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Susceptibility of Bean Genotypes to *Xanthomonas campestris* pv. *phaseoli* in Greenhouse Conditions

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SUMMARY

Plants of 17 bean genotypes were evaluated under greenhouse conditions for their reaction to *X. campestris* pv. *phaseoli*, the causal agent of common blight of beans, following leaf-spray inoculation with bacterial suspension (10^8 cfu/ml). The plants were evaluated based on the number of leaf lesions, and the disease severity index (DSI) was calculated. The evaluated genotypes showed various levels of susceptibility to *X. campestris* pv. *phaseoli* strain VS-1. The results of both experiments showed that the cultivar Oreol was the most resistant. The genotypes: KB 142, HR-45, Tisa and Panonski tetovac also showed low susceptibility with DSI values in the first trial ranging from 2.27 to 3.60. The same genotypes, with the exception of Panonski tetovac, were also categorized as low-susceptible to the bacterium in the second experiment, having the DSI values between 2.27 and 3.60. Most genotypes (Slavonski žutozeleni, Zlatko, Biser, Sremac, Naya Nayahit) were categorized as susceptible in the first experiment, including Panonski tetovac in the second one, while the genotypes Dvadesetica, Prelom and Oplenac displayed the highest susceptibility in both trials.

Keywords: Common blight; *X. campestris* pv. *phaseoli*; Bean genotypes; Disease resistance

INTRODUCTION

Xanthomonas campestris pv. *phaseoli*, the causal agent of common blight of beans, usually induces disease symptoms on bean leaves and pods. Leaf lesions develop into dark-brown or redish with growth, and are surrounded by narrow chlorotic halos, while pods are often girdled by red bands (Arsenijević et al., 1985; Arsenijević, 1997). The disease has become widespread in almost all bean growing regions (Lahman and Schaad, 1985), lowering the yield of susceptible cultivars as much as 50-80% (Wimalajeewa and Nancarrow, 1980). The bacterium is seedborne and soilborne. Furthermore, the ability of *X. campestris* pv. *phaseoli* epiphytic populations to survive on non-host weed plants (Arnaud-Santana et al., 1991; Gent et al., 2005) makes the pathogen difficult to control. In addition to regular cultural practic-

es and chemical treatments (Balaž, 1991; 2005) for disease control, the development of commercially acceptable bean cultivars resistant to *X. campestris* pv. *phaseoli* is one of the most suitable ways to overcome the disease (Kiryakov, 1999; Kiryakov and Genchev, 2000, 2003). Variability in the pathogenicity of *X. campestris* pv. *phaseoli* strains on different bean species and cultivars had been observed by Coyne and Schuster (1974). In their study, the highest resistance was demonstrated by *Phaseolus acutifolius* cultivars, as well as some cultivars and lines of *Phaseolus coccineus.* Ekpo and Saettler (1976) revealed two sources of resistance to *X. campestris* pv. *phaseoli*: GN Nebr. # 1 Sel. 27 (quantitatively inherited) and PI 207262 (controled by minor genes). Poryazov and Georgieva (1982) reported the existance of four groups of strains based on symptom appearance in three distinctive bean cultivars (Amboy, GN Ne sel. 27 and PI-150414). The cultivars GN Ne-1 sel.27 and PI-150414 showed high resistance to the first group of strains while they were susceptible to the fourth group. Based on the reaction of *P. acutifolius* to 30 investigated strains, Opio et al. (1996) determined eight pathogen races, and revealed the existance of natural sources of resistance to *X. campestris* pv. *phaseoli* in this bean species. Kiryakov and Genchev (2000) reported high to moderate resistance of several bean cultivars (XAN 159, HR 45, Oreol, PI 207262 and G.N. Jules) to as much as 80% of the strains tested. Moreover, Kiryakov and Genchev (2003) identified the bean line VAX 3-6 and cultivar XAN 159 as acceptable resistance gene donors.

In order to detect the sources of resistance to *X. campestris* pv. *phaseoli* the evaluation of a large number bean genotypes is necessary. Therefore, the objective of this study was to evaluate 17 bean genotypes most commonly grown in Serbia for their resistance to the causal agent of common blight of beans.

MATERIAL AND METHODS

Plant and bacterial material

Susceptibility to *X. campestris* pv. *phaseoli* was evaluated in 17 bean genotypes (Table 1). The experimental design was a randomised complete block with three replicates and five plants within each replicate. The experiment was performed twice.

The bacterial strain VS-1, previously determined as *X. campestris* pv. *phaseoli* (Todorović et al., 2006), was used for inoculation of bean plants. Bacterial cultures were maintained on YDC (Yeast extract – dextrose – $CaCO_3$) medium at 4°C. For inoculum preparation, bacteria were incubated on YDC at 26°C for three days. The bacterial inoculum was prepared by washing bacteria cultures with sterile distilled water. Concentration of bacteria in the suspension was adjusted to 10^8 cfu/ml using McFarland's scale and confirmed by a serial dilution plating method (Klement et al., 1990).

