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# RESULTS OF MONITORING OF THE POPULATION OF *Blumeria graminis* f.sp. *hordei* IN LATVIA IN 2009–2010

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*In 2009–2010, random samples of the causal agent of barley powdery mildew were collected in Daugavpils (south-eastern Latvia, Latgale region), Stende (north-western Latvia, Kurzeme region) and Priekuļi (north-eastern Latvia, Vidzeme region). Virulence frequency, complexity and pathotypes were calculated in the pathogen populations. Significant differences of virulence detected by the genes* Va1*,* Va3 *and* Va13 *occurred among samples of the pathogen population collected in different parts of Latvia. Nei index, Müller's index, Kosman index, Shannon index and Simpson index showed considerably higher diversity in Daugavpils and Stende during 2009–2010. In Daugavpils, the population of* Blumeria graminis *f.sp.* hordei *was particularly characterised by high diversity.*

**Key words:** Blumeria graminis *f.sp.* hordei*, barley, virulence, resistance, complexity, pathotype.*

## INTRODUCTION

Barley (*Hordeum vulgare* L.) has played a pivotal role in Old World agriculture since its domestication about 10 000 years ago (Piffanelli *et al*., 2004) and is the fourth most important cereal crop in the world, after wheat, maize and rice (Czembor, 2001; Anonymous, 2009b). Barley is also one of the most important cereals in Latvia. In 2008, the area of sown spring barley was 28% (147 000 ha) of the total cereal area in Latvia (Anonymous, 2009a). In 2009, of the area of barley decreased to 104 700 ha (Anonymous, 2009b).

The causal agent of barley powdery mildew, *Blumeria graminis* DC. f.sp. *hordei* Ėm. Marchal, is a windborne, biotrophic, fungal pathogen of cultivated and wild barley. It is particularly prevalent under cool conditions when the maximum daily temperature does not exceed 25 °C (Dreiseitl *et al*., 2006). Powdery mildew is one of the most destructive foliar diseases of barley in regions such as Europe (Czembor, 2001). In Latvia, where the climate is moderate, the pathogen develops in two stages: in the vegetation period, numerous conidia are formed, which ensure propagation and dispersal of the fungi; in autumn, cleistothecia with spores are formed on senescing parts of plants. The spores mature in the next spring and cause host infection, and powdery mildew epidemics.

Presently, powdery mildew-barley genetic interactions are considered as one of the host-pathogen systems that is well genetically characterised (McDermott *et al*., 1994). Harold Henry Flor proposed a gene-for-gene model for the genetic interaction between plant and pathogen, in which a dominant gene of the host interacts with a corresponding dominant virulence gene of the pathogen. Interactions between the two corresponding genes, the host resistance gene and the pathogen avirulence gene, induce a hypersensitive response, in this way providing resistance. Tools of molecular biology led to a hypothesis that a ligand from the pathogen interacts with a corresponding plant receptor, which triggers a defence response (Richter and Ronald, 2000).

More than 100 mildew resistance genes have been identified in barley cultivars, landraces, and wild or related *Hordeum* species. *Mlg* was the first gene introduced on a large scale in the 20$^{th}$ century (Brown and Jørgensen, 1991; Czembor, 2001). Many mildew resistance alleles have been identified on chromosome 4H and chromosome 1H of the barley genome. The *Mla* locus for barley resistance to powdery mildew is located on chromosome 1H (Schüller *et al*., 1992). Most genes in the *Mla* locus have lost their effectiveness during the recent few years in different regions of Europe, due to the high level of pathogenic variability of powdery mildew (Müller *et al*., 1996; Caffier *et al*., 1999; Limpert *et al*., 1999; Hovmøller *et al*., 2000; Dreiseitl, 2004a; 2004b; 2008; Kokina and Rashal, 2005a; 2007; 2012). Most of the powdery mildew resistance genes, including *Mla1, Mla3, Mla6, Mla7, Mla9, Mla12, Mla13, Mlat, Mlk, Mlg, Mlh* and *Mlra,* are commercially derived from the Mediterranean region (Czembor, 2001). Barley plants carrying recessive alleles of the *Mlo* locus are resis-

tant against all known isolates of powdery mildew fungus (Jørgensen, 1992; Piffanelli *et al*., 2004).

