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Unraveling possible association between quantitative trait loci (QTL) for partial resistance and nonhost resistance in food barley (*Hordeum vulgaris* L.)

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Many quantitative trait loci (QTLs) in different barley populations were discovered for resistance to *Puccinia hordei* and heterologous rust species. Partial resistance (host basal resistance) and nonhost resistance (nonhost basal resistance) in barley to leaf rusts are based on prehaustorial mechanism of resistance which is associated with papillae formation. They are mainly governed by genes with relatively small, quantitative effects, located on QTL. The genes for host basal resistance seem to play similar roles in basal resistance as those governing nonhost basal resistances. From different studies it was observed that these two resistance types are based on shared principles. Four and two quantitative trait loci-near isogenic lines (QTL-NILs), respectively, were developed for basal resistance and nonhost resistance QTLs using SusPtrit as recurrent parent. SusPtrit is a research line which is exceptionally susceptible to leaf rusts for which normally barley is a nonhost. They were infected with one homologous (*P. hordei* isolate 1.2.1) and three heterologous (*Puccinia* triticina isolate 'Flamingo', *P. hordei*-murini and *P. hordei*-secalini) leaf rusts in three replications at seedling stage to evaluate whether relatively large-effect QTLs show specificity in their reaction to homologous and heterologous rust isolates. The result showed that, the QTLs for host basal resistance and the QTL for nonhost basal resistance have a significant ($P<0.05$) effect on both homologous and heterologous rusts. Also, higher positive ($P<0.05$, $r = 0.98$) correlation was observed between different macroscopic and microscopic parameters measured indicating that there is a possible association between partial resistance quantitative trait loci and nonhost resistance quantitative trait loci. These give an indication that indeed, host and nonhost basal resistance are associated.

Key words: Partial resistance, nonhost resistance, near isogenic lines, barley.

INTRODUCTION

Naturally, most plant species are resistant to most pests and pathogen species. Such form of resistance is called non-host resistance which is commonly defined as resistance expressed by the whole plant species to all genetic variants of a non-adapted pathogen species or forma species (f.sp.) and represents the most durable form of plant resistance (Lipka et al., 2010; Schulze-Lefert and Panstruga, 2011). However, partially resistant genotypes can still be infected by the pathogen and show reduced rate of infection compared to susceptible

genotypes, which is an outcome of lower rate of colonization by the fungus (Niks and Rubiales, 2002; Niks and Marcel, 2009). Many factors contribute to nonhost resistance to unadapted pathogens, including constitutive defenses and induced defenses (Uma et al., 2011; Fan and Doerner, 2012). The possibility of transferring nonhost resistance to crop plants enabled the study of its association with host resistance. There are a number of examples of successful transfer of such resistance from nonhost to host species (Wulff et al., 2011). An important question will be whether there is association between genes potentially involved in nonhost resistance and host resistance and genes involved in nonhost resistance deploying them into host plants will result in durable resistance.

As QTLs can confer certain level of resistance by their cumulative effects, the adaptation of a pathogen and breakdown of each QTL resistance-allele is more difficult than breakdown of one monogenic resistance gene (Lindhout, 2002), particularly when each QTL-gene encodes a different gene product.

Near-isogenic lines (NILs) differing with regard to disease QTLs provide valuable material for a more detailed study into the genetic basis of quantitative resistance. Thus, development of such NILs allows the evaluation of a QTL in a nearly uniform genetic background, overcoming the difficulties of identifying QTL phenotypes (Marcel et al., 2007). QTL-NILs do not only provide a better estimate for the effect of single QTL alleles, but also provide a better insight into QTL x pathogen and QTL x environment interactions. Furthermore, QTL-NILs may provide a starting point for the unraveling of functional genes underlying these loci and possibly be useful for positional cloning (van Berloo et al., 2001).

For the purpose of evaluation of a QTLs genetic basis for quantitative resistance, five QTL-near isogenic lines (QTL-NILs) were developed by introgressing four host basal resistance QTLs and a nonhost resistance QTL into SusPtrit genetic background. SusPtrit is an experimental line which not only susceptible to the homologous rust, *P. hordei*, but also exceptionally susceptible to leaf rusts for which normally barley is a nonhost. The QTL-NILs developed allow us to study the possible association between the QTLs for host basal resistance and nonhost resistance (Marcel et al., 2007).

