al. 2000). Comparisons between lines were made by the Duncan test (Spain) or the Dunnett test (Germany).

Results

Reaction to leaf rust

Table 3 shows the macroscopic observations (IT, RLP and RIF) of the RLs and their recurrent parents with five isolates of leaf rust. The RLP of the partially resistant check Vada varied from 115 to 138% of L94, depending on the isolate. Golden Promise and Emir showed moderate levels of partial resistance.

Many RLs showed RLPs higher than their recurrent parents and as high as the partially resistant check Vada (Table 3). Regarding the Golden Promise RLs, 182Q20 showed a higher RLP and a lower RIF than Golden Promise and was similar to Vada with all isolates used. Two lines (38U4/1/3/10 and 38U4/1/3/8) showed high RLP to various isolates. Their partial resistance was higher than Golden Promise and as high as Vada. 53A8 and was resistant (IT = 5-6) to one isolate, but susceptible to the other four isolates (Table 3).

Six Emir RLs showed hypersensitive resistance to all or some of the isolates used. 102C2/14, 169P15 and 38P18 showed strong hypersensitive resistance (IT = 0-4) to all the

isolates. 119Y4 was resistant (IT = 3-4) to four isolates (CO-01, Al-02, TU-03 and IVP2000), but susceptible (IT = 8) to 1.2.1 isolate. 36L36 was resistant (IT = 6) to CO-01 and 1.2.1 isolates, but susceptible to the other three isolates. 219W4 was resistant (IT = 5-6) to one isolate, but susceptible to the other four isolates (Table 3).

The results of the microscopic observations are shown in Table 4. The high level of partial resistance in 182Q20, and to some extent in 38U4/1/3/8 and 38U4/1/3/10 was due to a high percentage of early aborted colonies without host cell necrosis. The resistance of 102C2/14, 169P15 and 38P18 was due to a high percentage of early aborted colonies associated with host cell necrosis. This result accords with the lower IT observed macroscopically (Table 3).

Reaction to powdery mildew

Table 5 shows the macroscopic observations on infection type (IT), disease severity (DS) and infection frequency (IF) of all the RLs and their parents using two isolates of powdery mildew in Spain. All three barley parents were highly susceptible to powdery mildew isolates (IT = 4), but they showed different levels of severity. Vada was the most susceptible parental line to CO-02 isolate, but it was only moderately susceptible to the CC1 isolate. 81882 was completely resistant to the CC1 isolate (IT = 0) and had a hypersensitive resistance (IT = 2) to the CO-02 isolate. Golden Promise and Emir gave similar susceptible reactions to both isolates. None of the Golden Promise RLs was more resistant than Golden Promise. Of the Emir RLs, 200A3 and 169P15 had lower DS and IF than Emir with CO-02 isolate, and with CC1 isolate they were moderately susceptible. 102C2/14 showed a DS and IF lower than Emir with CC1 isolate, but with CO-02 isolate it was moderately susceptible. 216U3 and 219W4 were completely resistant (IT = 0) to both isolates. 177L20 was fully resistant to the CC1 isolate, and only a few colonies were observed with the CO-02 isolate (IT = 0(4)).

All the eight tested RLs gave similar ITs to the 26 powdery mildew isolates used, with one or two exceptions (Tables 5 and 6). 212Y1 was not fully susceptible to some isolates and although 177L20, 216U3 and 219W4 were fully resistant to CC1 and CO-02 isolates, in Germany there was slight susceptibility of 177L20 and 219W4 to two isolates and of 216U3 to five isolates. More striking were the results from 200A3: in Spain ITs of 4 were recorded whereas in Germany it showed strong resistance to all but one of the 24 isolates.

81882 was more resistant than its parent, Vada, in seedlings tests to all isolates tested (Tables 5 and 6) and field tests (81882 MDS = 5.6, Vada MDS = 40.6). Golden Promise and two of its RLs, 53A8 and 182Q20, were fully susceptible to all 26 test isolates. Since 182Q20 was also susceptible in the field, it probably does not have any mildew resistance genes. Although 53A8 appeared to show higher partial resistance than Golden Promise in the field (MDS = 14.6 vs 40.7, respectively) the difference was not significant. In contrast, the third Golden Promise RL, 212Y1, was resistant to six of the isolates at the seedling stage and showed a significantly higher partial resistance (MDS = 13.8) than Golden Promise (MDS = 40.7). The RLs in an Emir background, 177L20, 200A3, 216U3, and 219W4, showed different reaction patterns from Emir, which only has *Mla12* resistance (Torp et al. 1978). Hence, they must contain other resistance genes or gene combinations. The genes of 177L20 and 219W4 are probably identical because of their similar reaction patterns to all isolates. In the field, only the RL 200A3 (MDS = 3.5) had a significantly higher resistance than Emir (MDS = 46.1).

Table 7 shows the microscopic observations of the resistant RLs and their parents with the two isolates. The resistant RLs generally showed high percentages of early aborted colonies not associated with host cell necrosis. 219W4 and 81882 showed a higher percentage of early aborted colonies associated with host cell necrosis with isolates CC1 and CO-02, respectively.