Host-pathogen evolution has has been a driver of biological diversity (Laine, 2006). The evolution of virulence involves the emergence of pathogens, host switch and host range expansion, and overcoming of host resistance, which may compromise the success of control strategies for diseases. Virulence evolution has an important role of pathogens in ecosystem composition and dynamics (Sacristán and Garcia-Arenal, 2008) and is considered as a force influencing diversity, including resistance, in plants (Brown, 2003). Genetic polymorphism of resistance in the host reduces the probability that a parasite infects the host (Thrall and Burdon, 2003).

There has been increased interest in the structure of *Blumeria graminis* f.sp. hordei local populations, because understanding the evolution of the pathogen requires recognition of variation within and between populations (Müller *et al*., 1996). Moreover, individual genetic structures for local populations of typical air-borne pathogens like the causal agent of barley powdery mildew can differ greatly from the average structure of aerial population (Dreiseitl and Wang, 2007). The evolutionary potential of the pathogen population influences the durability of disease resistance.

The overall goal of this study was to characterise the *B. graminis* f.sp. *hordei* population in Latvia. The specific objectives were: 1) to characterise the population in Latvia using standard European differentials supplemented by barley lines with "new" resistance genes; 2) to compare virulence frequencies, complexity and pathotypes, defined as combinations of virulences, on a set of differential hosts carrying different resistance genes, in samples collected in different parts of Latvia; 3) to evaluate the diversity and evolution potential of the population for further use in the barley breeding programme for resistance.

## MATERIAL AND METHODS

**Pathogen population and sampling**. In 2009–2010, random samples of *Blumeria graminis* f.sp. *hordei* isolates were obtained in three regions of Latvia. Among the sampling sites were commercial barley fields near Daugavpils (south-eastern part of Latvia, Latgale region), where samples were collected both in conidia and cleistothecia stages from unknown host genotypes in 2009–2010. Samples in conidia were collected before peak powdery mildew development. In the second sampling site, located near Talsi (north-western part of Latvia, Kurzeme region), samples of the pathogen were collected in breeding fields of the State Stende Cereal Breeding Institute, only in the cleistothecia stage in both years of investigation, from five barley varieties ('Annabell' and 'Druvis' in 2009 and 'Dzintara', 'Agra', 'Austris' and 'Druvis' in 2010). In the third sampling site, located in Priekuļi (north-eastern part of Latvia, Vidzeme region), pathogen samples were collected in breeding fields of the State Priekuļi Plant Breeding Institute in the cleistothecia stage from barley varieties 'Agra' and 'Rolfi'in 2009. The distance between Daugavpils and Stende is about 350 km, between Daugavpils and Priekuļi about 210 km, and between Stende and Priekuļi about 200 km. Dates and location of the sampling are presented in Table 1.

**Isolation and multiplication of single colonies**. For isolation and multiplication of single colonies from samples, both in conidia and cleistothecia, the first leaves of the universally susceptible barley variety 'Otra' were used. Plants were grown in a pot with soil at temperature 18–20 °C (12 h) under artificial light for 7–10 days.

Leaf segments of the host with single colonies of the pathogen that had developed from natural infections were placed in 100 mm plastic Petri dishes with 1% water agar and 0.004% benzimidazole and incubated for 20–24 h at 22–

Table 1

BARLEY POWDERY MILDEW SAMPLES COLLECTED IN LATVIA IN 2009–2010

| Location | Sampling year | Date of sampling | Barley genotype | Number of tested isolates | Life cycle stage of the pathogen (conidia or cleistothecia) | ID |
|---|---|---|---|---|---|---|
| Daugavpils | 2009 | June 28 | unknown | 40 | conidia | Da/09/con |
| | | July 29 | unknown | 40 | cleistothecia | Da/09/cl |
| | 2010 | June 28 | unknown | 40 | conidia | Da/10/con |
| | | August 5 | unknown | 40 | cleistothecia | Da/10/cl |
| Stende | 2009 | July 25 | 'Regatta' | 21 | cleistothecia | St/09/cl/1 |
| | | July 25 | 'Annabell' | 21 | cleistothecia | St/09/cl/2 |
| | | July 25 | 'Druvis' | 20 | cleistothecia | St/09/cl/3 |
| | | July 25 | 'Idumeja' | 20 | cleistothecia | St/09/cl/4 |
| | | July 25 | 'Merbin' | 21 | cleistothecia | St/09/cl/5 |
| | 2010 | August 5 | 'Dzintara' | 31 | cleistothecia | St/10/cl/1 |
| | | August 5 | 'Agra' | 23 | cleistothecia | St/10/cl/2 |
| | | August 5 | 'Austris' | 24 | cleistothecia | St/10/cl/3 |
| | | August 5 | 'Druvis' | 24 | cleistothecia | St/10/cl/4 |
| Priekuļi | 2009 | August 18 | 'Agra' | 30 | cleistothecia | Pr/09/cl/1 |
| | | August 14 | 'Rolfi' | 30 | cleistothecia | Pr/09/cl/2 |

