Dissimilarity of Barley Powdery Mildew Resistances Lomerit and Heils Hanna

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Abstract: The resistance Heils Hanna (HH) was postulated in several tens of 471 previously tested winter barley cultivars. In this paper, new tests on 29 of these cultivars are reported. Thirty-two reference isolates of *Blumeria graminis* f.sp. *hordei* held in the pathogen genebank at the Agricultural Research Institute in Kromeriz, Ltd. including a Japanese isolate and five Israeli isolates were used for response tests. However, the resistance HH conferred by the gene *Mla8* and herein characterised by reaction type 0 to an old Japanese isolate known as Race I was now postulated only in four cultivars. In the other 25 cultivars another resistance, characterised by reaction type 0 to Race I and also to two Israeli isolates, was detected. In addition to the two mentioned resistances, eight known (Bw, Dr2, Ha, IM9, Ln, Lv, Ra and Sp) resistances were found in the set examined. Lomerit was the only registered cultivar tested here in which the newly detected resistance was present alone, therefore, it is recommended that this resistance be designated Lo.

Keywords: *Blumeria graminis* f.sp. *hordei*; *Hordeum vulgare*; pathogen isolates; reaction types; resistance gene postulation; pedigree analysis

Barley (*Hordeum vulgare* L.) is an important crop in the Czech Republic and powdery mildew caused by the ascomycete fungus *Blumeria graminis* (DC.) E. O. Speer, f.sp. *hordei* emend. É. J. Marchal (anamorph *Oidium monilioides* Link) (= *Bgh*) is a predominating disease of this crop (DREISEITL 2011a).

In 2004, the cultivar Tadmor, which is highly susceptible to the powdery mildew pathogen and which was selected from a Syrian landrace Arabi Aswad (Ceccarelli, personal communication), was discussed for use in experiments with *Bgh*. In subsequent tests with more than 50 isolates, Tadmor exhibited susceptibility (reaction type 4 = RT4) to all isolates used, except the isolate known as Race I, to which it exhibited a phenotype of full resistance (RT0). Such a resistance spectrum (RS) was identical to RS of cultivars possessing the resistance HH designated according to an old cultivar

Heils Hanna. This resistance is controlled by the gene *Mla8* and can be detected by a postulation method using only some non-European isolates because no avirulent isolate has been found in the European pathogen population. Race I originates from Japan and it is one of a few known isolates that are avirulent to the gene *Mla8* (HIURA & HETA 1955). Since the tests of Tadmor produced identical RS as Heils Hanna, it was concluded that Tadmor also possesses the resistance HH.

In 2009, resistance of a part of a huge collection of wild barley (*Hordeum vulgare* subsp. *spontaneum* (C. Koch) Thell.) from the International Centre for Agricultural Research in the Dry Areas (ICARDA) genebank was tested. Tadmor was added to tests of these wild barley accessions and Israeli isolates collected from naturally growing wild barley (Dreseitle et al. 2006) were also used. It was found that cv. Tadmor, in contrast to cultivars with the

resistance HH, was characterised by RT0 not only to Race I, but also to two of five Israeli isolates used, which means that it possesses resistance other than HH. Tadmor comes from the Fertile Crescent region where a large number of barley powdery mildew resistances (FISCHBECK *et al.* 1976; JAHOOR & FISCHBECK 1987, 1993; DREISEITL & BOCKELMAN 2003) have their origin. Therefore,

the new resistance detected in this cultivar was not very surprising.

Resistance tests of the above-mentioned wild barley collection continued in 2010. For technical reasons, a set of 20 breeding lines of winter barley had to be exceptionally included in those tests and thus, these lines were tested also with isolates that had been used only for testing wild barley by

Table 1. Twenty-nine winter barley cultivars and their postulated resistances to the barley powdery mildew pathogen

