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RESISTANCE TO PVM IN POTATO PARENTAL LINES BRED IN MŁOCHÓW RESEARCH CENTER, IHAR

ABSTRACT

During last 22 years in the Potato Genetics and Parental Lines Department at Młochów Research Center more than 210 parental lines resistant to *Potato virus M* (PVM) have been bred. Genes controlling resistance to PVM originated from two sources *Solanum megistacrobium* (the gene *Rm*) or/and *S. gourlayi* (the gene *Gm*) were introduced into tetraploid breeding material. From these resistant lines 59 progenitors were chosen and offered to the potato breeders. The first resistant to PVM cultivar Triada and second one cv. Korona were registered in Poland in 1996 and 2002, respectively. At present 13 advanced breeding clones from different breeding stations derived from progenitors offered to breeders in 1986–1990 are evaluated in Preliminary Trials. One candidate cultivar tested in Registration Trials appeared to be resistant to PVM. Moreover 15 advanced breeding clones derived from progenitors offered during 1991–1995 were tested in Preliminary Trials. The mechanical and graft inoculations were used to identify the type of resistance to PVM in potatoes. The parental lines, which resistance to PVM is controlled by the gene *Rm* or/and *Gm*, express field resistance even under the high natural infection pressure. Increasing number of resistant to PVM new potato cultivars is expected.

Key words: potato, PVM, resistance genes *Rm*, *Gm*, reaction to infection

INTRODUCTION

In the production of seed and ware potatoes *Potato virus M* (PVM) is more dangerous in the Eastern Europe than in the Western Europe. The losses of tuber yield range from 10% to 75% in case of severe virus strain infecting very sensitive potato cultivar (Chrzanowska 1984). The level of resistance to PVM of potato cultivars grown in Poland is low and weakly differentiated: from 2 to 5 score (in 1–9 scale, 9=extremely resistant). The exception are cultivars: Triada and Korona registered in Poland, which show resistance to PVM under field conditions (score 7) (COBORU, 2001).

Polish potato cultivars infected in the field are often symptomless independently on pathogenicity of PVM strain (37% of cultivars are toler-

ant to PVM infection), what may cause difficulties in seed production. Most cultivars show severe symptoms, when the severe strain of PVM is spread in the field (Zagórska *et al.* 2000).

Two types of resistance to PVM in *Solanum* spp. were identified. One derived from *S. megistacrolobum* (the gene *Rm*) is connected with hypersensitive necrotic reaction (Dziewońska and Ostrowska 1977). The second (the gene *Gm*) derived from *S. gourlayi* is connected with resistance to infection, slow multiplication and slow systemic spread of virus in plants (Waś *et al.* 1980, Świeżyński *et al.* 1981).

This paper presents the results of breeding potato parental lines with resistance to PVM done at Młochów Research Center in 1980–2001. In total 214 parental lines resistant to PVM were propagated in the field. Two types of the resistance to PVM governed by the genes *Rm* and/or by *Gm* were characterized.

MATERIALS AND METHODS

Plant material

Two groups of plant material were tested for resistance to PVM: (1) 75 potato cultivars and 214 parental lines from Laboratory of Parental Lines and Breeding Methods at Młochów and (2) 13 progenitors (advanced parental lines) expected to be resistant to PVM and two Polish cultivars as a control: Klepa and Tokaj. Cv. Klepa reacts with very severe symptoms and cv. Tokaj reacts with very weak symptoms to infection with PVM.

Virus

Severe strain of PVM from potato cv. Uran maintained in the collection of viruses at Młochów was used for the inoculation of potato plants. PVM was multiplied in tomato plants *Lycopersicon esculentum* cv. Najwcześniejszy.

Methods of inoculation

Mechanical inoculation: Potato tubers were presprouted on light without chemicals breaking dormancy and without growth retardants. Tuber pieces with sprouts were excised and put into pots (10 cm of diameter) in the greenhouse. After two weeks ten plants of each cultivar/line were mechanically inoculated with PVM. Inoculum was prepared from sap of PVM infected tomato by grinding of leaves mixed with buffer 0.057 M K_2HPO_4 . Leaves of young potato plants were dusted with carborundum powder (400–600 mesh) and rubbed with a sponge dipped in the inoculum. Two weeks after inoculation plants were planted in the field and they were observed during vegetation period. Tubers were collected on the end of August. Six months later tubers were presprouted and planted in greenhouse for tuber indexing.