24 °C. Well-developed monopustules were transferred onto 25 mm long healthy primary leaves of 'Otra', which were placed on 1% water agar in Petri dishes and incubated at 18–20 °C (12 h) under artificial light for ten days.

For isolation of ascospores from cleistothecia, the host leaf segments with well-developed cleistothecia were placed on wet filter paper on the lid of a Petri plate and cultivated at 18–20 °C for 3–4 days. Then, the lids with mature cleistithecia were placed in Petri plates with 'Otra' 20-mm leaf segments on 1% water agar with 0.004% benzimidazole. When the filter paper had dried out, cleistothecia contracted and „shot" ascospores, infecting the host leaves. Host leaves infected by ascospores were incubated at 18–20 °C for 3–4 days and used for isolation of single colonies.

**Inoculation of differentials**. For testing of single colonies, the set of used differentials (Kølster *et al*., 1986) comprised nine near-isogenic Pallas lines, barley line *SI1* and three barley varieties 'Steffi', 'Goldie' and 'Meltan', which contained different genes for resistance to powdery mildew. Monopustule isolates were also tested on *mlo 5* (near isogenic Pallas line *P22*). A list of differentials used for detection of virulence genes in the population of *Blumeria graminis* f.sp. *hordei* in Latvia in 2009–2010 is presented in Table 2.

About 50–70 seeds of each differential were sown in plastic pots with sandy soil and were grown under laboratory conditions at 18–20 °C and natural light 10–14 days until well-developed first leaves appeared. Leaf segments with length approximately 20 mm were cut and placed on water agar with 0.004% benzimidazole in Petri dishes (100 mm in diameter). The number of Petri dishes was identical with the number of tested isolates.

Inoculation of differentials was conducted according by microinoculation (Dreiseitl, 1998). Monopustule isolates were drawn into a micropipette and dispensed into a setting tower, under which differentials 1% water agar with 0.004% benzimidazole in a Petri plate were exposed. The plates with inoculated differentials were incubated in chambers at temperature 18–20 °C in light with a photoperiod of 12 h.

Table 2

DIFFERENTIALS USED FOR DETECTION OF VIRULENCE GENES IN *Blumeria graminis* f.sp. *hordei* SAMPLES COLLECTED IN LATVIA IN 2009–2010

| Differentials | Main resistance genes |
|---|---|
| *P01* | *Mla1* |
| *P02* | *Mla3* |
| *P03* | *Mla6* |
| *P04B* | *Mla7* |
| *P08B* | *Mla9* |
| *P10* | *Mla12* |
| *P11* | *Mla13* |
| *P17* | *Mlk* |
| *P23* | *MlLa* |
| *SI1* | *Ml(SI)* |
| 'Steffi' | *Ml(St1), Ml(St2)* |
| 'Goldie' | *Mla12, MlLa, U* |
| 'Meltan' | *Mla13, Ml(Im9), Ml(Hu4)* |

**Virulence determination**. The infection type of differentials was detected 7–8 days after inoculation, according to a 0–4 point scale (Torp *et al*., 1978). Isolates with reaction type 0–3 were classified as resistant. Reaction type 4 was considered virulent on the corresponding resistance genes. Virulence frequencies, complexity (virulence gene number per genotype) and combinations of virulence genes in isolates were detected by pooling individual isolates from corresponding samples. Each pathotype was designated by the set of virulence genes present in a particular isolate.

**Data analysis**. Virulence frequency, complexity and pathotypes were calculated with the programme RASA. For characterisation of the diversity within populations and the distance between populations, Kosman indices were used (Kosman and Leonard, 2007). All diversity parameters were computed using the KOIND package (Kosman, 2002), which are based on the bootstrap method. Statistical significance of differences between all calculated parameters, including Kosman diversity KW and distance KB, were evaluated using the Student t-test at $\alpha = 0.05$.