Cultivar	No. rows	Pedigree	Resistance spectrum	Resistance code	
Alinghi (LP 6-225)	6	(LP 6-460 × 1665-24) × Lomerit 9		Lo IM9	
BE 601503	6	Lomerit × Carola 1		Lo	
Borwina (HVW 823/75)	6	(Valja × Vogesanger Gold) × Hohenthurm 7246 2		Lo Bw	
Florian (KM 999/04)	2	HVW 10803 × Monaco 15		HH Ln	
Kamil (KM-HM 2110)	6	Borwina × HVW 1044 2		Lo Bw	
Kromir (KM 252)	6	Selection from the Afghan barley K-20667 6		Lo Ra Dr2 Ha	
Kromoz (KM 908)	6	Erfa × HVW 759	5	Lo Ra Dr2	
Laurena (NORD 96515/26)	6	NORD 92544/1 × Carola	8	Lo Lv	
Laverda (SUR 01/3128)	6	(Ludmila × GW 1836) × Merlot	16	HH Lv	
Lomerit (LP 6-758)	6	(Askanova × Girte) × (Oceane × 1332-29)	1	Lo	
LP 6-627	6	HVW 2935 × Sorna	7	Lo Ra Ln	
LP 6-728	6	(Ikone \times Lomerit) \times Fridericus	11	Lo Sp	
LP 6-764	6	LP 6-355 \times Fridericus	1	Lo	
Lunet (SG-L 16)	6	$(Aschid \times Dea) \times Senta$	2	Lo Bw	
Luran (SG-L 725)	6	Okal × Lunet	3	Lo Bw Ra	
Luxor (SG-L 72)	6	Okal × Lunet	4	Lo Bw Ra Ha	
NORD 05109/19	6	NORD 20629/13 × Highlight	8	Lo Lv	
Okal (SG-L 27)	6	W 77 × HVW 860	4	Lo Bw Ra Ha	
Scarpia (BE 141601)	6	Carola \times (241685 \times Noveta)	10	Lo IM9 Ra	
SG-L 00/015/B1/A	6	Carola × Okal	13	Lo Sp IM9 Ha	
SG-L 00/015/B1/C	6	Carola × Okal	12	Lo Sp IM9	
SG-L 00/015/B9/A/09	6	Carola × Okal	11	Lo Sp	
SG-L 00/051/09	6	Hiro × Luran	4	Lo Bw Ra Ha	
SG-L 00/132/A/05	6	Seco-D 133-8b × SG-L 111		HH Ra Ha	
SG-L 99/079/09	6	Catania × Luran	5	Lo Ra Dr2	
SG-L 3423/D/07/B	6	Seco-D 133-3c × Luran 3		Lo Bw Ra	
Souleyka (NORD 03025/3)	6	Pelican × Laverda	16	HH Lv	
SZD 4127	6	(W781-1783 × Tundra) × Amarena	11	Lo Sp	
Wendy (NORD 02611/33	6	Palmyra × Laverda	8	Lo Lv	

then. Owing to that, it was found by chance but with a big surprise that 16 lines did not carry the expected resistance HH, which is often present in older spring barley cultivars, but other resistance which is identical to that in Tadmor. Therefore, the objectives of this research were to use a part of the current collection of the pathogen including five Israeli isolates to (*i*) test a set of selected winter barley cultivars which should possess the resistance HH or the resistance recently found in Tadmor, (*ii*) postulate resistances of these cultivars, and (*iii*) compare the obtained results with the pedigrees of the tested cultivars.

MATERIAL AND METHODS

Twenty-nine cultivars in which the resistance HH was previously postulated (Dreiseitl 2007) were tested (Table 1). Thirty-two reference isolates of *Bgh* held in the pathogen genebank at the

Agricultural Research Institute Kromeriz, Ltd. were used for response tests. Resistances were postulated by comparing the responses of the tested cultivars with cultivars possessing genes from known resistance sources. Further details of the method used including the testing procedure and evaluation can be found in DREISEITL (2011b).

RESULTS

Twenty-nine cultivars tested with 32 mildew isolates resulted in 16 RSs (Table 1). To characterise these RSs, 10 isolates including Race I avirulent to all cultivars, and an Israeli isolate J-462 avirulent to most of the tested cultivars were selected (Table 2). The resistance HH, characterised by avirulence (RT0) to one isolate only (Race I), was detected in four cultivars, however in all cases in combination with other resistances. Therefore, RS0 characterising the alone resistance HH was

Table 2. Sixteen resistance spectra produced by 29 winter barley cultivars inoculated with 10 isolates of *Blumeria* graminis f.sp. hordei

Resistanc	e	Isolate of Blumeria graminis f.sp. hordei										
Spectrum	code	Race I	J-462	A-G/05	EmA30	21/99	I-248/99	J-20/04	I-158/09	M-246/09	O-283/09	
$\overline{0^1}$	НН	0	4	4	4	4	4	4	4	4	4	
1	Lo	0	0	4	4	4	4	4	4	4	4	
2	Lo Bw	0	0	2-3	4	4	4	4	4	4	2-3	
3	Lo Bw Ra	0	0	2-3	0-1	4	4	4	4	4	2-3	
4	Lo Bw Ra Ha	0	0	2-3	0-1	4	4	4	4	4	2	
5	Lo Ra Dr2	0	0	2	0-1	4	4	4	4	4	4	
6	Lo Ra Dr2 Ha	0	0	2	0-1	4	4	4	4	4	2	
7	Lo Ra Ln	0	0	4	0-1	4	1-2	1-2	1-2	1-2	1-2	
8	Lo Lv	0	0	1-2	1-2	1-2	1-2	1-2	4	1-2	1-2	
9	Lo IM9	0	0	1-2	1-2	1-2	1-2	1-2	4	4	4	
10	Lo IM9 Ra	0	0	1-2	0-1	1-2	1-2	1-2	4	4	4	
11	Lo Sp	0	0	0	4	4	4	0	4	4	4	
12	Lo Sp IM9	0	0	0	1-2	1-2	1-2	0	4	4	4	
13	Lo Sp IM9 Ha	. 0	0	0	1-2	1-2	1-2	0	4	4	2	
14	HH Ra Ha	0	4	2-3	0-1	4	4	4	4	4	2-3	
15	HH Ln	0	4	4	1-2	4	1-2	1-2	1-2	1-2	1-2	
16	HH Lv	0	1-2	1-2	1-2	1-2	1-2	1-2	4	1-2	1-2	

¹Resistance spectrum produced by the standard spring barley cultivar Pallas

included in Table 2. In 25 cultivars the resistance characterised by RT0 to two isolates (Race I and J-462) was found and in three of these cultivars (Lomerit, BE 601503 and LP 6-764) this resistance was present alone. Besides the two mentioned resistances, six well-known (Bw, Dr2, Ha, IM9, Ra, Sp) and two newly described resistances (Ln and Lv) (Dreiseitl 2011c, d) were found in the set examined. Thus, in the set of cultivars a total of 10 resistances were detected that were present in individual cultivars in 15 various combinations (Table 1).