Therefore, this study was conducted to evaluate whether relatively large-effect QTLs show association in their reaction to homologous and heterologous rust isolates, and also, to evaluate their mechanism of resistance at tissue and cell level, especially whether the resistance is based on hypersensitivity or non-hypersensitivity.

Table 1. QTL-NILs used in this study.

QTL-NILs	Donor line
Vada-Rphq2	Vada
Vada-Rphq3	Vada
L-94-Rphq2	L-94*Vada
L-94-Rphq3	L-94*Vada
Su-Rphq2	SusPtrit*Vada
Su-Rphq3	SusPtrit*Vada
Su-Rphq11	SusPtrit*Steptoe
Su-Rphq16	SusPtrit*Dom
Su-Qnh.L	SusPtrit*L94
Su-Qnh.V	SusPtrit*Vada

MATERIALS AND METHODS

All experiments were conducted in Plant Research International (PRI) laboratory and greenhouse at Wageningen University, The Netherlands.

Plant materials

Near isogenic lines (NILs) with SusPtrit genetic background (Table 1) and having resistance QTLs, *Rphq2*, *Rphq3*, *Rphq11, Rphq16* and *Rnhq* (*Rnhq-V* and *Rnhq-L*), were used for this study.

The parental lines for each respective NILs were used as a reference. In addition, L94-NILs (L94-*Rphq2* and *-Rphq3*) and Vada-NILs (Vada-*rphq2*, and *-rphq3*) were included as well. For the histology assays, host plants corresponding to the rust species under observation were added as a reference.

Inoculum

Four isolates of rust fungi were used in infection studies (Table 2). They were multiplied on their respective host species. Urediniospores were collected and dried in desiccators for 5 to 7 days before used for inoculation.

Genotyping QTL-NILs

Genotyping of each QTL-NILs was performed before sowing to confirm the introgression of donor resistant allele by using molecular markers flanking the QTL region (Table 3). For this purpose DNA of each QTL-NILs was isolated following the CTAB isolation method according to Wang et al. (1993).

Phenotyping QTL-NILs with homologous rust isolate

Seeds of QTLs-NILs were sown in 37 × 39 cm boxes in two rows along with reference lines. Depending on the availability of seeds, 1 to 2 seeds were sown for each NIL. The secondary leaves were clipped out and the fully grown primary leaves were fixed horizontally with adaxial side up in an inoculation tower and

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Table 2. Rust isolates used in this study.

Pathogens	Host plant	Common name
P. hordei isolate 1.2.1	*Hordeum vulgare*	Barley leaf rust
P. hordei-murini	*H. murinum*	Wall barley leaf rust
P. hordei-secalini	*H.secalinum*	Meadow barley leaf rust
P. triticina isolate "Flamingo"	*T.aestivum*	Wheat leaf rust

Table 3. List of markers used in genotyping of QTL-NILs.

Name(s)	Chrom.	Type	Tm (°C)	RE(s)	Linked QTL	Source
besV76P5D5AR	2H	ASPCR	56	-	*Rphq2*	
k00345	2H	CAPS	56	*Sdu I*	*Rphq2*	Marcel et al., 2008
scP15M51-204	2H	SCAR	56		*Rphq2*	Marcel et al., 2007
ABG388	6H	CAPS	58	*Nla III*	*Rphq3*	Marcel et al., 2007
WBE201	6H	CAPS	58	*Mnl I*	*Rphq3*	Marcel et al., 2007
GBM1212	6H	SSR			*Rphq3*	Marcel et al., 2007
HVM14	6H	SSR			*Rphq3*	Marcel et al., 2007
GBS0512	2H	CAPS	58	*Aci I - (Hpy99 I)*	*Rphq11*	Marcel et al., 2007
TC134748	2H	CAPS	47	*ApoI = XapI*	*Rphq11*	Rice synteny
GBM1062	2H	SSR	55		*Rphq11*	Marcel et al., 2007
GBMS244	2H	SSR	55		*Rphq11*	Marcel et al., 2007
DsT-33	5H	SCAR	45	-	*Rphq16*	Marcel et al., 2007
Scsnp03275_2	5H	CAPS	65	*BglII*	*Rphq16*	Marcel et al., 2007
GMS002	5H	SSR			*Rphq16*	
MWG2031	1H	CAPS	55	*Mwo I*	*Rnhq*	Kikuchi et al, 2004
SKT1	1H	CAPS	60	*Alu I*	*Rnhq*	Kikuchi et al., 2004
WBE101	1H	CAPS	52	*HpyCH4 IV*	*Rnhq*	Marcel et al., 2007
GBM1303	1H	SSR			*Rnhq*	Marcel et al., 2007