Graft inoculation: Five plants of each line expected to be resistant to PVM were inoculated by grafting with scions from tomato plants infected with PVM. Grafted plants were placed in plastic boxes for two weeks to keep higher humidity. Four weeks after grafting new potato lateral shoots were tested serologically. Tubers from grafted plants were collected. After six months of storage tubers were cut and necrotic symptoms were observed. Tubers of grafted plants were collected and stored in low temperature (8–10°C) for six months.

Disease assessment

Disease symptoms were observed during experiment. Serological test was performed by DAS–ELISA. The polyclonal antibodies against PVM produced in IHAR, Dept. of Seed Production Laboratory in Gdańsk were used.

RESULTS

Reaction of Polish potato cultivars to the severe strain of PVM after mechanical inoculation of plants

Severity of symptoms was estimated on daughter tuber progeny of plants infected with severe strain of PVM (Zagórska *et al.* 2000). Potato cultivars and breeding clones could react with malformation of leaves and dwarfing of plants (Fig. 1). Plants of 28 cultivars out of 76 tested showed mild symptoms independently on virus strain, plants of 12 cultivars did react with very severe symptoms after infection with severe strain of PVM. Plants of other 36 cultivars expressed variable reaction, sometimes with severe symptoms (Table 1).



Fig. 1 The samples of three resistant to PVM parental lines (left), no disease symptoms and three susceptible cultivars (right), severe mosaic, malformation of leaves and dwarfing of secondary infected plants are visible

Table 1

**Reaction of 76 potato cultivars after mechanical inoculation
of plants with severe strain of PVM**

Level of resistance*	Severity of disease symptoms:					
	Mild		Moderate		Very severe	
7	Triada [®] , Korona					
5	Aksamitka	Ruta	Albina [®]	Kuba	Aster	Harpun
	Irga	Tokaj	Baszta	Orlik	Bard	Mila
	Koral		Bryza		Drop	
3 – 4.5	Accent	Jasia	Anielka [®]	Lawina	Alicja	
	Arkadia	Kos	Balbina	Łucja	Barycz	
	Beata	Lord	Bekas [®]	Lena [®]	Beata	
	Danusia	Perkoz	Bila	Maryna [®]	Cedron	
	Denar	Rywal	Bzura	Mors	Panda	
	Glada	Sante	Cykada [®]	Rybitwa		
	Gloria	Wawrzyn	Fregata	Salto		
	Ibis	Wigry	Frezja	Sumak		
	Irys	Wiking	Grot	Tara		
	Jantar		Ikar [®]	Vital [®]		
			Jagna	Wolfram		
		Koga				
2 – 2.5	Kolia	Omulew	Ania	Meduza	Klepa	
	Oda	Orłan	Dunajec	Muza	Nimfy	
			Hinga [®]	Vistula [®]		
Total number (%)	28 (37%)		36 (47%)		12 (16%)	

* – according to the scale 1–9, where 9 = extremely resistant, evaluated by Dr. J. Pietrak (COBORU 2001), [®] – cultivar with moderate disease symptoms, but sporadically reacting with very severe

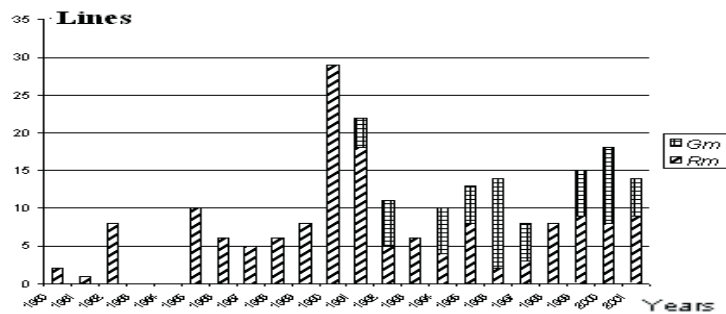


Fig. 2 Sources of resistance to PVM in 214 parental lines bred in IHAR, Młochów Research Center during 1980–2001

Progress in breeding of potato parental lines resistant to PVM

During twenty two years of breeding, 214 parental lines from the Laboratory of Parental Lines and Breeding Methods at Młochów have been

identified as highly resistant to *PVM* (Fig. 2). Resistance of these lines was confirmed after inoculation in greenhouse condition (Fig. 1). Till 1990 resistant lines carried only the gene *Rm*. In 1991 the gene *Gm* have been introduced into breeding material. At present, some of parental lines may possess both the *Rm* and *Gm* genes, but this was not established.