Discussion

The present study clearly indicates that *H. bulbosum* is an important and useful source of partial and hypersensitive resistance to barley leaf rust and powdery mildew, confirming observations of Thomas and Pickering (1983), Xu and Snape (1989), Pickering (1992), Xu and Kasha (1992), Pickering et al. (1995) and Singh et al. (2003).

Different resistance reactions were observed among the RLs and their barley parents. Many RLs showed low ITs and/or longer LPs to one or more isolates used in the study. It seems that the introgressed DNA segments from *H. bulbosum* contain minor and major gene(s) for partial and hypersensitive resistance to leaf rust and powdery mildew.

Resistance to leaf rust

Several RLs showed high RLPs and low RIFs against leaf rust. The high level of RLP in 182Q20 against all isolates of leaf rust used was remarkable since it was higher than its recurrent parent (Golden Promise) and as high as the partially resistant check Vada. 182Q20 contains a DNA fragment from *H. bulbosum* located distally on chromosome 2HL (R. Pickering, unpublished) and this fragment may contain some minor genes that confer the high level of partial resistance present in 182Q20. The high level of PR to all isolates used was due to a very high level of early aborted colonies without host cell necrosis, and may indicate a durable form of resistance. 53A8 Golden Promise RL showed hypersensitive resistance to one isolate of leaf rust (TU-02) indicating that this DNA fragment on chromosome 4HL may harbour some specific major gene(s) for leaf rust resistance. The hypersensitive resistance of 53A8 to isolate TU-02 was due to a high level of early aborted colonies with host cell necrosis.

Although three Emir and Golden Promise RLs carry a distal *H. bulbosum* DNA fragment on chromosome 2HL conferring leaf rust resistance, 182Q20 showed a different

reaction from 38P18 and 102C2/14, indicating that this DNA fragment is not exactly the same size in all three RLs, or that the *H. bulbosum* parent contains different alleles or, finally, that resistance alleles transferred from *H. bulbosum* to *H. vulgare* do not behave identically in different genetic backgrounds as they do in the *H. bulbosum* background (Xu and Kasha 1992).

Resistance to powdery mildew

There was little resistance among the Golden Promise RLs to powdery mildew in the seedling tests, indicating that there are no effective minor or major gene(s) for resistance against powdery mildew in the introgressed *H. bulbosum* DNA fragments. However, 212Y1 was significantly more resistant than Golden Promise in the field. Furthermore, the non-significant trend towards partial resistance in 53A8 has been borne out in field trials in New Zealand, Denmark and the United Kingdom (unpublished data).

Several Emir RLs were, however, resistant (low IT) to one or many isolates of powdery mildew. Their resistance against powdery mildew was due to a high percentage of early aborted colonies without host cell necrosis. 219W4 Emir RL, with a distal introgression on chromosome 7HL (R. Pickering, unpublished), gave a hypersensitive reaction to one isolate of leaf rust (TU-03); it was also more resistant to ten isolates of powdery mildew indicating that the *H. bulbosum* DNA fragment in 219W4 has resistance genes against at least two barley foliar diseases.

The Vada RL 81882 and the Emir RL 200A3 showed effective hypersensitive resistance in seedlings to all isolates tested as well as partial resistance. Their reaction patterns in the seedling test differed from the patterns of all Pallas differential lines. They must, therefore, carry new and effective resistance genes that could be used for developing mildew-resistant cultivars.

From our results we can conclude that *H. bulbosum* is an important source of resistance against powdery mildew and leaf rust. Effective major gene(s) for resistance against leaf rust can be transferred from *H. bulbosum* to *H. vulgare* since many RLs showed resistance to the most virulent isolate TU-03. For future research, it will be important to study allelism among genes located on chromosome 2HL, which confer hypersensitive resistance to leaf rust in some of the Emir RLs and non-hypersensitive resistance in 182Q20 Golden Promise RL. Preliminary inheritance and allelism studies indicate that the alleles conferring resistance to powdery mildew in 177L20, 216U3 and 219W4 are simply inherited and allelic, although 216U3 was susceptible to five isolates compared with susceptibility to only two isolates for 177L20 and 219W4. These differences may be due to the heterozygous nature of the common *H. bulbosum* parent. We aim to continue inheritance studies of these resistance gene(s) and determine their relationship to other mapped resistance genes to establish how many new major resistance gene(s) to powdery mildew and leaf rust are available for breeders.

Acknowledgments

We gratefully acknowledge the Spanish Agency for International Cooperation (AECI), CICYT projects AGF99-1036-CO1 and AGL2005-01781 for financial support in Spain. R. Pickering acknowledges the financial support of the Foundation for Research, Science and Technology (New Zealand).

References

Backes, G., L. H. Madsen, H. Jaiser, J. Stougaard, M. Herz, V. Mohler, and A. Jahoor, 2003:
Localisation of genes for resistance against *Blumeria graminis* f.sp. *hordei* and *Puccinia graminis* in a cross between a barley cultivar and a wild barley (*Hordeum vulgare ssp. spontaneum*) line. Theor. Appl.Genet. **106**, 353 - 362.