## RESULTS

**Virulence frequency**. In 2009–2010, 425 single isolates of the causal agent of barley powdery mildew, collected in different parts of Latvia, were tested on 13 differentials. Frequencies of virulence genes found in different parts of Latvia are presented in Table 3. In both years, no virulences were found against resistance genes from *SI1* and against *mlo5*. In contrast, the frequencies of virulences against the single resistance gene *Mla9* and against combined resistance genes *Mla6, Mla7, Mlk* and *Ml(La)* were high in both years of investigation in all samples and varied from 60.0% to 97.0%. In samples collected in Stende, between 2009 and 2010, frequencies of the genes *Va1, Va3* and *Va13* significantly decreased from 40.5% to 20.0%, from 40.5% to 18.0% and from 52.3% to 24.0%, respectively. In contrast,

Table 3

FREQUENCY OF VIRULENCE GENES (%) FOUND IN LATVIA IN 2009–2010

| Virulence genes | Da/09 | Da/10 | St/09 | St/10 | Pr/09 |
|---|---|---|---|---|---|
| *Va1* | 46.2 | 52.5 | 40.5 | 20.0 | 48.3 |
| *Va3* | 45.0 | 50.0 | 40.5 | 18.0 | 51.7 |
| *Va6* | 71.3 | 70.0 | 94.2 | 86.2 | 85.0 |
| *Va7* | 65.0 | 60.0 | 86.0 | 81.4 | 70.0 |
| *Va9* | 66.3 | 62.5 | 97.0 | 88.7 | 73.3 |
| *Va12* | 75.0 | 72.5 | 65.1 | 55.3 | 83.3 |
| *Va13* | 42.5 | 46.3 | 52.3 | 24.0 | 41.6 |
| *Vk* | 78.8 | 78.8 | 86.6 | 82.5 | 76.6 |
| *V(La)* | 83.8 | 81.3 | 71.8 | 64.7 | 75.0 |
| *V(SI)* | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| *V(St)* | 23.8 | 23.8 | 37.2 | 36.9 | 21.7 |
| *V(Go)* | 22.5 | 23.8 | 39.1 | 40.8 | 23.3 |
| *V(Me)* | 20.0 | 20.0 | 39.1 | 39.5 | 28.3 |

increases in frequencies of *Va1* from 46.2% to 52.5%, *Va3* from 45.0% to 50.0% and *Va13* from 42.5% to 46.3% were observed in Daugavpils. Virulences from '*Steffi'* and *'Goldie'* were significantly higher in samples collected in Stende.

**Virulence complexity and diversity parameters**. The lowest virulence complexity detected in Latvia in 2009–2010 (two virulence genes) was found in one pathotype collected in Daugavpils. The highest virulence complexity represented by 12 virulence genes was found in 11 pathotypes collected in Daugavpils and Stende. In both years of investigation, the mean complexity varied from 6.33 ± 0.30 to 10.00 ± 0.44. In Daugavpils, a tendency of increasing mean complexity in cleistothecia samples was observed. In 2009–2010, mean complexity of isolates in samples collected on different host genotypes differed between Stende and Priekuļi (Table 4). In total, 95 pathotypes were detected in 425 isolates. Among different samples and years, number of pathotypes with frequency more than 5% varied from 2 to 6 and number of pathotypes per sample from 2 to 27. The lowest number of pathotypes was detected in the Stende population, while in populations of Daugavpils and Priekuļi high numbers of pathotypes were found. The highest richness 0.83 was found in Priekuļi in samples collected in 2009 on 'Rolfi', and the lowest 0.09 in Stende in 2009 in samples collected on 'Annabell'

In both years of investigation, the pathotype *a7 a9 a12 k La* was dominant in Daugavpils only. In 2009, pathotypes *a1 a3 a6 a9 a13 k; a6 a7 a9 a12 k la St Go Me* and *a1a3 a6 a7 a9 a12 a13 k La* were dominant in Stende. In contrast, in 2010 other pathotypes (*a6 a7 a9 St Go Me; a6 a7 a9 St Go Me; a3 a13 k Go* and *a1 a3 a6 a7 a9 a12 a13 k la St Go Me*) had highest frequency (Table 5). The pathotype *a1 a3 a13* were detected in Daugavpils only. During 2009–2010, a clear tendency to increasing of richness was observed in isolates from Stende.