inoculated with 3.5 g of freshly collected spores of *P. hordei* isolate 1.2.1 diluted 10 times with lycopodium spores to obtain uniform spore distribution. The inoculated boxes were placed in a humidity chamber to incubate the spores overnight for eight hours at 100% relative humidity in the dark at 18°C. After incubation, the inoculated boxes were transferred to a greenhouse compartment where the temperature is set at 14 ± 3°C with 30 to 70% relative humidity. The experiment was carried out in two replications.

The latent period (LP) was measured one week after inoculation. The LP50S was then calculated with the following formula:

$$LP50 = T1 + (T2 - T1) \times \frac{(N100-2-N1)}{(N2-N1)}$$

T1 = the time just before 50% of the pustules are mature; T2 = the time just after 50% of the pustules are mature; N1 = number of mature pustules at T_1; N2 = number of mature pustules at T_2; N100/2 = half of the total mature pustules number.

To normalize the results, the relative latency period (RLP50S) was calculated relative to the LP50S of SusPtrit which set at 100.

Phenotyping QTL-NILs with heterologous rust isolates

The QTL-NILs were grown as described above except that in this case susceptible host plants were included. Ten to twelve days after sowing, completely unfolded primary leaves were fixed horizontally with the adaxial side up, and inoculated with 10 mg of spores per box using a settling tower (Atienza et al., 2004). For each QTL-NIL and reference lines, two seedlings were inoculated of which one seedling was sampled for histological studies. The second seedling was used for macroscopic phenotyping. To avoid cross contamination of rusts, the settling tower and other tools were cleaned with 70% ethanol before and after use. Latency period (LP) was measured six days (*Phs*) and eight days (*Phm* and *P. triticina*) after inoculation. Additionally, the level of infection was quantified by estimating the following traits: Infection frequency (IF, pustules/cm^2), flecks (F, non- sporulating infection sites/cm^2), frequency of visible infection sites (VIF, IF/total amount of visible infection sites/cm^2), and TotF (total amount of visible infection sites/cm^2) by using a metal frame with 1 cm^2 window. RLP50S and Relative Infection Frequency (RIF) were calculated by setting the RLP50S and RIF of SusPtrit to 100 as stated above. Analysis of variance for both RLP50S and RIF was carried out using GenStat statistical software (11.1th edition). All genotypes were grown in one box and the experiment was carried out in two replications.

Histological evaluation of pathogenesis

Histology is the study of the microanatomy of cells and tissues of plants. It is commonly performed by examining cells and tissues under microscope. The specimen having been sectioned (cut into a

