Mapping QTLs and major resistance genes to clubroot (*Plasmodiophora brassicae*) in *Brassica napus*

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ABSTRACT

The presence of two types of resistance to *Plasmodiophora brassicae* has been recognized in *Brassica napus*. High level of resistance to *P. brassicae* is isolate-specific and controlled by major genes. And according to the isolate, a partial resistance under polygenic control has been detected in rapeseed. The relationships between genetic components responsible for both types of resistance were investigated using a set of 110 doubled haploid lines from the cross Darmor-*bzh* x Yudal and 92 doubled haploid lines from the cross Stellar x Drakkar. Segregating progenies were tested against four different single spore isolates of *P. brassicae*. Three major specific genes, named *Pb-Bn1, Pb-Bn2 and Pb-Bn3,* efficient against 3 different isolates were mapped on 3 linkage groups of the consensus rapeseed genetic map. QTL analysis identified eight genomic regions implied in the resistance. Both qualitative and quantitative components exhibited an isolate specificity. In two chromosomic regions, colocalizations between two of the major specific-genes and QTLs implied in resistance to other isolates were found. This result suggests that a major-gene conditioning a high level of resistance may have a weaker effect that contributes, in association with other QTLs, to the expression of a partial resistance to other isolates.

Key words: rapeseed – mapping – qualitative resistance – quantitative resistance

INTRODUCTION

Clubroot of crucifers, caused by the obligate biotroph *Plasmodiophora brassicae* W., is a world damaging disease of rapeseed (*Brassica napus* var. *oleifera*). The development of resistant varieties is the most efficient option for protecting crops against *P. brassicae*. At the intraspecific level, several sources of resistance have been found in cultivars of swede and in fodder rape. This resistance is described as qualitative, being mainly controlled by 3 to 5 isolate-specific major genes (Crute et al, 1983; Gustafsson and Fält, 1986). In rapeseed, information about either host-pathogen interaction or the genetic basis of the resistance remains however limited. Results from the screening of rapeseed lines with several single spore isolates (SSI) of *P. brassicae* indicated that, according to the isolate, the resistance expression can vary from high to intermediate levels in a same host genotype (Manzanares-Dauleux *et al*, 2000a). Quantitative Trait Loci (QTL) analysis showed that partial resistance harboured by Darmor-*bzh*, a winter dwarf line, to a *P. brassicae* SSI was under control of several genetic factors. One of these QTL was mapped at the same position that a major gene of Darmor-*bzh* controlling high level of resistance to a different isolate of *P. brassicae* (Manzanares-Dauleux *et al*, 2000b). In order to gain insight into the relations between loci associated with partial and high-level resistance in rapeseed, we report here on the detection and mapping of other genomic regions with major and minor effects on clubroot resistance.

MATERIALS AND METHODS

Two segregating populations were analysed: a set of 110 doubled haploid lines (DH) from the cross Darmor-*bzh* x Yudal and a set of 92 DH from the cross Stellar x Drakkar. Plant material, genetic maps established from the two populations and the consensus rapeseed genetic map are described in Lombard and Delourme (2001). Four SSI belonging to 3 different *P. brassicae* pathotypes (Manzanares-Dauleux *et al*, 2001) were used: K92-16 (pathotype P4), Pb137-522 (P7), Ms6 and eH (kindly provided by J. Siemens, Berlin) (P1). Inoculation methods, scoring symptoms and disease index (DI) were performed as described previously (Manzanares-Dauleux *et al*, 2000b). QTLs were detected, using Mapmaker/QTL, on the genetic maps

established from the two DH populations and then located on the consensus rapeseed genetic map.

RESULTS

The DH progeny Darmor-*bzh* x Yudal was tested against isolates Pb137-522, K92-16 and eH. Darmor-*bzh* showed a high level of resistance to Pb137-522 and eH isolates and a partial resistance to isolate K92-16. The DH progeny Stellar x Drakkar was tested against isolate Ms6; Stellar was partially resistant to this isolate. Yudal and Drakkar were highly susceptible to all isolates tested.

QTL analysis identified 8 genomic regions implied in resistance. These QTLs fell on seven of the 19 rapeseed chromosomes (Fig 1). The number and weights of QTLs detected by isolate and the LOD scores are summarised in Table 1. In a multiple QTL model and according to the isolate, these loci accounted for 35.1% to 98.3 % of the phenotypic variation. For all the additive QTLs detected in the interactions with Pb137-522 and K92-16 isolates, the alleles of "Darmor-*bzh*" were associated with resistance. In the interactions with eH and Ms6 isolates, the resistance alleles at one and four QTLs, respectively, were from the susceptible parent. Three QTLs identified in the interactions with isolates Pb137-522, eH and Ms6 had a strong effect on resistance explaining individually 98.1%, 84.2% and 66.1%, respectively, of the variability observed. These 3 major resistance loci were named Pb-*Bn*1, Pb-*Bn*2 and Pb-*Bn*3. In two genomic regions, colocalizations between Pb-*Bn*1 and Pb-*Bn*2 and QTLs implied in resistance to other isolates were found.

Table 1. Clubroot resistance QTLs detected by interval mapping in the two DH populations for the DI resistance criteria with four *P. brassicae* isolates: position, LOD score, individual effect and contribution to the resistance variation.