- Edwards, H. H, 1993: Light affects the formation and development of primary haustoria of *Erysiphe graminis hordei* in leaf epidermal cells of *Hordeum vulgare*. Physiol. Mol. Plant Path. **42**, 299-308.
- Haugaard, H., D. B. Collinge, and M. F. Lyngkjær, 2002: Mechanisms involved in control of *Blumeria graminis* f. sp. *hordei* in barley treated with mycelial extracts from cultured fungi. Plant Path. **51**, 612-620.
- Jahoor, A., and G. Fischbeck, 1993: Identification of new genes for mildew resistance of barley at the *Mla* locus in lines derived from *Hordeum spontaneum*. Plant Breeding **110**, 116-122.
- Kintzios, S., A. Jahoor, and G. Fischbeck, 1995: Powdery-mildew-resistance genes *Mla29* and *Mla32* in *H. spontaneum* derived winter-barley lines. Plant Breeding **114**, 265-266.
- Kølster, P., L. Munk, O. Stølen, and J. Löhde, 1986: Near isogenic barley lines with genes for resistance to powdery mildew. Crop Sci. **26**, 903-907.
- McNeal, F. H., C. F. Konzak, E. P. Smith, W. S. Tate, T. S. Russell, 1971: A uniform system for recording and processing cereal research data. USDA, Agricultural Research Service ARS, Washington, D.C., p. 34 121.
- Moll, E., K. Flath, and H. P. Piepho, 2000: Testing of crop cultivars for resistance to noxious organisms at the Federal Research Centre, Part 3. Mitteilungen aus der Biologischen Bundesanstalt für Land- und Forstwirtschaft, Berlin-Dahlem, pp128.
- Moseman, J. G, 1965: Genetic studies with cultures of *Erysiphe graminis* f. sp. *hordei* virulent on *Hordeum spontaneum*. Trans. Brit. Mycol. Soc. **48**, 479-489.
- Niks, R.E, 1982: Early abortion of colonies of leaf rust, *Puccinia hordei*, in partially resistant barley seedlings. Can. J. Bot. **60**, 714-723.

- Niks, R. E., and D. Rubiales, 1994: Avirulence factors corresponding to barley genes *Pa3* and *Pa7* which confer resistance against *Puccinia hordei* in rust fungi other than *P. hordei*.
 Physiol. Mol. Plant Path. 45, 321 331.
- Parlevliet, J. E., 1975: Partial resistance of barley to leaf rust, *Puccinia hordei* I. Effect of cultivar and development stage on latent period. Euphytica **24**, 21-27.
- Pickering, R.A., 1992: Monosomic and double monosomic substitution of *Hordeum bulbosum*L. chromosomes into *H. vulgare* L. Theor. Appl. Genet. 84, 466-472.
- Pickering, R., and P.A. Johnston, 2005: Recent progress in barley improvement using wild species of *Hordeum*. Cytog. Genome Res. **109**, 344-349.
- Pickering, R. A., A. M. Hill, M. Michel, G. M. Timmerman-Vaughan, 1995: The transfer of a powdery mildew resistance gene from *Hordeum bulbosum* L. to barley (*H. vulgare* L.) chromosome 2 (2I). Theor. Appl. Genet. **91**, 1288-1292.
- Pickering, R. A., B, J. Steffenson, A. M. Hill, and I. Borovkova, 1998: Association of leaf rust and powdery mildew resistance in a recombinant derived from a *Hordeum bulbosum* x *Hordeum bulbosum* hybrid. Plant Breeding **117**, 83-84.
- Pickering, R. A., S. Malyshev, G. Kunzel, P.A. Johnston, V. Korzun, M. Menke, and I. Schubert, 2000: Locating introgressions of *Hordeum bulbosum* chromatin within *H. vulgare* genome. Theor. Appl. Genet. 100, 27-31.
- Rubiales, D., and T. L. W. Carver, 2000: Defence reactions of *Hordeum chilense* accessions to three formae speciales of cereal powdery mildew fungi. Can. J. Bot. **78**, 1561-1570.
- SAS Institute, 1988: SAS user guide: Statistics. SAS Institute, Cary, N.C.
- Singh, A. K., B. G. Rossnagel, G. J. Scoles and R. A. Pickering, 2003: Inheritance of scald resistance from barley lines 4176/10/n/3/2/6 and 145L2. Can. J. Plant Sci. **83**, 417-422.