Experimental conditions and disease evaluation

Bean seedlings were transplanted into 10-cm diametar plastic pots containing substrate „B medium course" (Floragard, Germany) and kept on benches in a greenhouse. At the stage of first trifoliate leaf, the plants were inoculated by spraying with bacterial suspension (10^8 cfu/ml) (Kiryakov and Genchev, 1994). After inoculation the plants were kept under a plastic cover for 48 hours to provide 100% relative humidity. After cover removal, humidity was 75% in both experiments. Temperature was 20°C in the first experiment, and 28°C in the second trial. The results were analysed separately for each trial.

Fourteen days after inoculation, bean plants were classified into nine groups according to a scale based on leaf area covered by lesions, e.i. the percentage of chlorotic or necrotic leaf area (Kiryakov and Genchev, 1994; Kiryakov, 1999): 1 = no lesions; 2 = no lesions, up to 2% of leaf area covered by chlorosis or necrosis; 3 = up to 5% leaf lesions and 10% covered by chlorosis or necrosis; 4 = 10% lesions and 25% covered by chlorosis or necrosis; 5 = 20% lesions and 25% covered by chlorosis or necrosis, 6 = 30% lesions and 50% covered by chlorosis or necrosis; 7 = 40% lesions and 100% covered by chlorosis or necrosis; 8 = 50% lesions and 100% covered by chlorosis or necrosis; and 9 = more than 50% lesions and 100% covered by chlorosis or necrosis (Figures 1-9).

Disease was evaluated on the basis of disease severity index (DSI). The results were processed by analyses of variance and differences among tomato genotypes tested by Duncan's multiple range test.

RESULTS

The evaluated genotypes showed various levels of susceptibility to *X. campestris* pv. *phaseoli* strain VS-1 (Table 1).

Figure 1. 1 = no lesions
Slika 1. 1 = nema pega

Figure 2. 2 = no lesions, up to 2% of leaf area covered by chlorosis or necrosis
Slika 2. 2 = nema pega, ali hloroza ili nekroza zahvataju 2% lisne površine

Figure 3. 3 = up to 5% leaf lesions and 10% covered by chlorosis or necrosis
Slika 3. = do 5% pega na listu i 10% hloroze ili nekroze lista

Figure 4. 4 = 10% lesions and 25% covered by chlorosis or necrosis
Slika 4. 4 = do 10% pega i 25% hloroze ili nekroze

Figure 5. 5 = 20% lesions and 25% covered by chlorosis or necrosis
Slika 5. 5 = do 20% pega i 25% nekroza ili hloroza

Figure 6. 6 = 30% lesions and 50% covered by chlorosis or necrosis
Slika 6. 6 = do 30% pega i do 50% hloroza ili nekroza

Figure 7. 7 = 40% lesions and 100% covered by chlorosis or necrosis
Slika 7. 7 = do 40% pega, tj. do 100% hloroza ili nekroza

Figure 8. 8 = 50% lesions and 100% covered by chlorosis or necrosis
Slika 8. 8 = do 50% pega, odnosno 100% hloroza ili nekroza

In the first experiment, disease severity index of the tested bean genotypes varied between 1.27 and 8.80. The genotypes Oreol, KB 101, KB 100, A 55 and Darina showed low susceptibility to strain VS-1 with DSI values ranging between 1.27 and 2.00. The genotypes HR-45, KB 142, Tisa and Panonski tetovac were categorized as low susceptible since their DSI values were between 2.73 and 3.93, while the genotypes Slavonski žutozeleni, Zlatko, Biser, Naya Nayahit and Sremac were designated as susceptible (with DSI ranging between 4.27 and 5.47). Based on DSI values (5.47-8.80), Prelom, Dvadesetica and Oplenac were the most susceptible bean cultivars.

Figure 9. 9 = more than 50% lesions and 100% covered by chlorosis or necrosis
Slika 9. 9 = preko 50% pega na listu, tj. 100% nekroza ili hloroza lista

In the second experiment, DSI values of the bean genotypes tested ranged between 1.40 and 8.80. Oreol, KB 101, KB 100, A 55 and Darina were moderately resistant based on DSI between 1.40 and 1.87 whereas the genotypes KB 142, HR-45 and Tisa were low susceptible (DSI between 2.27 and 3.60). Most genotypes (Panonski tetovac, Slavonski žutozeleni, Zlatko, Biser, Sremac, Naya Nayahit) with DSI values ranging from 4.07 to 5.33 were categorized as susceptible in the second experiment, while Dvadesetica, Prelom and Oplenac showed the highest susceptibility.