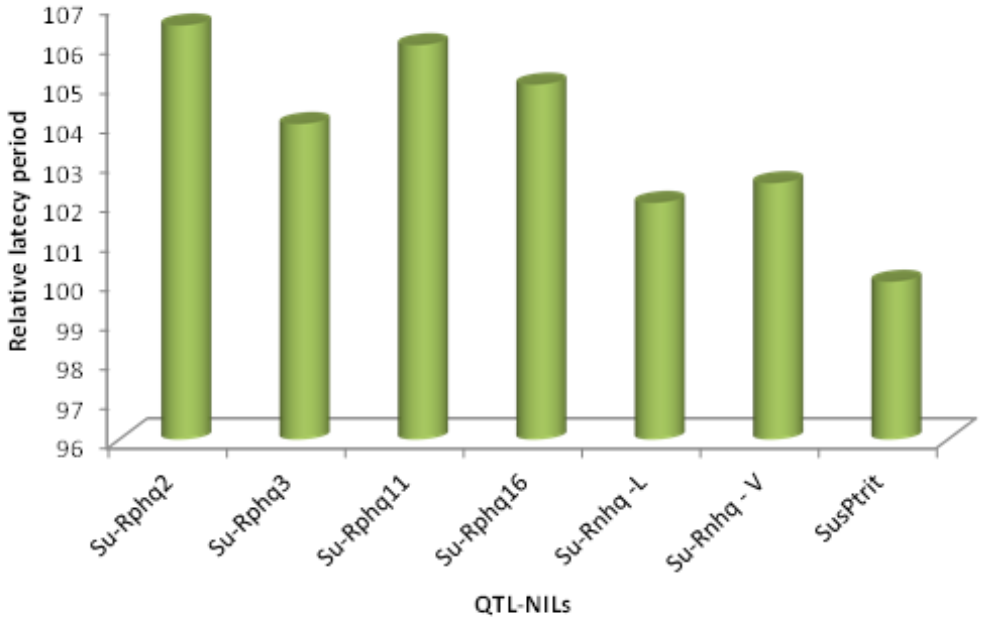

Figure 1. RLP50S of *P. hordei* isolated 1.2.1 on SusPtrit NILs and the reference lines at seedling stage (Source my own result of research data 2012).

thin cross section), stained, and mounted on a microscope slide.

Preparation of leaf samples for fluorescence microscopy

Seven days after inoculation, leaf segments of approximately 2 to 3 cm long cut out from the middle of primary leaf. These leaf segments were prepared as whole mount for fluorescence microscopy, except that Uvitex 2B (Ciba-Geigy) was used instead of Calcofluor. The leaf segments were immediately fixed and bleached by boiling for 1.5 min in a water bath in lactophenol-ethanol (1:2 v/v). After the leaves were bleached, the lactophenol-ethanol was poured off and they were washed once for 30 min in ethanol (50%) and in 0.05 N NaOH (2 g/l), respectively one after the other. The washed leaf segments were rinsed three times in water and soaked for 30 min in 0.1 M Tris/HCl buffer (pH 8.5). After 5 min of staining in a solution of 0.1% Uvitex in the same buffer, they were rinsed thoroughly for four times in water and then washed for 30 min in a solution of 25% glycerol. Finally, to prepare the slides, small drops of glycerol were added on the slide before the leaf samples were put along the longitudinal axis of the slide and the leaves samples were embedded on slide with the adaxial side facing up. Then the slide cover was carefully placed on the samples.

Observation of infection units under UV-microscope

For ease of inspection, different classes of infection units were set based on status of infection unit, where an infection unit is described as non-penetrating (NP), early aborted (EA) and established (Jafary et al., 2006). The detailed observation and scoring were done with a 10×10 and 40×10 magnification. The preparations were screened starting from one of the corners and moving horizontally along longitudinal axis of the leaves. The outmost stomata rows were excluded from observation to avoid possible border effects. Also overlapping and infection points close to air bubble were ignored. The infection hyphae were scored as "established" type (more than six haustorial mother cells) or "early aborted" type (having six or less haustorial mother cells). The data collected were analyzed using GenStat statistical software.

RESULTS AND DISCUSSION

Effects of PR and nonhost resistance QTLs towards homologous rust *P. hordei* isolate 1.2.1

QTL-NILs with PR and nonhost resistance QTLs had longer LP and RLP50S than susceptible check line, SusPtrit (Figure 1).

Against *P. hordei* isolate 1.2.1, *Rphq2*, *Rphq3, Rphq11* and *Rphq16* significantly ($P<0.05$) increase the LP observed in Su-*Rphq2*, Su-*Rphq3,* Su-*Rphq11* and Su-*Rphq16* as compared to SusPtrit. The effect of *Rphq3* in the seedling stage was smaller than that of the other three partial resistant (PR) QTLs (Figure 1). This is in agreement with the previous studies (Qi et al., 1999; Marcel et al., 2007). On the other hand, the effects of non-host QTLs (both on Su-*Rnhq-L* and Su-*Rnhq-V*) were not statistically significant. However, they have positive effect on resistance towards *P. hordei*.