- Steffenson, B. J., Y. Jin and C. A. Griffey, 1993: Pathotypes of *Puccinia hordei* with virulence for the barley leaf rust resistance gene *Rph7* in the United Stats. Plant Dis. 77, 867-869.
- Thomas, H. M., and R. A. Pickering, 1983: Chromosome elimination in *Hordeum vulgare* X *H. bulbosum* hybrids 2. Chromosome behaviour in secondary hybrids. Theor. Appl. Genet. 44, 141-146.
- Torp, J., H.P. Jensen, and J.H. Jørgensen, 1978: Powdery mildew resistance genes in 106 northwest European spring barley varieties. Kgl. Vet. Landbo. Årsskr. **1978**, 75-102.
- Walther, U., H. Rapke, G. Proeseler, and G. Szigat, 2000: *Hordeum bulbosum* a new source of disease resistance - transfer of resistance to leaf rust and mosaic viruses from *H*. *bulbosum* into winter barley. Plant Breeding **119**, 215 - 218.
- Xu, J., and K. J. Kasha, 1992: Transfer of a dominant gene for powdery mildew resistance and DNA from *Hordeum bulbosum* into cultivated barley (*H. vulgare*). Theor. Appl. Genet. 84, 771-777.
- Xu, J., and J. W. Snape, 1989: The resistance of *Hordeum bulbosum* and its hybrids with *H. vulgare* to common fungal pathogens. Euphytica **41**, 273-276.
- Zadoks J. C., T. T. Chang and C. F. Konzak, 1974: A decimal code for the growth stages of cereals. Weed Res. 14, 415-421.

Lina anda	U unlagua porent	U hulbogum parant	Intrograssion logation
Line code 81882 ¹	H. vulgare parent		Introgression location
	Vada	S1	2HS
38U4/1/3/8	Golden Promise	2920/4	5HL, 6HS
38U4/1/3/9	Golden Promise	2920/4	6HS
38U4/1/3/10	Golden Promise	2920/4	6HS, 7HL
38U16	Golden Promise	2920/4	5HL
53A8 ¹	Golden Promise	2920/4	4HL
$182Q20^{1}$	Golden Promise	A17/1	2HL
$212Y1^{1}$	Golden Promise	2920/4	6HS, 7HS
102C2/14	Emir	2032	2HL
119Y4	Emir	2920/4x2929/1	6HS, 7HS, 7HL
171J1	Emir	2920/4X2929/1	6HS, 7HS
$177L20^{1}$	Emir	A17/1	7HL
181P158	Emir	A17/1	4HL
200A3 ¹	Emir	A17/1	2HS
120G4	Emir	2920/4x2929/1	6HS, 7HS
129F2	Emir	2920/4	4HL
169P15	Emir	A17/1	4HL
170R1	Emir	2920/4X2929/1	6HS
36L36	Emir	2920/4	2HS
38P18	Emir	2032	2HL
203S1	Emir	A17/1	5HL
216U3 ¹	Emir	A17/1	7HL
219W4 ¹	Emir	A17/1	7HL

Table 1. Barley recombinant lines (RLs) with introgressed DNA fragment from Hordeum

bulbosum used in the study.