Characterization of reaction to *PVM* of potatoes with resistance controlled by the genes *Rm* and *Gm*

Under natural infection pressure parental lines which possess the genes *Rm* or *Gm* are not infected with *PVM* (Table 2). Also they are not infected with *PVM* after mechanical inoculation of young plants in the greenhouse/field conditions. Very rarely individual plants (with the gene *Gm*) could be infected.

Reaction of parental lines resistant to *PVM* according to applied method of inoculation

Table 2

Method of inoculation	Plants of parental lines derived from parents carrying the genes			
	<i>Rm</i>		<i>Gm</i>	
	Primary infection	Secondary infection	Primary infection	Secondary infection
Natural infection under field condition	0	0	0	0
Mechanical inoculation, greenhouse and field conditions	0	0	0	Sporadically mild symptoms low A_{405} values
Graft inoculation, greenhouse condition	Above 20% of plants with necrotic reaction	0 or total necrosis of plants	0	

0 = No symptoms on plants and the virus was not detected using ELISA

Reaction of resistant potatoes after graft inoculation with *PVM*

The sample of parental lines with expected genes *Rm* or/and *Gm* were graft inoculated with *PVM* (Table 3). The mean A_{405} values in ELISA for resistant lines were low (up to 0.11). In some individual plants the value increased up to 0.38 towards 0.8 and 1.1 in plants of susceptible controls (cvs Tokaj and Klepa). Lines possessing the gene *Rm* show necrotic lesions on lateral shoots and top necrosis. In tubers collected from grafted plants distinct necrotic lesions were observed (Fig. 3) and plants grown from them are often dwarf and stunt (Fig. 4).

The best indicator of the presence of the gene *Rm* in potato genotypes were necroses appearing in tubers stored for six months after graft inoculation of mother plants.

Utilization of progenitors in breeding of potatoes

From progenitors resistant to *PVM* offered to Polish breeders three cultivars (Triada, Tokaj and Korona), were developed. In addition one

Table 3

**Reaction of parental lines resistant to PVM after grafting
(5 plants of each parental line were grafted)**

Parental line	Genes expected in parental line	Number of plants with necrotic reaction on lateral shoots	A ₄₀₅ value in ELISA		Symptoms in tubers from grafted plants	Presumptive presence of gene(s) in parental line
			Mean	Max		
99 × ^A 20	<i>Gm Rm</i>	0	0.07	0.24		<i>Gm</i>
99 × ^A 43	<i>Gm Rm</i>	0	0.05	0.06		<i>Gm</i>
99 × ^A 15	<i>Gm,Rm</i>	5	0.04	0.11	tn	<i>Rm</i>
99 × ^A 11	<i>Gm,Rm</i>	4	0.03	0.05	tn	<i>Rm</i>
99 × ^B 18	<i>Gm,Rm</i>	4	0.03	0.11	tn	<i>Rm</i>
99 × ^B 61	<i>Rm</i>	4	0.10	0.38	tn	<i>Rm</i>
99 × ^A 8	<i>Gm,Rm</i>	3	0.04	0.21	tn	<i>Rm</i>
99 × ^A 10	<i>Gm,Rm</i>	3	0.02	0.03	tn	<i>Rm</i>
99 × ^B 42	<i>Gm,Rm</i>	3	0.03	0.04	tn	<i>Rm</i>
99 × ^A 57	<i>Gm,Rm</i>	2	0.06	0.13	tn	<i>Gm,Rm</i>
99 × ^A 19	<i>Gm,Rm</i>	1	0.11	0.26	tn	<i>Gm,Rm</i>
99 × ^A 64	<i>Gm,Rm</i>	0	0.04	0.05	tn	<i>Gm,Rm</i>
99 × ^B 21	<i>Gm,Rm</i>	0	0.03	0.04	tn	<i>Gm,Rm</i>
cv. Klepa		5 ¹	0.80	1.10		
cv. Tokaj		5 ²	0.70	0.80		
Healthy plants			0.05	0.06		

tn= necrotic lesions in tubers

¹ – with severe mosaic, malformation of leaves

² – with mild mosaic



Fig. 3 Potato tubers collected from grafted plants of three resistant parental lines carrying the gene *Rm* – numerous necrotic spots in flesh are distinct



Fig. 4 The healthy potato plant (left) and the plant grown from the tuber showing necroses (right). This type of symptoms is characteristic for the hypersensitive reaction of plant to PVM

candidate cultivar and 28 advanced breeding clones were tested in 2000 in official trials (Table 4).