SusPtrit NIL with PR QTLs had longer LP than SusPtrit, indicating that, these QTLs contributed to the effect on PR. Furthermore, it had an effect on resistance in Vada and L94 NILs (Data not shown). On the other hand, *Rnhq* QTL on Su-*Rnhq-V* had relatively more positive effect on resistance than that of Su-*Rnhq*-L. Likewise, both Su-*Rnhq-V* and Su-*Rnhq*-L had higher RLP50S than SusPtrit, indicating that *Rnhq* had an effect towards homologous rust.

Table 4. Mean relative latency period (RLP), relative infection frequency (RIF), percentage of non-penetrating (NP), early aborted (EA) and established (Est.) infection units by *P. triticina, P. hordei-murini* and *P. hordei-secalini* of tested lines.

QTL-NILs	*P. triticina* (Flamingo)					*P. hordei-murini*					*P. hordei-secalini*				
	RLP	**RIF**	**NP**	**EA**	**Est.**	**RLP**	**RIF**	**NP**	**EA**	**Est.**	**RLP**	**RIF**	**NP**	**EA**	**Est.**
Vada-*rphq2*	106.6^{ab}	6.4^{a}	24.2^{e}	69.5^{f}	4.7^{a}	103.4^{a}	9.1^{a}	15.4^{cd}	70.6^{f}	8.0^{a}	109.5^{c}	4.6^{a}	17.3^{a}	76.3^{g}	8.0^{a}
Vada-*rphq3*	117.3^{d}	7.2^{a}	24.5^{e}	66.4^{f}	9.3^{b}	103.5^{a}	11.0^{a}	16.0^{d}	73.8^{ef}	10.6^{b}	114.1^{c}	15.2^{b}	15.5^{a}	74.1^{fg}	10.4^{bc}
L94-*Rphq2*	113.9^{cd}	38.8^{b}	14.0^{bcd}	48.0^{e}	19.8^{c}	104.7^{a}	30.0^{b}	11.7^{bc}	68.9^{e}	10.0^{ab}	103.7^{ab}	42.5^{de}	12.0^{a}	70.0^{ef}	9.0^{ab}
L94-*Rphq3*	108.0^{bc}	33.4^{b}	12.0^{bc}	39^{d}	24.5^{d}	103.3^{a}	55.9^{e}	11.7^{bc}	59.6^{d}	14.3^{c}	102.7^{ab}	32.6^{c}	11.0^{a}	64.5^{de}	12.3^{cd}
Su-*Rphq2*	106.5^{ab}	37.8^{b}	16.0^{cd}	34.5^{cd}	24.5^{d}	104.5^{a}	30.3^{b}	14.7^{cd}	56.1^{cd}	14.8^{c}	104.9^{b}	31.2^{c}	14.6^{a}	63.3^{d}	12.0^{cd}
Su-*Rphq3*	106.0^{ab}	55.8^{c}	17.0^{d}	31.5^{bc}	25.8^{de}	10.9^{a}	39.7^{cd}	12.3^{bcd}	56.0^{cd}	15.9^{cd}	102.0^{ab}	29.4^{c}	12.5^{a}	56.3^{c}	14.7^{e}
Su-*Rphq11*	106.4^{ab}	57.7^{c}	16.0^{cd}	32.3^{bc}	25.9^{de}	104.2^{a}	31.2^{bc}	13.2^{bcd}	52.7^{c}	17.2^{d}	104.0^{ab}	39.1^{d}	14.5^{a}	57.0^{c}	13.9^{de}
Su-*Rphq16*	104.4^{ab}	63.0^{c}	15.0^{cd}	28.0^{b}	28.5^{e}	102.4^{a}	40.7^{d}	14.3^{cd}	45.2^{b}	20.3^{e}	102.2^{ab}	40.8^{be}	14.0^{a}	55.5^{c}	15.1^{e}
Su-*Rnhq-L*	100.6^{a}	64.2^{c}	10.0^{ab}	22.5^{a}	33.7^{f}	103.2^{a}	55.3^{e}	10.3^{ab}	37.8^{a}	26.1^{f}	102.0^{ab}	57.1^{f}	9.0^{a}	39.8^{b}	25.6^{g}
Su-*Rnhq-V*	102.6^{ab}	64.2^{c}	13.0^{bcd}	30.0^{bc}	28.3^{e}	103.0^{a}	44.5^{d}	12.2^{bcd}	38.5^{a}	24.7^{f}	102.9^{ab}	45.7^{e}	12.0^{a}	43.2^{b}	22.4^{f}
SusPtrit	100.0^{a}	100.0^{d}	7.0^{a}	21.0^{a}	36.0^{f}	100.0^{a}	100.0^{f}	7.0^{a}	34.2^{a}	29.7^{g}	100.0^{a}	100.0^{g}	11.2^{a}	26.4^{a}	30.9^{h}
Mean	**106.6**	**50.0**	**15.3**	**38.4**	**23.7**	**103.6**	**40.7**	**12.6**	**54.5**	**17.4**	**104.1**	**41.5**	**13.1**	**56.9**	**15.8**
CV (%)	**2.8**	**7.6**	**13.4**	**6.4**	**5.5**	**1.8**	**10.1**	**14.0**	**6.2**	**5.3**	**1.9**	**6.2**	**15.4**	**4.5**	**6.9**
LSD (5%)	**6.6**	**8.9**	**4.5**	**5.4**	**2.9**	**4.0**	**9.0**	**3.9**	**4.3**	**2.0**	**4.5**	**6.0**	**4.4**	**5.7**	**2.3**