¹ tested in Germany with 24 isolates of powdery mildew

Pathogen	Isolate	Country of origin	Virulence/avirulence factors
Leaf rust	CO-01	Spain	Rph1,2,4,6,8,12/3,5,7
	AL-02	Spain	Rph1,2,3,4,5,6,8,9,12/7
	1.2.1	Holland	Rph1,2,4,5,6,8,9/3,7,12
	IVP200	Holland	Rph1,2,4,5?,6,8,9,12/3,7
	TU-03	Tunisia	Rph1,2,3,4,5,6,7,8,9,12/7
Powdery	CO-02	Spain	<i>Mla1,a7,a8,a9,a10,a12,a22,a23,k,p,g,La,h/a3,a6,a13,a14,t,o5</i>
mildew	CC1	UK	<i>Mla7,a8,a9,a10,a12,a13,k,p,t,g,La,h/a1,a3,a6,a14,a22,a23,o5</i>
	1	Denmark	<i>Mla22,ra,k,nn,p,La/a1,a3,a6,a14,a7,a9,a10,a12,a13,at,g,h,o5</i>
	2	Denmark	Mla12,a22,nn,p,La,h/a1,a3,a6,a14,a7,a9,a10,a13,ra,k,at,g,o5
	3	Denmark	Mla1,a22,nn,p,at,La,h/a3,a6,a14,a7,a9,a10,a12,a13,ra,k,g,o5
	4	Germany	Mla6,a14,a22,ra,nn,p,La,h/a1,a3,a7,a9,a10,a12,a13,k,at,g,o5
	5	Denmark	Mla6,a14,a22,ra,nn,p,g,h/a1,a3,a7,a9,a10,a12,a13,k,at,La,o5
	6	Denmark	Mla9,a10,a22,k,nn,p,La,h/a1,a3,a6,a14,a7,a12,a13,ra,at,g,o5
	7	Denmark	Mla6,a14,a22,ra,nn,p,g,La,h/a1,a3,a7,a9,a10,a12,a13,k,at,o5
	8	Germany	Mla6,a14,a7,a12,a22,ra,nn,p,La,h/a1,a3,a9,a10,a13,k,at,g,o5
	9	Germany	Mla6,a14,a7,a22,ra,k,nn,p,g,La,h/a1,a3,a9,a10,a12,a13,at,o5
	10	Germany	Mla6,a14,a7,a12,a22,ra,nn,p,g,La,h/a1,a3,a9,a10,a13,k,at,o5
	11	Denmark	Mla3,a14,a6,a7,a22,ra,nn,p,g,La,h/a1,a9,a10,a12,a13,k,at,o5
	12	Denmark	<i>Mla7,a9,a10,a13,ra,k,nn,p,g,La,h/a1,a3,a6,a14,a12,a22,at,o5</i>
	13	Germany	Mla1,a7,a10,a12,ra,nn,p,g,La,h/a3,a6,a14,a9,a13,a22,k,at,o5
	14	Germany	Mla6,a14,a7,a10,a13,ra,k,nn,p,at,La,h/a1,a3,a9,a12,a22,g,o5
	15	Germany	Mla3,a6,a14,a7,a22,ra,nn,p,g,La,h/a1,a9,a10,a12,a13,k,at,o5
	16	Germany	Mla6,a14,a7,a13,a22,ra,k,nn,p,at,g,La,h/a1,a3,a9,a10,a12,o5
	17	Denmark	Mla6,a14,a7,a9,a12,a22,ra,k,nn,p,g,La,h/a1,a3,a10,a13,at,o5
	18	Denmark	<i>Mla3,a7,a9,a10,a12,ra,k,nn,p,at,g,La,h/a1,a6,a14,a13,a22,o5</i>
	19	Germany	<i>Mla6</i> , <i>a14</i> , <i>a7</i> , <i>a10</i> , <i>a12</i> , <i>a13</i> , <i>ra</i> , <i>k</i> , <i>nn</i> , <i>p</i> , <i>at</i> , <i>g</i> , <i>La</i> , <i>h</i> / <i>a1</i> , <i>a3</i> , <i>a9</i> , <i>a22</i> , <i>o5</i>
	20	Denmark	<i>Mla6</i> , <i>a14</i> , <i>a7</i> , <i>a9</i> , <i>a10</i> , <i>a12</i> , <i>a13</i> , <i>ra</i> , <i>k</i> , <i>nn</i> , <i>p</i> , <i>g</i> , <i>La</i> , <i>h</i> / <i>a1</i> , <i>a3</i> , <i>a22</i> , <i>at</i> , <i>o5</i>
	21	Germany	<i>Mla3,a6,a14,a7,a12,a13,a22,ra,nn,p,at,g,h/a1,a9,a10,k,La,o5</i>
	22	Germany	<i>Mla3,a6,a14,a7,a9,a10,a12,ra,k,nn,p,g,La,h/a1,a13,a22,at,o5</i>
	23	Austria	<i>Mla6</i> , <i>a14</i> , <i>a7</i> , <i>a9</i> , <i>a10</i> , <i>a12</i> , <i>a13</i> , <i>ra</i> , <i>k</i> , <i>nn</i> , <i>p</i> , <i>at</i> , <i>g</i> , <i>La</i> , <i>h</i> / <i>a1</i> , <i>a3</i> , <i>a22</i> , <i>o5</i>
	24	Germany	<i>Mla6,a14,a7,a9,a10,a12,a13,a22,ra,k,nn,p,g,La,h/a1,a3,at,o5</i>

Table 2. Virulence / avirulence factors of the leaf rust and powdery mildew isolates

									Ise	olates						
Barley	Genetic		CO-0)1		AL-0)2		TU-()3		1.2.	1		IVP20	000
Line	Background ¹	IT^2	RLP ³	RIF ³	IT	RLP	RIF	IT	RLP	RIF	IT	RLP	RIF	IT	RLP	RIF
Emir		9	$102 de^4$	97a	9	108c	105abc	9	107ef	131a	9	104cde	107ab	9	96e	60efg
102C2/14	Emir	1	<u>_</u> 5	—	4	—	—	1	—	—	1	—	—	1	—	—
119Y4	Emir	3	—	—	4	—	—	3	_	—	8	104cde	84bcd	4	—	—
171J1	Emir	9	103cd	50de	9	108c	98abc	9	115bc	45f	9	112b	74cde	9	104cde	94abcde
177L20	Emir	9	102de	88ab	9	111abc	105abc	9	106f	79bcde	9	103cde	80cd	9	105cde	68def
181P158	Emir	9	102de	75bc	9	114a	96abc	9	112cde	67cdef	8	110bc	70de	9	104cde	63efg
200A3	Emir	9	106bc	61cd	9	112abc	130a	9	109def	56def	7	110bc	55ef	9	114bc	115ab
120G4	Emir	7	107b	66c	9	110bc	121ab	9	116bc	56def	9	110bc	75cde	9	107cd	114ab
129F2	Emir	9	106bc	75bc	9	112ab	109abc	9	113cd	89bc	9	107bcd	64def	9	107cd	90abcde
169P15	Emir	1	_	_	1	_	_	1	_	_	1	_	_	0	—	_
170R1	Emir	9	107b	47e	9	111abc	114abc	9	119b	39f	9	110bc	50ef	9	108cd	122a
36L36	Emir	6	_	_	9	115a	82bcde	9	103fg	64cdef	6	_	_	9	114bc	66def
38P18	Emir	1	_	_	4	_	_	1	_ 0	_	1	_	_	1	—	_
203S1	Emir	9	102de	64cd	9	110bc	91abcd	9	108def	53def	9	103cde	84bcd	9	104cde	93abcde
216U3	Emir	9	101de	71c	9	112abc	83abcde	9	103fg	80bcd	9	103de	111a	9	104cde	110abc
219W4	Emir	9	106b	47e	9	114a	62e	6	_	_	9	107bcde	97abc	9	107cde	81bcde
Vada		9	115a	29f	9	116a	81abcde	9	127a	51ef	9	138a	44f	9	119ab	70def
L94		9	100e	100a	9	100d	100abc	9	100g	100b	9	100e	100abc	9	100de	100abcd

