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DISTRIBUTION AND RACE COMPOSITION OF DOWNY MILDEW (*Plasmopara halstedii* (Farl.) Berl. and de Toni) IN BULGARIA

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SUMMARY

Downy mildew caused by the fungus *Plasmopara halstedii* is the main disease on sunflower in Bulgaria. In recent years a number of authors have reported the occurrence of new more virulent races of the pathogen. According to other authors these races demonstrate resistance to the fungicides used up to now. This fact is rather alarming and imposes the necessity of annual researches with the aim of following the changes in the downy mildew race variability.

In the period 1995-1997 downy mildew isolates were collected from the following locations: Bourgas, Boyanovo, Karnobat, Ognyanovo, Selanovtsi, Kroushari, Lovech, Koubrat, Brashlyan, Sitovo, Tervel, Targovishte, IWS "Dobroudja" and Dobrich. The samples were assessed for virulence on a set of sunflower differential - lines under greenhouse conditions.

The obtained results do not reveal a great race variability of downy mildew population in Bulgaria. In the period of study two races of the pathogen were identified: race 1 which infects the differential lines without genes for resistance to the pathogen. It is distributed in all sunflower production areas of the country. The other one is race 2. It is of limited distribution and has been registered in individual fields of north-east and north-west Bulgaria. It attacks the differential lines carrying the resistance gene *Pl-1*.

Key words: downy mildew, distribution, race composition

INTRODUCTION

Downy mildew in sunflower caused by the pathogenic fungus *Plasmopara halstedii* (Farl.) Berl. and de Tony is the main disease on sunflower in Bulgaria. Control over it is presently realized with the help of breeding, by developing resistant varieties and hybrids, or by pre-sowing treatment of seeds with metalaxyl-containing fungicides.

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In recent years a number of authors have reported the occurrence of new, more virulent races of the pathogen which overcome the resistance of the varieties and hybrids introduced into practice.

According to Maširević (1992), a new downy mildew race (race 4) has been established in Yugoslavia, which attacks all genotypes with resistance gene *Pl-1*, *Pl-2*, *Pl-4* and *Pl-5*. Tourvieille *et al*. (1988) have registered that a new race for the conditions of France is distributed in the region of Indre (race B) - a downy mildew race that attacks the genotypes with the *Pl-1* gene for resistance. Later researches (Mouzeyar *et al*., 1994) have shown, that the race composition of the pathogen in France is enriched with other two races - C and D. The investigations of Rashid (1993) on the physiological specialization of downy mildew in west Canada reveal a rich race diversity of the pathogen. The author has determined six races (races 1, 2, 3, 4, 6 and 7); races 3 and 4 are the most widely distributed, but the most virulent race reported is race 2. In their studies on the pathogen variability in Hungary, Virányi and Gulya (1993) report five races - 1, 2, 3, 4 and 7 or 9. A rich diversity in the downy mildew races in North and South America has been established by Gulya *et al*. (1991) who distinguished seven races - 1, 2, 3, 4, 5, 6 and 7. Summarizing the race variability of downy mildew in Europe, Virányi and Gulya (1996) reported 9 races. In their opinion, there are 3 races in Germany (1, 4 and 5), 6 races in Hungary (1, 2, 3, 4, 8 and 9), 4 races in France (1, 4, 6 and 8), 3 races in Yugoslavia (1, 4 and 8), 2 races in Bulgaria (1 and 4), 2 races in Italy (1 and 3), 2 races in Romania (1 and 9), and 2 races in Spain (1 and 8).

According to other authors these races demonstrate resistance to the fungicides used up to now (Virányi *et al*., 1992).

The facts mentioned above show that sunflower downy mildew is forming new races in the process of its evolution, which imposes the necessity to carry out systematic studies on the distribution and race variability of the pathogen.

MATERIALS AND METHODS

In order to determine the race variability of sunflower downy mildew in Bulgaria and the distribution of the individual races, expeditions for collecting inoculum (fungus conidiospores) from the main sunflower production areas were organized in the period 1995-1997. The locations where downy mildew attacks were registered and from which we collected samples are given in Table 1.

In 1995 samples from 11 locations in the country were collected: Bourgas, Boyanovo, Karnobat, Ognyanovo, Selanovtsi, Koubrat, Brashlyan, Sitovo, Targovishte, IWS “Dobroudja” and Dobrich.