Table 4
Utilization of progenitors resistant to PVM in potato breeding programs

Year(s)	Number of progenitors resistant to PVM offered to breeders	Resistance genes present in progenitors	Cultivars or advanced breeding clones derived from progenitors up to 2000	Year of registration
1985	4	<i>Rm</i>	cv. Triada (7) cv. Tokaj (5)	1996 1998
1986 – 1990	18	<i>Rm</i>	cv. Korona (7) One candidate cultivar* in Registration Trials 13 advanced breeding clones in Preliminary Trials in 2000	
1991 – 1995	14	<i>Rm, Gm</i>	15 advanced breeding clones in Preliminary Trials in 2000	2002
1996 – 2000	23	<i>Rm, Gm</i>		
Total	59	–	Three cultivars, one candidate cultivar, 28 advanced breeding clones	

* P. 90.743/33 resistant to PVM, with the gene *Rm* derived from PW 155

DISCUSSION

PVM is more economically important virus in Eastern Europe than in other parts of the world (Jeffries 1998). Potato cultivars are mostly susceptible to *PVM* what makes easier spreading of virus in grown potatoes.

Several variants of *PVM* strains could be distinguished by the symptoms induced on potato and test plants and by serological relationship (Wetter 1972, Kowalska 1978, Cavileer *et al.* 1998, Chrzanowska *et al.*

2000). Disease symptoms caused by *PVM* ranged from very slight to severe, depending on virus strain and potato variety. Leaflet malformation, rolling of the top of plant and even necroses on petioles and stems could be observed (Zagórska *et al.* 2000, Chrzanowska and Zagórska 2001).

Mild strains of *PVM* cause mild symptoms on all potato cultivars. Severe strain of *PVM* used for the inoculation of potatoes could cause severe symptoms on about 30% of cultivars grown in Poland (Chrzanowska and Zagórska, 1996)

Two types of resistance to *PVM* have been introduced to potato parental lines bred in the Potato Genetics and Parental Lines Department at Młochów and offered to breeders since 1985 (resistance based on the gene *Rm*) and since 1991 (resistance based on the gene *Gm*). Such potato parental lines are resistant to *PVM* in natural infection pressure and after mechanical inoculation. At the first stages of selection of resistant parental lines the spray gun technique is used for inoculation of seedlings (Siczka 2001). The reaction of parental lines possessing the genes: *Rm* and *Gm* has been described using graft inoculation of plants.

Thirteen parental lines were selected previously as resistant to *PVM*. Twelve of them have parents carrying both genes for resistance. Only one clone (99-X^B-61) was originated from parent hypersensitive to *PVM* carrying the gene *Rm* (Table 3).

After graft inoculation with *PVM* of nine parental lines the necrotic symptoms were observed on lateral shoots. From one to five grafted plants showed symptoms. Four parental lines did not show necroses on lateral shoots. The resistance of two of them is probably based on the presence of the gene *Gm* (99-X^A-20, 99-X^A-43). In two others (99-X^A-64, 99-X^B-21) the presence of necrotic lesions in tubers may indicated the action of the gene *Rm*. This reaction is in agreement with previous observations described by Chrzanowska (1988).

According to Mietkiewska (1999) in potatoes possessing both genes (*Rm* and *Gm* together) after graft inoculation of plants the presence of the gene *Gm* may suppress the hypersensitive reaction to *PVM* governed by the gene *Rm*. In such case the necrotic symptoms on lateral shoots are not visible.

The results presented in this paper pointed out that symptoms on lateral shoots after grafting may be not sufficient to determine the presence of the gene *Rm*. Observation of necroses in tubers collected from grafted plants is recommended in addition for proper discrimination of the source of resistance to *PVM*.

Till now it is evident that only cultivars: Triada and Korona and candidate cultivar P.90.743/33 tested in Registration Trials are resistant to *PVM*. Increasing number of resistant clones/cultivars is expected in near future as 28 advanced breeding clones tested in official trials originate from progenitors governing resistance to *PVM* were offered to breeders since 1986 (Table 4).

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