Table 3. Infection type (IT), relative latency period (RLP), and relative infection frequency (RIF) of five isolates of *Puccinia hordei* on barley recombinant lines with DNA segments introgressed from *Hordeum bulbosum*

Continue Table 3.

									Isolate	es						
Barley	Genetic		CO-01	l		AL-02	2		TU-03	;		1.2.1			IVP20	00
Line	Background ¹	IT^2	RLP ³	RIF^3	IT	RLP	RIF	IT	RLP	RIF	IT	RLP	RIF	IT	RLP	RIF
Vada		9	115a ⁴	29c	9	116a	81ab	9	127a	51b	9	138a	44c	9	119a	70b
81882	Vada	8	101b	50b	9	116a	82ab	9	111b	71b	8	124b	32c	9	115b	71b
L94		9	100b	100a	9	100b	100a	9	100c	100a	9	100c	100a	9	100d	100a
Golden Promise	2	9	102de	77b	9	107b	81ab	9	105cd	127a	9	106cde	81bc	9	107d	60b
38U16	Golden Promise	8	110bc	71b	9	103de	82ab	9	110c	61b	9	105cde	86ab	9	116c	57bc
53A8	Golden Promise	9	110bc	75b	9	108cd	80ab	5	5	—	9	103de	67cd	9	122ab	51bc
182Q20	Golden Promise	9	112ab	27d	9	121a	41d	0(9)	—	_	9	140a	7g	9	119bc	37c
38U4/1/3/8	Golden Promise	9	103de	24d	8	124a	66bc	9	121ab	24b	8	123b	25f	7	120bc	61b
38U4/1/3/9	Golden Promise	9	107c	48c	9	118a	67bc	9	114bc	44b	9	114bcd	59de	9	122ab	58bc
38U4/1/3/10	Golden Promise	9	113ab	47c	9	116a	78abc	9	109cd	51b	9	117bc	48e	9	117bc	54bc
212Y1	Golden Promise	9	104d	73b	9	118a	57cd	9	109cd	58b	9	111cde	66cd	9	126a	64b
Vada		9	115a	29cd	9	116a	81ab	9	127a	51b	9	138a	44e	9	119bc	70b
L94		9	100e	100a	9	100c	100a	9	100d	100a	9	100e	100a	9	100e	100a
			(138h)	(54)		(140h)	(55)		(180h)	(18)		(169h)	(69)		(135h)	(69)

¹ RLs are separated to groups according to their genetic background. ² IT on a scale of 0 to 9 (McNeal et al. 1971). ³ Relative latency period (RLP) and relative infection frequency (RIF) referred to L94 = 100 %. The actual values for L94 with each isolate are indicated in the table between brackets.

⁴Data with the same letter per column per group do not differ significantly (Duncan, $P \le 0.05$). ⁵Could not be determined because of low number of uredia due to low IT.

