Characterization of Clubroot Resistance in Recent Winter Oilseed Rape Material

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ABSTRACT

Resistance breeding is a major method to control clubroot disease. Recently, new oilseed type cultivars of *B. napus* have been released carrying clubroot resistance, eg cv. Mendel. A threat to these new resistances will be the adaptability of the pathogen causing clubroot, *Plasmodiophora brassicae*, which is showing great variability. Different resistance sources can be used for *B. napus* breeding: Dominant, strong but race-specific genes from *B. rapa*, recessive but mainly unspecific genes from *B. oleracea* or a number of dominant, but weak and race-specific genes present in *B. napus* fodder types. Our work focuses on the characterization of clubroot resistance in recent oilseed rape using *P. brassicae* isolates that have been selected for representing different pathotypes. For this purpose, we have established a new set of tester lines carrying race-specific, dominant resistance genes originating from *B. rapa*, which have been introduced into *B. napus*. First results indicate race-specific resistance genes in the new cultivars.

INTRODUCTION

Clubroot disease incidences in oilseed rape are increasing due to the wide spread of the crop. As agricultural control means are poorly efficient, resistance breeding is used to ensure yield and presence of the crop in endangered areas. However, the great variability of the pathogen is a major challenge to these efforts. To characterize race-differentiation in *P.brassicae* sets of differential hosts have been introduced, in particular the ECD-Set (Buczacki et al. 1975). This set comprises 5 hosts differing for clubroot resistance for each of the *Brassica* species *oleracea*, *rapa* and *napus*. Other sets have been used that are usually composed of less hosts, in particular from *B.rapa* or *B.napus*, where resistance appears to be in general race-specific. A number of problems have to be faced when working with these tester sets:

- *B.rapa* and *B.oleracea* are outbreeding species; their differential hosts are not lines, but heterogenic
- *B.napus* differentials are usually more homogenous, but many isolates overcome their resistance
- At least the ECD-hosts are complex resistance genotypes. Compatible races always need to have more than one virulence factor. Therefore, the presence of races carrying eg. only 1 virulence factor for the *B.rapa* resistance will be overlooked.
- Defined pathotypes to select the correct differential genotypes according to the postulate are not available.

Our work is focused on the characterization of clubroot resistance in newly developed *B.napus* lines. Resistance in this material originates from resynthesized *B.napus* forms, having clubroot resistance from *B.rapa* and/or *B.oleracea*. In previous work a *B.napus* backcross population segregating for the dominant resistance genes that originate from the *B.rapa* differential host ECD-04 had been characterized (Diederichsen and Sacristan 1996, Diederichsen et al. 1996). A selection from this population is now being used to determine virulence properties of *P.brassicae* isolates and the identity of resistance loci.

MATERIALS AND METHODS

Plant material and *P.brassicae* isolates were as described by Diederichsen et al. 1996. Seeds of the *B.napus* cvs. Sparta and Mendel were kindly supplied by the respective breeders. All greenhouse experiments and data handling were as described by Diederichsen et al. 1996. Maintenance of the selected backcross individuals has been made by vegetative *in vitro* propagation.

RESULTS AND DISCUSSION

When testing the backcross population with the isolates e, k, h, or 1, different segregation ratios could be observed depending on the isolate (Table 1). Isolate 1 (a single spore derived isolate, SSI) never caused any symptoms on the *B.rapa* hosts ECD-01, 02, 03 or 04 and is assumed to carry no specific virulence. This is confirmed by the segregation ratio, which indicates 3 dominant resistance genes, as it has to be expected according to the postulated resistance genes for the resistance donor ECD-04.

The same plants segregated in a 1:1 ratio when tested with the 2 field isolates e or k, resp., indicating 1 efficient gene towards both. A number of BC individuals showed differential interactions with both isolates, while another group of plants was attacked by both isolates. This indicates that the isolates k and e carry 2 virulence factors each, one of these factors is shared by both of them.

The SSI h attacked slightly more than 25% of the BC plants, indicating 2 efficient genes. All plants that were susceptible to h were also susceptible to isolate e, whereas susceptible plants with isolate h were usually resistant to k, except for those plants that were also susceptible to isolate 1. Thus, isolate h carries 1 virulence factor which is shared with isolate e, but not with k. As isolate k regularly attacks the ECD-host 01 completely, it can be assumed to have vir_{b+c}. Isolate h should carry 1 different factor, which is vir_a. Isolate e should then be vir_{a+b} or - less likely according to reactions with ECD-hosts - vir_{a+c}.

Table 1. Segregation analysis of a *B.napus* backcross population after multiple resistance testing of cuttings against 4 different *P.brassicae* isolates

P.brassicae Isolate	Resistant individuals	Susceptible individuals	Expected segregation	Number of efficient genes	X^2
e	84	89	1 : 1	1	0.14
h	121	55	3 : 1	2	3.67
k	101	76	1 : 1	1	3.53
1	153	31	7 : 1	3	3.18

Table 2 summarizes the composition of our BC-population based tester set. In total 31 BC individuals have been selected for this purpose.

Table 2. *B.napus* differentials and used *P.brassicae* isolates – postulated virulence and resistance genotypes

Resistance genotype*	BC individuals	Virulent *P.brassicae* isolates	Postulated virulence factors
A B C	30, 42, 163, 195	(none)	-
A - C	24, 74, 135, 171	(none)	-
- B C	5, 49, 70, 138	k	vir_{b+c}
- - C	15, 39, 54, 102	k	vir_{b+c}
A B -	11, 26, 40, 187	e	vir_{a+b}
A - -	1, 55, 82, 175	h, e	vir_a
- B -	62, 80, 140	k, e	vir_b
- - -	12, 165, 166, 182	1, h, k, e	(none)

* Designation of resistance genes according to Wit 1964.

Some more recent *B.napus* cultivars (Mendel, Sparta) were tested with the mentioned isolates. Both cultivars exhibited no complete resistance (Table 3). When applying a cut-off point of DI=25 to distinguish resistant vs. susceptible reactions, Sparta was susceptible to all 3 isolates, although a quantitative reduction of susceptibility was still present compared to fully susceptible hosts (eg ECD-05 or ECD-07, DI=100). Cv. Mendel, a hybrid cultivar, was resistant towards the isolates 1 and e, the very small number of fully diseased plants amongst a population of plants without symptoms is likely to be explained by the hybridity which is between 90 to 100%. The

resistance in cv. Mendel has to be regarded as race-specific, as the isolate k showed a compatible reaction. Therefore, it should be assumed that Mendel's resistance is not based on the gene A, but most likely on both B and C. However, also cv. Mendel did not show a completely susceptible reaction with k, indicating additional resistance genes.

Table 3. Disease indices (DI) of cvs. Mendel and Sparta with 3 *P.brassicae* isolates

cv. Sparta		cv. Mendel	
Isolate	DI	Isolate	DI
a*	84	1	7
e	63	e	12
k	53	k	45

*= Field isolate; SSI h was isolated from this population

Using the tester set it should be possible to designate the resistance genes in these 2 cultivars more precisely and also of other cultivars, such as cv. Tosca, which is now ongoing. The tester set will also help us to identify most informative *P.brassicae* isolates. These are prerequisites for the identification of most suitable additional resistance genes that can serve for gene pyramiding in breeding.

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