Nei index, Müller's index, Kosman index, Shannon index and Simpson indexes showed higher diversity in Daugavpils and Stende during 2009–2010 (Table 6). In 2009, the Kosman distance between Daugavpils and Priekuļi populations was 0.102, between Daugavpils and Stende 0.215 and between Stende and Priekuļi — 0.190. In 2010, a higher Kosman distance (0.304) between Daugavpils and Stende was observed. A significant Kosman distance (0.318) between years of investigation was observed in Stende, but was low (0.101) in Daugavpils. The genetic distances between populations in 2009 and 2010 was 0.159 (Table 7).

## DISCUSSION

Local populations of *Blumeria graminis* f.sp. *hordei* can change due to mutation, migration, recombination and direct selection, and therefore new dangerous pathotypes spread rapidly (Dreiseitl, 2000). In Latvia, evaluation of virulence was begun in 1981, which was the first study of this kind in the Baltic States. Until 1994, observations were carried out only in the central part of Latvia (Rashal and Tueryapina, 1996; Tueryapina *et al*., 1997). Since 1995, a regular study of the genetic structure of the pathogen was conducted in the south-eastern part of Latvia, in the Latgale region (Kokina and Rashal, 2004; 2005a; 2005b; 2007; 2008). The pathogen had not been studied previously in other parts of the country and there were no available data on virulence frequencies, distribution, complexity and pathotypes, although in Latvia, there are many domestic and foreign varieties grown.

Table 4

COMPARISON OF DIFFERENT POPULATIONS OF *Blumeria graminis* f.sp. *hordei* IN LATVIA IN 2009–2010

| Parameter | Da/09/ con | Da/09/ cl | Da/10/ con | Da/10/ cl | St/09/ cl/1 | St/09/ cl/2 | St/09/ cl/3 | St/09/ cl/4 | St/09/ cl/5 | St/10/ cl/1 | St/10/ cl/2 | St/10/ cl/3 | St/10/ cl/4 | Pr/09/ cl/1 | Pr/09/ cl/2 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| No. of isolates | 40 | 41 | 40 | 40 | 21 | 21 | 20 | 21 | 21 | 31 | 23 | 24 | 24 | 30 | 31 |
| Total number of pathotypes | 27 | 22 | 26 | 24 | 6 | 2 | 2 | 4 | 4 | 10 | 12 | 11 | 12 | 18 | 25 |
| No. of pathotypes with frequency higher than 5% | 3 | 5 | 2 | 5 | 5 | 2 | 2 | 2 | 3 | 4 | 3 | 5 | 6 | 6 | 2 |
| Mean complexity [2] | 8.00 ±0.28 | 9.00 ±0.43 | 9.01 ±0.25 | 10.00±0 .44 | 7.33 ±0.32 | 7.00 ±0.32 | 7.50 ±0.34 | 7.80 ±0.40 | 7.10 ±0.31 | 7.32 ±0.23 | 7.35 ±0.53 | 6.33 ±0.30 | 6.88 ±0.41 | 7.86 ±0.22 | 8.20 ±0.25 |
| The highest virulence complexity/number of isolates [1] | 9/4 | 12/3 | 9/1 | 12/3 | 9/4 | 9/14 | 9/10 | 9/13 | 9/5 | 9/3 | 12/5 | 9/3 | 10/2 | 9/6 | 9/1 |
| The lowest virulence complexity/number of isolates | 2/1 | 3/3 | 3/4 | 3/3 | 3/1 | 6/7 | 6/10 | 3/1 | 3/1 | 3/1 | 5/1 | 3/1 | 5/11 | 5/1 | 4/3 |
| Frequency of the dominant pathotype (%) | 25.0 | 12.5 | 22.5 | 12.5 | 28.6 | 66.7 | 50.0 | 65.0 | 52.4 | 45.2 | 30.4 | 20.8 | 20.8 | 20.0 | 16.7 |
| Richness [2] (number of pathotypes/ number of isolates) | 0.67 | 0.55 | 0.65 | 0.60 | 0.28 | 0.09 | 0.10 | 0.20 | 0.20 | 0.32 | 0.52 | 0.45 | 0.5 | 0.60 | 0.83 |