									Isola	ates						
	Genetic		CO-0	1		AL-0	2		TU-03	3		1.2.1			IVP200	00
Barley line	Background ¹	$EA+^2$	EA- ²	CS^2	EA+	EA-	CS	EA+	EA-	CS	EA+	EA-	CS	EA+	EA-	CS
Vada		0.3a ³	40.3a	0.016b	0.0a	4.8a	0.135b	0.0b	28.8ab	0.019b	0.3a	31.3a	0.035c	0.7a	20.0a	0.115b
81882	Vada	2.2a	31.3a	0.014b	0.0a	3.6a	0.113c	5.2a	23.6b	0.016b	0.0a	18.8b	0.064b	0.0a	19.5a	0.142b
L94		0.0a	0.0b	0.075a	0.0a	0.0a	0.222a	1.1ab	0.0c	0.054a	0.0a	0.0c	0.184a	0.0a	0.0b	0.284a
Golden Promise		0.6b	2.8d	0.049b	0.0a	0.0b	0.138b	0.0c	24.1b	0.040b	0.7b	0.0e	0.117b	2.5a	9.3bc	0.159b
53A8	Golden Promise	0.7b	16.5c	0.025c	0.0a	0.0b	0.134c	70.6a	0.0c	4	0.0b	3.5de	0.099b	5.8a	4.4c	0.103bc
182Q20	Golden Promise	9.9a	85.1a	0.011e	0.0a	26.1a	0.075bc	7.7b	88.7a	0.008c	3.0a	74.3a	0.073c	0.0a	45.1a	0.105bc
38U4/1/3/8	Golden Promise	0.6b	32.3b	0.013de	0.0a	28.2a	0.065bc	0.0c	32.1b	0.016c	1.8ab	30.7b	0.056c	2.6a	34.0ab	0.105bc
38U4/1/3/9	Golden Promise	2.1b	9.7cd	0.016de	0.0a	3.7b	0.103bc	0.0c	35.7b	0.018c	0.0b	20.5c	0.072c	0.0a	29.1ab	0.101bc
38U4/1/3/10	Golden Promise	7.9a	16.2c	0.026c	1.3a	31.1a	0.079bc	0.0c	16.2b	0.016c	0.0b	16.9c	0.056c	0.0a	16.6bc	0.093bc
212Y1	Golden Promise	1.2b	5.4cd	0.022cd	0.0a	6.8b	0.101bc	2.5c	31.0b	0.019c	0.0b	9.0d	0.075c	0.7a	51.1a	0.084c
Vada		0.3b	40.3b	0.016de	0.0a	4.8b	0.135bc	0.0c	28.8b	0.019c	0.3b	31.3b	0.035d	0.7a	20.0bc	0.115bc
L94		0.0b	0.0d	0.075a	0.0a	0.0b	0.222a	1.1c	0.0c	0.054a	0.0b	0.0e	0.184a	0.0a	0.0c	0.284a

Table 4. Microscopic components of resistance to five isolates of *Puccinia hordei* in barley recombinant lines (RLs) with DNA segments introgressed from *Hordeum bulbosum*

Continue Table 4.

									Isolates	5						
Barley	Genetic		CO-01			AL-02	2		TU-03	5		1.2.1			IVP20	00
line	Background ¹	$EA+^2$	EA- ²	CS^2	EA+	EA-	CS	EA+	EA-	CS	EA+	EA-	CS	EA+	EA-	CS
Emir		0.7e ³	2.7de	0.041b	0.0d	0.0b	0.028a	0.8d	9.1bc	0.091a	1.0d	0.0c	0.225a	0.0c	0.7d	0.235ab
102C2	Emir	95.5a	4.1cde	4	38.4c	2.1b	_	95.2ab	4.8bc	_	96.5a	3.5bc	_	61.2a	11.0b	—
119Y4	Emir	51.6c	24.3b	—	1.9d	0.0c	—	34.1c	4.3bc	—	0.0d	0.0c	0.113c	2.0c	1.0d	—
169P15	Emir	67.1b	7.0cde	—	100a	0.0c	—	85.8b	12.0b	—	87.7b	3.3bc	—	40.3b	1.0d	—
36L36	Emir	36.9d	11.9c	—	0.0d	0.0c	0.140b	10.0d	11.3b	0.049ab	21.0c	2.4bc	—	0.0c	1.8cd	0.219abc
38P18	Emir	95.0a	3.7cde	—	61.3b	0.7c	—	100a	0.0c	_	94.0ab	5.7bc	—	59.0a	11.7b	—
219W4	Emir	0.0e	2.6de	0.038b	1.3d	0.0c	0.142b	4.3d	4.3bc	_	1.0d	7.1b	0.119c	0.0c	0.0d	0.178bc
Vada		0.3e	40.3a	0.016c	0.0d	4.8a	0.135b	0.0d	28.8a	0.019b	0.3d	31.3a	0.035d	0.7c	20.0a	0.115d
L94		0.0e	0.0e	0.075a	0.0d	0.0c	0.222a	1.1d	0.0c	0.054ab	0.0d	0.0c	0.184b	0.0c	0.0d	0.284a

¹RLs are separated to groups according to their genetic background. ² Expressed are percentage of early aborted colonies associated with plant cell necrosis (EA+), percentage of early aborted colonies without plant cell necrosis (EA-) and colony size in mm² (CS). ³ Data with the same letter per column per group do not differ significantly (Duncan, $P \le 0.05$). ⁴ CS could not be measured because of plant cell necrosis.