DISCUSSION

In resistance tests conducted here the set of 29 cultivars exhibited 16 RSs. It can be deduced from 13 observed RSs that 25 of the tested cultivars share a so far unknown resistance, present in three cultivars in the absence of other resistance genes (RS1). Lomerit was the only registered cultivar in which no other resistance was found besides the resistance effective against Race I and isolates J-462 and S-016 (the latter one is not shown in Table 2).

Pedigrees of cultivars that produced 13 abovementioned RSs suggest that the corresponding resistance must be present in many old cultivars, but it has never been detected (Brown & Jør-GENSEN 1991) despite its remarkable phenotype expressed by RT0. Thus, it can be supposed that the effectiveness of this resistance to the European pathogen population, which is determined by the frequency of avirulent isolates, must be extremely low not only in the Czech Republic where 0% of the avirulence has been found (DREISEITL 2008, 2011e) but also in the rest of Europe. This is apparently an explanation why this "very old" resistance was detected only on the basis of the tests presented herein, and only due to using non-European isolates. Out of the registered cultivars tested here, this resistance is present alone only in Lomerit, bred by Lochow-Petkus GmbH, Germany, and registered there in 2001. Therefore, according to agreed guidelines (Boesen et al. 1996), the resistance characterised by RS1 was designated as Lo.

The resistance Lo can be theoretically based on two or more genes with the same phenotype (RT0), it means for instance on the gene *Mla8*, which confers the resistance to Race I, and on the second gene which would control the resistance to isolates J-462 and S-016. It would be similar,

for example, to hypothetical RS at accumulating the resistances Ln and Lv detected here in one cultivar. Both these resistances are characterised by an identical phenotype (RT1-2), and if they were present together in one cultivar, the corresponding RS would not indicate that it is determined not by one gene but by two genes with an identical phenotype. However, the resistance Lo was detected in 25 cultivars that were bred by crossing and selecting, and in none of them were for example RT4 after inoculation with Race I and RT0 after inoculation with isolates J-462 and S-016 found, which would be expressed in the case of more genes conferring this resistance. Thus, the resistance Lo is likely to be determined by one gene, though the presence of two or more genes in strong coupling cannot be excluded.

As mentioned above, the resistance Lo, like the resistance HH, against the European pathogen population is assumed to be very weak and therefore the value of both resistances is negligible from the aspect of their contribution to the resistance of grown cultivars. Nevertheless, information about the presence of the resistance Lo or HH can be of considerable importance in genetic studies. A project aiming at locating newly identified resistance genes in the barley genome has recently been discussed. A large number of resistance genes to the powdery mildew pathogen in barley that have been localized so far are at the Mla locus (Jør-GENSEN 1994; DREISEITL et al. 2007; ŘEPKOVÁ et al. 2006, 2009a, b; Řepková & Dreiseitl 2010; Seeholzer et al. 2010; Teturová et al. 2010). Therefore, it is important to define the relation of new genes just to this locus. From this point of view, Lomerit with its resistance HH conferred by the gene Mla8 postulated at that time showed to be a very good candidate for such study.

The method used here is classical (Brückner 1964) and still highly effective for postulation of resistance genes against plant disease pathogens (Dreiseitl & Steffenson 2000; Czembor 2002; Kolmer 2003; Lillemo et al. 2010; Zhang et al. 2010; Goyeau & Lannou 2011; Silvar et al. 2011). As any method, it has also some constraints that can be demonstrated in full using the example of Lomerit. This cultivar was first tested in 1998, when it was considered to possess no specific resistance to the powdery mildew pathogen. However, later in 1998, Race I was brought from Risø National Laboratory, Denmark, where it had been used since 1980 (Jørgensen & Jensen 1983). In subsequent

tests, Lomerit exhibited the resistance against this isolate, and because its resistance phenotype was identical to the HH resistance phenotype (RT0), the original conclusion about its resistance was corrected (it contains the resistance HH). From 1999 through 2009, 94 different isolates were used for testing, while there was no difference in RS between the resistance HH and Lo. So, it was a unique case when both resistances were characterised not only by the identical RS, but also by the identical phenotype (RT). The dissimilarity of these resistances was first determined when two Israeli isolates were employed, 12 years after the first tests of Lomerit. On the one hand, it demonstrates certain constraints of the method, on the other hand, it also documents the importance of studying the global pathogen population as well as alternation of isolates at postulating specificresistance genes.

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