Effects of PR and nonhost resistance QTLs towards heterologous rust isolates

The macroscopic observation revealed that all SusPtrit NILs had lower RIF and longer LP as compared to SusPtrit. The RIF of SusPtrit NILs with PR QTLs ranges from 38% (Su-*Rphq2*) to 63% (Su-*Rphq16*) on seedlings infected by *P. triticina*. As shown in Table 4, lower RIF were observed in lines with *Rphq2*. Nonhost resistance QTLs, showed nonhost resistance on Su-*Rnhq-V,* having 45% RLF for both *Phm* and *Phs,* while the percentage was the same with Su-*Rnhq-L* for *P. triticina*.

Concerning, microscopic observations, the distribution of infection units type based on counting of ≥50 infection units per plant revealed differences between QTL-NILs and SusPtrit for *P. triticina, Phs,* and *Phm*. These differences could be in proportion of NP, EA, formation of sporogenic tissue and colony size.

On the SusPtrit NILs it was observed that the percentage of non-penetrating (NP) infection units (this non-penetration of infection units was recognized by the absence of substomatal vesicle (SSV)) of *P. triticina* and *Phm* on average, were almost twice as high as in the susceptible line, SusPtrit. For *Phs* on the other hand, the percentage of NP infection units showed no significant differences (Table 4). Higher percentage of NP was observed in Su-*Rphq3* for *P. triticina*, while for *Phm* and *Phs* higher values were in Su-*Rphq2.* In case of nonhost SusPtrit NILs, Su-*Rnhq-V* had higher percentage of NP than that of Su-*Rnhq-L* for the three rusts species tested (Table 4).

Of the four infection unit classes studied in this experiment, it seems that EA and established infection units appeared to be the parameters with the largest contrasts between lines to see the existing variation among the NILs containing PR QTLs (*Rphq2, Rphq3, Rphq11, Rphq16*), non-host QTL (*Rnhq*-V and *Rnhq*-L) and susceptible line, SusPtrit. Thus, higher proportions of EA were observed on NILs containing *Rphq2* QTL except in Vada-*rphq3*, for all inappropriate rust species. The occurrence of large proportions of early abortion (EA) combined with host cell necrosis has been reported as a typical feature of nonhost reaction. On non-host plant, infection units are arrested between formation of haustorial mother cell (HMC) and first haustoria often accompanied with limited cell collapse. In nonhost resistance analysis of *Arabidopsis* to *Phytophthora infestans* infection, penetrated epidermal cells with rapid hypersensitive response was observed, although no symptoms could be detected (Huitema et al.,

Table 5. Correlation coefficients (*r*) among infection parameters measured from four rust fungi.