		Isolates											
	Genetic		CC1			CO-()2						
Barley line	Background ¹	IT^2	DS^3	IF^{3}	IT	DS	IF						
Vada		4	$14a^4$	25a	4	75a	91a						
81882	Vada	0	0b	0 b	2	41b	54b						
Golden Promise		4	23ab	35abc	4	56ab	64b						
38U4/1/3/10	Golden Promise	4	25ab	35abc	4	63a	72ab						
38U16	Golden Promise	4	27ab	31bc	4	54ab	64ab						
53A8	Golden Promise	4	15b	22bc	4	48ab	53b						
182Q20	Golden Promise	4	18b	15c	4	51ab	67ab						
38U4/1/3/8	Golden Promise	4	31a	56a	4	66a	82a						
38U4/1/3/9	Golden Promise	4	25ab	38ab	4	66a	79a						
212Y1	Golden Promise	4	18b	17bc	4	46b	62ab						
Emir		4	34ab	67a	4	45bc	57b						
102C2/14	Emir	4	21c	27c	4	35cd	46bcd						
119Y4	Emir	4	34ab	54ab	4	56ab	77a						
171J1	Emir	4	26bc	32bc	4	63a	81a						
177L20	Emir	0	0d	0d	0(4)	3e	5e						
181P158	Emir	4	26bc	46abc	4	36cd	48bcd						
200 A3	Emir	4	22bc	40bc	4	24d	38cd						
120G4	Emir	4	31abc	55ab	4	65a	80a						
129F2	Emir	4	25bc	39bc	4	27d	43bcd						
169P15	Emir	4	24bc	44abc	4	23d	35d						
170R1	Emir	4	26bc	56ab	4	65a	77a						
36L36	Emir	4	40a	69a	4	32cd	49bcd						
38P18	Emir	4	24bc	50abc	4	42bc	52bc						
203S1	Emir	4	25bc	28c	4	42bc	57b						
216U3	Emir	0	0d	0d	0	0e	0e						
219W4	Emir	0	0d	0d	0	0e	0e						

Table 5. Infection type (IT), disease severity (DS), and infection frequency (IF), of two isolates of powdery mildew on barley recombinant lines (RLs) with DNA segments introgressed from Hordeum bulbosum

¹ RLs are separated to groups according to their genetic background. ² Infection type (IT) on a scale of 0-4 (Moseman 1965).

3 Disease severity (DS) estimated as the percentage of leaf area covered by powdery mildew colonies, infection frequency (IF) calculated as number of powdery mildew colonies per cm².

⁴ Data with the same letter per column per group do not differ significantly (Duncan, $P \le 0.05$).

Barley line	Gene(s)													Isola	ate										
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
Vada	MlLa	3	3	4	3	2	3	3	3	3	3	4	4	4	3	3	3	4	4	3	4	2	4	4	3
81882		1	1	2	1	0	0	1	2	2	2	2	1	1	1	2	2	2	2	1	0	0	2	1	1
Vada	MlLa	3	3	4	3	2	3	3	3	3	3	4	4	4	3	3	3	4	4	3	4	2	4	4	3
81882		1	1	2	1	0	0	1	2	2	2	2	1	1	1	2	2	2	2	1	0	0	2	1	1
Golden Promise	none	4	4	4	4	4	4	3	4	4	4	4	4	4	4	4	3	4	4	4	4	4	4	4	4
53A8		4	4	4	4	3	3	4	3	4	4	4	3	4	4	4	4	4	4	4	4	4	3	4	4
182Q20		4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	3	3	4	4
212Y1		4	2	4	3	2	3	2	3	3	3	3	3	3	4	3	4	3	4	3	2	2	2	3	3
Emir	Mla12	1	3	3	2	1	3	2	3	2	4	2	2	4	2	2	2	4	4	4	3	3	4	4	4
177L20		0	2	0	0	0	2	0	1	0	2	0	0	1	2	0	0	4	2	3	2	2	0	2	2
200A3		0	0	0	0	0	0	0	0	1	0	0	0	1	0	0	0	3	2	0	0	2	2	2	2
216U3		0	1	0	4	0	2	0	2	0	2	0	4	1	0	1	1	3	2	3	3	0	0	2	0
219W4		0	0	0	1	0	1	0	1	0	0	0	0	2	1	0	0	3	0	3	0	1	0	0	0

Table 6. Infection type¹ on eight barley recombinant lines with DNA segments introgressed from *Hordeum bulbosum* after inoculation with 24 isolates of *Blumeria graminis* f.sp. *hordei*.

¹ Infection type (IT) on a scale of 0-4 were 0-2 = resistant and 3-4 = susceptible (Moseman 1965).

		Isolates											
	Genetic		CC1		_	CO-02							
Barley line	Background	$EA+^1$	EA- ¹	\mathbf{EST}_{1}	EA+	EA-	EST-						
Vada		$2.4a^{2}$	72.9b	24.7a	0.0b	76.3b	23.7a						
81882	Vada	3.3a	96.7a	0.0b	6.0a	84.1a	9.9b						
Emir		2.0c	71.7c	26.3a	0.0a	89.6b	10.4a						
177L20	Emir	7.1b	91.1a	1.8b	0.0a	95.2a	4.8b						
216U3	Emir	8.4b	90.8a	0.8b	0.0a	97.7a	2.3c						
219W4	Emir	18.2a	78.9b	2.9b	1.8a	98.2a	0.0c						

Table 7. Microscopic components of resistance to Blumeria graminis f.sp. hordei in barley recombinant lines (RLs) with introgressed segments from Hordeum bulbosum.

¹ Expressed as percentages of early aborted colonies associated with host cell necrosis (EA+), percentage of early aborted colonies without host cell necrosis (EA-) and established colonies without host cell necrosis (EST-). ² Data with the same letter per column do not differ significantly (Duncan, $P \le 0.05$).