In 1996 downy mildew attacks were registered in the following locations: Boyanovo, Selanovtsi, Krushari, Lovech, Tervel, Targovishte, IWS “Dobroudja” and Dobrich.

Table 1: Race variability of downy mildew in Bulgaria for the period 1995-1997

Origin of downy mildew	Race / Year					
Location / District	1995		1996		1997	
	1	2	1	2	1	2
Bourgas / Bourgas	+	-				
Boyanovo / Yambol	+	+	+	-	+	+
Karnobat / Bourgas	+	-				
Ognyanovo / Plovdiv	+	-				
Selanovtsi / Pleven	+	-	+	-	+	-
Krushari / Dobrich			+	-	+	-
Lovech / Lovech			+	-		
Koubrat / Razgrad	+	-				
Brashlyan / Russe	+	-			+	-
Sitovo / Silistra	+	+			+	+
Tervel / Razgrad			+	-	+	-
Targovishte / Razgrad	+	-	+	-		
Dourankulak/ Dobrich					+	-
Pleven / Pleven					+	-
Byala / Russe					+	-
IWS / Dobrich	+	+	+	+	+	+
Dobrich / Dobrich	+	+	+	+	+	+

The samples collected in 1997 were from the following 11 locations: Boyanovo, Selanovtsi, Krushari, Brashlyan, Sitovo, Tervel, Dourankulak, Pleven, Byala, IWS "Dobroudja" and Dobrich.

The inoculum was gathered from infected sunflower plants in commercial crops and from different varieties and hybrids in variety maintenance and testing stations. The same was kept through periodical infection (at 15-day intervals) of plants of Peredovik variety which is susceptible to all mildew races. In order to avoid mixing-up of the inoculum, the mildew-infected plants were grown in sterilized soil, in containers covered with glass cylinders. A differentiation key was used in order to specify mildew races, consisting of the following lines - differentials:

Peredovik	- Pl_0
RHA-266	- Pl_1
HA-61	- Pl_1, $Pl_{2/3}$
DM-2	- Pl_2 , Pl_5
HA-335	- $Pl_{6\ +.....}$
HA-337	- $Pl_{7\ +.....}$

One-hundred plants were infected from each differential line after the following method: seeds from differentials were placed for germination at the temperature of 22-23°C. At the length of radicles of 0.7-1.0 cm, the seedlings were infected with a suspension of fungus zoospores with an approximate concentration of 600 000 spores/ml. The soaking of seedlings took five hours, a period of time necessary for an effective infection, at the temperature of 15-16°C. The seedlings infected in this way were planted in

vegetational containers (2 x 50 seedlings) with a row of uninfected seedlings of Peredovik variety planted in each of these containers to check soil purity. In order to avoid mixing-up of the inoculum, through flight of spores, the containers were covered with plexiglass lids after the infection.

The response of the differential lines was reported after a 15-day incubation period. Since the differentials employed are homozygous as regards their mildew response, they indicate two types of response - resistant (R) and sensitive (S).

Table 2: Differential lines and their reaction to sunflower downy mildew in the period 1995-1997

Races in Bulgaria	Reaction of differential lines						Sample / isolate	
	Peredovik *Pl-0*	RHA-266 *Pl-1*	HA-61 *Pl -2/3*	DM-2 *Pl-2/5*	HA-335 *Pl-6+..*	HA-337 *Pl-7+..*	Number	%
1995								
1	S	R	R	R	R	R	11	73.3
2	S	S	R	R	R	R	4	26.7
1996								
1	S	R	R	R	R	R	8	80
2	S	S	R	R	R	R	2	20
1997								
1	S	R	R	R	R	R	11	73.3
2	S	S	R	R	R	R	4	26.7
1995-1997								
1	S	R	R	R	R	R	30	75.0
2	S	S	R	R	R	R	10	25.0

RESULTS AND DISCUSSION

The results obtained indicate that the sunflower mildew race diversity in Bulgaria is not very high. From the samples used in the period 1995-1997, with each one of them representing a single isolate, two pathogen races were established (Table 1, Table 2):

Race 1 - it infects only the Peredovik variety, with the rest of the lines of the differentiation key being resistant towards it. According to pathogenicity it corresponds to race 1 (European) from the world classification.

Race 2 - it overcomes the Peredovik variety resistance and the HA-266 bearing the resistance gene *Pl-1*. According to pathogenicity this race is identical with race 2 (Red River) from the world classification. These results do not differ from our research on race composition of sunflower mildew for the period of 1991-1994 (Shindrova, 1995).