Parameter	RLP *Ph*	RIF*Pt*	EA*Pt*	RIF*Phs*	EA*Phs*	RIF*Phm*	EA*Phm*
RLP *P. hordei* isolate 1.2.1	1.00						
RIF *P. triticina*'Flamingo'	-0.81	1.00					
EA *P. triticina*'Flamingo'	0.85	-0.84	1.00				
RIF *P. hordei-secalini*	-0.87	0.93	-0.86	1.00			
EA *P. hordei-secalini*	0.94	-0.91	0.90	-0.93	1.00		
RIF *P. hordei-murini*	-0.92	0.94	-0.86	0.98	-0.92	1.00	
EA *P. hordei-murini*	0.81	-0.79	0.87	-0.80	0.91	-0.76	1.00

RLP = relative latency period; RIF = relative infection frequency; EA = percentage of early abortion.

2003). Therefore, HR that cannot be detected on visual symptoms may be observed at the single cell level in Type I NHR, which is probably related to low infection probability and an extremely rapid HR on non-host plants.

The established colonies were classified according to their phase of development into two categories, namely, those with sporogenic tissues (WST) and without sporogenic tissues (WOST). There is a significant variation between QTL-NILs and reference line, in proportion of established infection units with and without sporogenic tissues at seven days after inoculation (Data not shown).

Significantly larger proportions of established colonies with sporogenic tissues were observed in SusPtrit as compared to SusPtrit NILs with PR and non-host QTLs. Microscopical observations made in this study on the mechanism of non-host resistance confirm that for a large part of the resistance is pre-haustorial. Large proportions of colonies were arrested before HMC formation (Table 4). From all tested NILs, lower percentages of established infection units (*P. triticina* 49.0%, *Phm* 29.5% and *Phs* 23.6%) were observed in NIL with *Rphq2*.

In NILs with *Rnhq* (Su-*Rnhq-V* and Su-*Rnhq-L*), the proportions of established colonies with sporogenic tissues were lower than in SusPtrit infected with all three inappropriate rust species, while the percentage is higher than that of SusPtrit for established colonies without sporogenic tissues (Data not shown).

Correlations between measured components of resistances for tested rusts

The association in prehaustorial resistance was quantified by calculating correlation coefficients (*r*) among the infection parameters measured for partial resistance and nonhost resistance. Higher negative correlation was observed between RLP of *P. hordei* isolate 1.2.1 and RIF of *P. triticina, Phm* and *Phs*, indicating that some of the genes for resistance to these rusts are either linked or have pleiotropic effects. The higher negative correlation was also observed between parameters measured from those tested inappropriate rust fungi (Table 5).

Furthermore, the higher positive correlation (*r* = 0.81 to 0.94) was also observed between RLP of *P. hordei* isolate 1.2.1 and EA of *P. triticina* isolate 'Flamingo', *Phm* and *Phs* (Table 5). Correlation between RIF of *P. triticina* isolate "Flamingo" and RIF of *Phm* and *Phs* was also higher. Higher positive association was observed between different parameters measured (RLP, RIF, and EA), indicating that there is a possible association between partial resistance quantitative trait loci and nonhost resistance quantitative trait loci. As stated in Qi et al. (1998, 1999), Niks et al. (2000) and Jafary et al. (2006), this significant association between parameters for PR to *P. hordei* and the loci for resistance to heterologous rusts revealed that these two traits are associated with each other.

Conclusion

The result of current study illustrated that PR QTLs, *Rphq2, Rphq3, Rphq11 and Rphq16* and the non-host resistances QTL, Rnhq, in barley, have effects on both partial resistance towards homologous leaf rust and non-host resistance towards heterologous leaf rusts. Furthermore, higher positive correlation was observed between parameter of relative latent period in seedling stage and proportion of early abortion at infection sites, which indicated that there is a possible association between partial resistance QTLs and non-host resistance QTLs.

Conflict of Interests

The authors have not declared any conflict of interests.

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