DISCUSSION

Difficulties involved in controlling bacterial diseases are well known. The best results in controlling bacterial common blight of beans can be achieved by growing resistant bean cultivars, but satisfactory results in *X. campestris* pv. *phaseoli* control have also been obtained by using copper-based compounds (Balaž, 2005). Imports of new high-yield bean cultivars to Serbia have been extensive over the past few years as the range of bean products widened. One of the consequences is a lack of data regarding the susceptibility/resistance of

Table 1. Disease severity index of bean genotypes – experiments I and II
Tabela 1. Indeks oboljenja kod različitih genotipova pasulja – eksperimenti I i II

Experiment I – Eksperiment I		Experiment II – Eksperiment II	
Genotypes – Genotipovi	DSI* IO	Genotypes – Genotipovi	DSI* IO
Oreol	1.27 a	Oreol	1.40 a
KB 101	1.40 a	KB 101	1.47 a
KB 100	1.60 ab	KB 100	1.47 a
A 55	1.93 ab	A 55	1.73 ab
Darina	2.00 ab	Darina	1.87 ab
HR-45	3.00 bcd	HR-45	2.60 abc
KB 142	3.60 cde	KB 142	2.93 bcd
Tisa	3.80 cde	Tisa	3.60 cde
Panonski tetovac	3.93 cdef	Panonski tetovac	4.07 def
Slavonski žutozeleni	4.27 defg	Slavonski žutozeleni	4.40 ef
Zlatko	4.27 defg	Zlatko	4.40 ef
Biser	4.60 efg	Biser	4.47 ef
Naya Nayahit	5.33 fg	Naya Nayahit	5.07 fg
Sremac	5.47 g	Sremac	5.33 fg
Prelom	5.47 g	Prelom	6.20 g
Dvadesetica	7.27 h	Dvadesetica	7.73 h
Oplenac	8.80 i	Oplenac	8.80 h

DSI - Disease severity index (average of 15 plants)
*Values followed by different letters differ significantly (t-test; p<0.05)
IO - Indeks oboljenja (prosek od 15 biljaka)
*Vrednosti označene različitim slovima razlikuju se statistički značajno (t-test; p<0.05)

most locally grown bean genotypes to the causal agent of common blight of beans (Balaž, 1990).

Susceptibility of bean cultivars has been thoroughly investigated by a large number of authors (Stappa, 1934, as cited in Balaž, 1990; Kiryakov, 1999). However, most references on the sources of resistance and selection focused on *Pseudomonas savastanoi phaseolicola*, both in our country and worldwide (Coyne et al., 1967; Balaž, 1985, 1990). Greenhouse evaluation of the susceptibility of bean genotypes grown in Serbia has shown that most of them are susceptible to *X. campestris* pv. *phaseoli*.

DSI values were found to be very similar in both experiments. The results of our trials showed that Oreol was the most resistant cultivar, its DSI values ranging from 1.27 (I exp.) to 1.40 (II exp.). Moderate resistance was found in the genotypes: KB 101, KB 100, A 55 and Darina (Table 1) in both trials. An earlier investigation of susceptibility of some bean genotypes to *X. campestris* pv. *phaseoli* under field conditions (Balaž et al., 2005) had also shown that Oreol was resistant to *X. campestris* pv. *phaseoli*, as well as the genotypes Xan-208 and Xan-159. Balaž (1990) has also reported the resistance of Oreol to *Pseudomonas savastanoi* pv. *phaseolicola*, the causal agent of halo blight of bean in greenhouse conditions. Supporting the results of our study, Kiryakov and Genchev (2000) had also reported high to moderate resistance of the bean cultivars Oreol and HR-45 to as much as 80% of the tested strains. The authors concluded that symptom development in these cultivars depended on the bacterial strain, bean genotype, inoculum concentration and leaf age.

In our study, most genotypes (Slavonski žutozeleni, Zlatko, Biser, Sremac, Naya Nayahit) were categorized as susceptible in the first experiment, and Panonski tetovac in the second one, while the genotypes Dvadesetica, Prelom and Oplenac showed the highest susceptibility in both trials. Similar results had been acquired by Balaž et al. (2005) under field conditions.

The results of this investigation show that most bean genotypes commonly grown in Serbia are susceptible to *X. campestris* pv. *phaseoli*.

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Osetljivost genotipova pasulja na *Xanthomonas campestris* pv. *phaseoli* u zaštićenom prostoru

REZIME

Ispitivana je reakcija 17 genotipova pasulja i boranije na *X. campestris*. pv. *phaseoli*, prouzrokovača obične bakteriozne plamenjače pasulja u uslovima staklenika nakon inokulacije biljaka prskanjem bakterijskom suspenzijom (10^8 ćel/ml). Reakcija biljaka ocenjivana je na osnovu broja pega na lišću i izračunat je indeks oboljenja (IO). Proučavani genotipovi su ispoljili različit nivo osetljivosti na soj bakterije VS-1. U oba ogleda najveću otpornost je ispoljila sorta oreol. Slabo osetljivim genotipovima u prvom ogledu pripadali su genotipovi KB 142, HR-45, tisa i panonski tetovac sa IO 2,73-3,93. Isti genotipovi, osim panonskog tetovca, i u drugom ogledu su svrstani u kategoriju slabo osetljivih (IO 2,27 do 3,60). Najveći broj genotipova pripadao je kategoriji osetljivih (slavonski žutozeleni, zlatko, biser, sremac, Naya Nayahit) u prvom ogledu, a u drugom, pored navedenih genotipova, spada i panonski tetovac. Najveću osetljivost u oba ogleda ispoljile su sorte dvadesetica, prelom i oplenac.

Ključne reči: Obična plamenjača; *X. campestris* pv. *phaseoli*; genotipovi pasulja; otpornost