[1] different pathotypes were detected in the tested isolates

[2] all values for different populations and years were significantly different

Table 5

DOMINANT PATHOTYPES IN THE POPULATION OF *Blumeria graminis* f.sp. *hordei* IN LATVIA IN 2009–2010

| Pathotype | Number of corresponding isolates | | | | |
|---|---|---|---|---|---|
| | Da-2009 | Da-2010 | St-2009 | St-2010 | Pr-2009 |
| *a1 a3 a6 a7 a9 a12 k La* | | | 6 | 14 | |
| *a6 a7 a9 a12 k La* | 15 | | | 7 | 11 |
| *a6 a7 a9 a13 St Go Me* | | | 11 | 5 | |
| *a6 a7 a9 St Go Me* | | | | 5 | |
| *a1 a3 a 13 k Go* | | | | 5 | |
| *a1 a3 a6 a7 a9 a12 a13 k La St Go Me* | | | | 5 | |
| *a1 a3 a6 a9 a13 k* | | | 10 | | |
| *a6 a7 a9 a12 k La St Go Me* | | | 13 | | |
| *a1 a3 a6 a7 a9 a12 a13 k La* | | | 14 | | |
| *a7 a9 a12 k La* | 5 | 14 | | | |

Da-09, Daugavpils in 2009; Da-10, Daugavpils in 2010; St-09, Stende in 2009; St-10, Stende in 2010; Pr-09, Priekuļi in 2009

Table 6

PARAMETERS OF DIVERSITY WITHIN POPULATIONS OF *Blumeria graminis* f.sp. *hordei* IN LATVIA IN 2009–2010

| Parameter of diversity | Da-2009 | Da-2010 | St-2009 | St-2010 | Pr-2009 |
|---|---|---|---|---|---|
| Total number of isolates | 80 | 80 | 103 | 102 | 60 |
| Nei index | 0.374 | 0.384 | 0.341 | 0.355 | 0.350 |
| Müller's index | 0.379 | 0.389 | 0.344 | 0.358 | 0.355 |
| Kosman expected index | 0.566 | 0.600 | 0.531 | 0.557 | 0.519 |
| Shannon normalized index | 0.717 | 0.729 | 0.473 | 0.680 | 0.734 |
| Simpson index | 0.937 | 0.944 | 0.866 | 0.939 | 0.930 |

Da-09, Daugavpils in 2009; Da-10, Daugavpils in 2010; St-09, Stende in 2009; St-10, Stende in 2010; Pr-09, Priekuļi in 2009

Table 7

KOSMAN'S ASSIGNMENT-BASED DISTANCE (KOSMAN, 1996) BETWEEN POPULATIONS OF *Blumeria graminis* f.sp. *hordei* IN LATVIA IN 2009–2010

| Populations | Da-10 | St-09 | St-10 | Pr-09 | 2010 |
|---|---|---|---|---|---|
| Da-09 | 0.101 | 0.215 | 0.203 | 0.102 | |
| Da-10 | | 0.211 | 0.304 | 0.107 | |
| St-09 | | | 0.318 | 0.190 | |
| St-10 | | | | 0.193 | |
| 2009 | | | | | 0.159 |

Da-09, Daugavpils in 2009; Da-10, Daugavpils in 2010; St-09, Stende in 2009; St-10, Stende in 2010; Pr-09, Priekuļi in 2009

In 2009–2010, virulence frequencies of *Va6, Va7, Va9, Va12, Vk* and *VLa* were similarly high in all locations. This is likely due to the lack of a geographic barrier, and a change in one location can spreads to other regions (Zhu *et al*., 2010). Genes *Mla6, Mla7, Mla9, Mla12, Mlk* and *MlLa* can be considered as typical unnecessary resistance genes in Latvia. These resistance genes, used in approximately 700 cultivars, were gradually overcome by new pathogen pathotypes in a period of four to five years in Europe (Czembor, 2000). Significant differences of virulence in the pathogen population between samples collected in different parts of Latvia were detected for *Va1, Va3* and *Va13*. Significantly lower frequencies of *Va1, Va3* and *Va13* were detected in Stende in 2010. A clear tendency of increasing frequency of these genes was observed in some previous years in South-Eastern Latvia (Kokina and Rashal, 2004; Kokina and Rashal, 2005a; Kokina and Rashal, 2006; Kokina and Rashal, 2008). In many European countries, *Va1, Va3* and *Va13* had moderate to high frequency approximately ten years ago (Hovmøller *et al*., 2000). Presently, these genes are still effective in Central and Eastern Asia, for example, in winter barley regions of China. Dreiseitl and Wang (2007) found no isolates that could overcome *Mla1, Mla3, Mla13, Mla6, Mla7, Mla9, Mla12* and *MlLa*. Virulence frequencies of *Mlk* were very low. In the Zhu *et al*. (2010) study, no isolate was found to be virulent to *Mla1, Mla3, Mla6, Mla9, Mla13* and the virulence frequency to *Mla7, Mla12* and *Mlk* was very low.