Isolate	Linkage Group	Length (cM)	Position (cM)	LOD	Weight	R^2p (%)	R^2t (%)
DH progeny 'Darmor-*bzh'* x 'Yudal'							
522	LG4	8.3	5.3	91.0	90.9	98.1	
	LG2	11.8	0.0	3.85	35.4	15.0	
				91.3			98.3
16	LG4	8.3	2.3	4.8	12.3	19.4	
	LG15	3.7	0.0	3.4	10.4	13.4	
				9.8			35.1
eH	LG15	5.9	2.9	42.4	61.2	84.2	
	LG9	3.4	0.0	2.7	-20.9	10.6	
	*LG2	10.5	2.0	+3.8	-10.4		
	*LG717	1.2	0.0	+2.8	-8.7		
	*LG12	1.5	0.0	+2.1	-7.8		
				53.4			90.2
DH progeny 'Stellar' x 'Drakkar'							
Ms6	LG19	7.6	2.9	18.0	44.6	66.1	
	LG4	8.1	4.4	2.8	21.1	15.9	
	*LG717	8.2	4.1	+1.6	-16.0	9.1	
				25.8			80.8

R^2p is the percentage of variation explained by each QTL

R^2t is the percentage of variation explained by all the additive QTLs detected for one isolate

* QTLs detected with a multiple QTL model; the increase in the total LOD with the new QTL is indicated with the sign '+'.

DISCUSSION

The results of this study indicate that the resistance response of two segregating rapeseed populations to different *P. brassicae* isolates showed both qualitative and quantitative components, all of these exhibiting a isolate-specificity. We have identified and located on the linkage groups LG4, LG15 and LG19 three major genetic factors controlling resistance to three different single spore isolates. These genes may be identical to one of the genes previously identified in the *B. napus* ECD hosts (Gustafsson and Fält 1986); it is not possible however to confirm with the available published data. These loci were also identified by QTL analysis using quantitative evaluation of response reactions and they mapped at the same position. Two of the

major locus, *Pb-Bn1* and *Pb-Bn2,* maintained a residual effect (acted as a QTL) against other isolates. This result suggests that a major gene conditioning a high level of resistance may have a weaker effect that contributes, in association with other QTLs, to the expression of a partial resistance to other isolates.

Work is in progress to study the efficiency of different genetic combinations, including the major and minor resistance genes, on resistance to different *P. brassicae* populations. If major resistance genes provide some residual effect, pyramiding these genes, and combining with quantitative resistance and other management tactics, may provide an additional component for reducing the risk of major losses due to clubroot of crucifers.

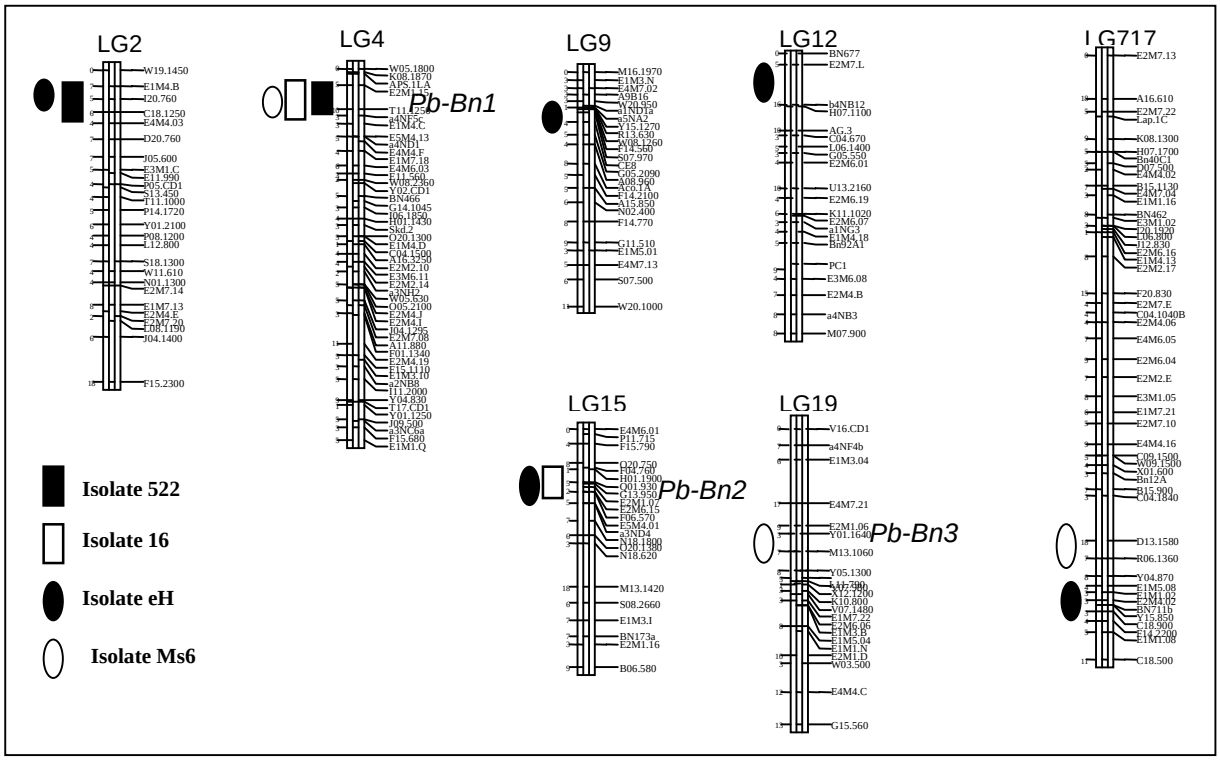

Fig. 1. Locations on the consensus rapeseed genetic map of the major resistance genes and the additive QTLs contributing to clubroot resistance identified in the cross Darmor-*bzh*xYudal and in the cross StellarxDrakkar. The QTL length is in proportion to the confidence interval. Different symbols are used for the four isolates analysed.

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