Race 1 and race 2 were registered in each of the research years:

In 1995 we worked with 15 samples altogether. Race 1 was isolated from 11 of them, which is 73.3% of the analyzed isolates, and race 2 from 4 (26.7%).

During the following year (1996) we analyzed 10 samples. Eight of them (80.0%) corresponded to race 1 and 2 (20.0%) to race 2 of the pathogen.

The number of samples collected and studied in 1997 was 15, with 11 of them (73.3%) corresponding according to pathogenicity to race 1 and 4 (26.7%) to race 2 of the pathogen.

Figure 1: Distribution of downy mildew races in Bulgaria: (-race 1, **-race 2, ***-races 1+2)*

The data in Table 1 and Figure 1 give an idea of the distribution area of both races. They show that during the research period race 1 had an over-all incidence. It was isolated in all sunflower growing areas in Bulgaria. Race 2 has a more limited distribution. It was found only in Sitovo, Dobrich and IWS "Dobroudja" regions, *i.e.*, in northestern Bulgaria, southern Bulgaria and in Boyanovo (District of Yambol), respectively.

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EXTENSION Y RAZAS DEL MILDIU (*Plasmopara halstedii* (Farl.) Berl. y de Toni) EN BULGARIA

RESUMEN

El mildiu causado por el hongo *Plasmopara halstedii* es la más grave enfermedad del girasol en Bulgaria. Durante los últimos años, el número más grande de investigadores comunicó sobre la aparición de nuevas razas de este patógeno, que son más virulantes que las anterioraes. Según otros autores, estas razas muestran la resistencia a los fungicidas que se utilizaban hasta ahora. Este hecho es alarmante e impone la necesidad de investigar continuamente para seguir las variabilidades de razas del mildiu.

Durante el periodo 1995-1997, los aislados del mildiu eran coleccionados en las localidades: Bourgas, Boyanovo, Karnobat, Ognyanovo, Selanovtsi, Kroushari, Lovech, Koubrat, Brashlyan, Sitovo, Tervel, Targovishte, IWS "Dobroudja" y Dobrich. La virulencia de las muestras coleccionadas era determinada por medio del grupo de lineas diferenciales del girasol, cultivadas en las condiciones de invernáculo.

Los resultados obtenidos indican que la variabilidad de razas del mildiu en Bulgaria no es grande. Dos razas del patógeno fueron identificadas durante la investigación. Esas son la raza 1 que es susceptible de infectar las lineas diferenciales que no poseen los genes de resistencia al patógeno, y la raza 2, cuya extensión es limitada a los campos singulares en Bulgaria del Nordeste y Noroeste. Esta raza ataca las lineas diferenciales que poseen el gen para la resistencia *Pl-1*.

ÉTENDUE ET RACES DE LA ROUILLE (*Plasmopara halstedii* (Farl.) Berl. and de Toni) EN BULGARIE

RÉSUMÉ

La rouille provoquée par le champignon *Plasmopara halstedii* est la maladie la plus importante du tournesol en Bulgarie. Ces dernières années, un grand nombre de chercheurs ont annoncé l'apparition de nouvelles races de ce pathogène plus virulentes que les espèces connues jusqu'à maintenant. D'autres auteurs indiquent que ces races montrent une résistance aux fongicides utilisés jusqu'à maintenant. Ce fait est alarmant et il impose une recherche continue qui aura pour but de suivre la variabilité de ces races de rouille.

Entre 1995 et 1997, des isolats de rouille ont été recueillis dans les localités suivantes: Bourgas, Boyanovo, Karnobat, Ognyanovo, Selanovtsi, Kroush-

ari, Lovech, Koubrat, Brashlyan, Sitovo, Tervel, Targovishte, IWS "Dobroudja" et Dobrich. La virulence des échantillons recueillis a été confirmée grâce aux groupes des lignes différentielles de tournesol cultivés dans les conditions de serre.

Les résultats obtenus indiquent que la variabilité de la race de la rouille en Bulgarie n'est pas importante. Deux races de pathogènes ont été identifiées au cours des recherches. Il s'agit de la race 1 qui est de nature à infecter les lignes différentielles qui ne possèdent pas de gènes de résistance envers le pathogène et la race 2, dont l'étendue est limitée à certains champs en Bulgarie du nord-ouest et du nord-est. Cette race attaque les lignes différentielles qui possèdent le gène de résistance *Pl-1*.