In 2009–2010, a tendency of increasing *V(Me), V(St)* and *V(Go)* frequency was observed. The presence of the these virulences in the Latvian population of the pathogen can be explained by the fact that the corresponding pathotypes spread from Western Europe, where increasing trends were detected in 2000 already (Hovmøller *et al*., 2000). It is quite possible that 'Steffi', 'Goldie' and 'Meltan' resistance genes will lose their effectiveness in the near future, but several resistance genes are still present in these varieties.

Mlo resistance does not correspond to a gene-for-gene system. Since 1979, the barley *mlo* gene has been defined as highly effective and Mlo resistance confers nearly total resistance against fungal penetration attempts. Cytological investigation has shown in Mlo genotypes the ability to form effective papillae at sites of attempted penetration (Jarosch *et al*., 2003). Presently, this resistance is still effective, and is the most used resistance in spring barley grown through-

out Europe. In 2009–2010, this resistance was effective also in Latvia, and only some sporadic pustules of the pathogen were detected on the differential with *mlo5*.

Breeding for resistance is a cheap and environmentally safe approach to reduce loss in yield caused by powdery mildew of barley, which can reach 20% in Europe (Czembor, 2000). However, breeding for resistance depends on having gene pools from which new genes can be introduced into existing cultivars, and new none-specific resistance sources are needed. Barley line *SI1* was considered as a new resistance source for the control of powdery mildew (Hovmøller *et al*., 2000). Not any isolate with virulence to *SI1* was detected in Latvia in 2009–2010, as well asnor in previous years. Further observation of this virulence is necessary in Latvia and elsewhere in Europe.

According to Limpert (2008), for wind-dispersed nomadic diseases (including barley powdery mildew), although pathotypes are dispersed anywhere, predominantly dispersal in Europe is from West to East in the direction of prevailing winds. Complexity or the number of virulence genes per pathogen accumulate in this direction, because of migration and selection (Limpert and Bartoš, 2002). This is also apparent in Latvia, where the lowest complexities was observed in Stende, located in the Western part of Latvia. Mean complexity accumulates in the Eastern direction, shown by a higher level in Priekuļi and even higher in Daugavpils.

In Daugavpils, the population of *Blumeria graminis* f.sp. *hordei* had particularly high diversity. In 1995, the pathotype *a1 a3 a13* was detected in Latvia for the first time. In 2009–2010, this new and dangerous pathotype was found only in Daugavpils. This might be due to possible gene flow (spore migration from neighbouring Lithuania and Belarus). Also, in south-eastern Latvia, where Daugavpils is located, the climatic conditions are harsher, with lower winter temperatures. This might explain the favourable selection of new genotypes with possibly higher viability, but more vulnerable genotypes. Based on data presented here, it is possible to choose the best strategy for resistance breeding under Latvian conditions and to create new varieties with durable resistance.

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## MIEŽU MILTRASAS IZSRAISĪTĀJA *Blumeria graminis* f.sp. *hordei* POPULĀCIJAS MONITORINGS LATVIJĀ, 2009–2010

Miežu miltrasas izraisītāja paraugi tika ievākti 2009.–2010. gadā. Daugavpils apkārtnē (Latgale), Stendē (Kurzeme) un Priekuļos (Vidzeme). Katrā populācijā noteiktas patogēna virulences gēnu frekvences, virulences gēnu skaits katram individuālajam izolātam un patotipi. Ievērojamas frekvenču atšķirības starp populācijām konstatētas virulences gēniem *Va1, Va3* un *Va13*. Populāciju raksturojošie rādītāji (Neja, Millera, Kosmana, Šenona un Simpsona indeksi) liecina par ievērojamu daudzveidības palielināšanos Latgalē un Kurzemē pētījuma laikā. Daugavpils apkārtnē ievāktajiem paraugiem tika konstatēta īpaši liela patotipu daudzveidība.