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Fusarium oxysporum Schl. as a Pathogen to Some Leguminous Plants

Fusarium oxysporum Schl. patogenem niektórych roślin motylkowatych

The most dangerous plant pathogens of many agricultural crops are soil-borne fungi. One of such species is *Fusarium oxysporum* Schl., which occurs in various climatic zones and infects the plants of many families (Booth, 1971; Kwaśna et al. 1991). Within this species, Booth (1971) distinguished 70 formae speciales, while Armstrong and Armstrong (1981) distinguished about 120 formae speciales and specific races which parasitize particular hosts. The Latin name of a host was used to define the names of the formae speciales of *F. oxysporum* (Snyder, Hansen, 1940; Booth, 1971; Armstrong and Armstrong, 1981). Consequently, formae speciales which parasitize pea — *F. oxysporum* Schl. f. sp. *pisi* Hans., bean — *F. oxysporum* Schl. f. sp. *phaseoli* Kend. Snyd, and soybean — *F. oxysporum* Schl. f. sp. *glycine* Amst. Amst. (Armstrong and Armstrong, 1965; Booth, 1971) were created. According to Armstrong and Armstrong (1975; 1981), some formae speciales have a number of hosts, for example *F. oxysporum* Schl. f. sp. *vasinfectum* (Ath.) Snyd. Hans. causing vascular wilt of *Gossypium hirsutum* L. is pathogenic to plants in the families of *Malvaceae, Solanaceae* and *Fabaceae.* The same authors as well as Correll (1991) stated that a formae speciales could comprise races with subtle differences in disease severity towards the host.

At the Department of Phytopathology of the Agricultural University of Lublin the fungus *F. oxysporum* was frequently isolated from plants such as

pea (*Pisum sativum* L.), runner bean (*Phaseolus coccineus* L.), soybean (*Glycine max* (L.) Merrill), lentil (*Lens esculenta* Moench.) and mung bean (*Vigna radiata* (L.) Wilczek).

The purpose of the studies was to determine pathogenicity of particular strains of *F. oxysporum* for the plants of all the above mentioned species.

MATERIALS AND METHODS

LABORATORY TRIAL

The object of the studies were culture filtrates from 50 strains of *F. oxysporum* i. e. 10 strains isolated from the internal tissues of the base of stem from each of the following: plants *P. sativum, Ph. coccineus, G. max, L. esculenta* and *V. radiata.* The strains of this fungus were obtained from plants of the first four species grown in monoculture for 6 — 7 years in the same field. Only the strains of *F. oxysporum* from *V. radiata* came from plants in the third year of cultivation. SNA liquid medium was the substrate for obtaining culture filtrates (Nirenberg, 1976). The way of obtaining culture filtrates and the performance of the experiments were the same as in the case of studies referring to *Phaseolus vulgaris* L. (Sułek-Pięta, 1983). The plants of particular species were placed in culture filtrates diluted at the proportions the concentrations of 1:2, 1:3 and 1:4 (water:filtrate) for each examined strain. Control plants were placed in to sterilized distilled water. After 24 and 48 hours of the experiment the reaction of the plants of a particular species to the presence of culture filtrates of fungus was determined.

TRIAL IN GROWTH CHAMBER

The same plant species as in laboratory tests as well as 5 strains of *F. oxysporum* (1 from each plant species) were used in a growth chamber trial. After 24 hours a clear and complete wilt symptoms of the host was observed under the influence of the culture filtrates of the above-mentioned strains irrespective of their concentration. The garden soil infested with a particular strain of *F. oxysporum* was used as a substrate for seed germination and the growth of plants of particular species. A hundred seeds of each plant species were sown into 4 pots filled with the soil infested with the experimental strain of *F. oxysporum.* The plant species were studied in combinations with each fungus strain. In that experiment the plants grew in a temperature of 20-24°C and 18-hour glow-light. After 28 days the growth and healthiness of the seedlings were determined. In order to determine the disease of the roots and stem basis of the examined plants a 5-degree scale was used (Pięta, 1983), while for the calculation of the value of pathogenicity index the formula of Mc Kinney was used (Łacicowa, 1969).

FIELD TRIAL

In this series of studies the fields of a 6-year-old monoculture of *P. sativum*, 7-year-old monoculture of *Ph. coccineus*, *G. max* and *L. esculenta* and 3-year-old monoculture of *V. radiata* were used. In each of those fields 100 seeds of all the species were sown on 4 plots (4 repetitions). During the growing season, observations were carried out which determined the number and healthiness of the plants, and after harvest also the weight of the seed yield. The manner of determining healthiness of plants and the mycological analysis were described in the earlier paper concerning *Phaseolus vulgaris* L. (Pięta, 1988).

RESULTS

CHARACTERISTIZATION OF THE STRAINS OF *FUSARIUM OXYSPORUM*

Both macro- and microscopic studies of the strains of *F. oxysporum* used to obtain culture filtrates revealed differences in the appearance of colonies and microscopic properties. Colonies with well-developed aerial mycelium and white-red colour dominated among examined cultures. The reverse of those colonies was dark red (85%). The colonies with a low, white-pink mycelium and pink-violet revers constituted 15%. Chlamydospores were formed on the mycelium of the colonies. Twenty percent of the examined strains formed microconidia and numerous macroconidia, as well. On the other hand, the remaining strains formed only single macroconidia and numerous microconidia. No relation was found between the type of the spores and the colour of the colony or its reverse.

LABORATORY TRIAL

The results obtained in laboratory trials showed that after 24 hours the plants of the studied species wilted at greater concentrations of culture filtrates of *F. oxysporum* (Tab. 1). Besides, in the combinations of the experiment at the concentrations of filtrate of 1:3 and 1:4, darkening of vascular bundles was observed in all plants of the examined species. 100% wilt took place after 48 hours after the experiment was set, regardless of the concentration of the culture filtrate and no matter which strain it came from. Besides, leaves of *L. esculenta* underwent necrosis, while in the remaining species this symptom occurred only on the leaf margin to the half of the leaf blade. Control plants showed no disease symptoms.

Tab. 1. Plants reaction to filtrate cultures of fungi after 24 hours

Filtrates	*Pisum sativum*			*Phaseolus coccineus*			*Glicine max*			*Lens esculenta*			*Vigna radiata*		
	Concentration of filtrates														
	1:2	1:3	1:4	1:2	1:3	1:4	1:2	1:3	1:4	1:2	1:3	1:4	1:2	1:3	1:4
Fusarium oxysporum from *P. sativum*	–	–	–	–	–	–	+	+	–	+	•	–	+	+	–
Fusarium oxysporum from *Ph. coccineus*	•	•	•	–	–	–	–	–	–	+	+	–	+	–	–
Fusarium oxysporum from *G. max*	+	–	–	+	–	–	–	–	–	+	•	–	+	•	–
Fusarium oxysporum from *L. esclenta*	+	–	–	•	–	–	+	•	–	–	–	–	•	•	–
Fusarium oxysporum from *V. radiata*	•	•	–	•	–	–	–	–	–	•	•	–	–	–	–
Control (sterilized water distilled)	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

– wilt, + lack of wilt, • partial wilt

TRIAL IN GROWTH CHAMBER

The plants with inhibited growth and the wilt symptoms were found in all treatment combinations in growth chamber. 28 days after the experiment was set the smallest number of plants with the greatest proportion of diseased plants, irrespective of relation to the presence of the strain of *F. oxysporum* in the substrate, were found in *V. radiata* (Fig. 1). Besides, pathogenicity indices for this species turned out to be the greatest except a combination with the strain of *F. oxysporum* isolated from *L. esculenta* (Tab. 2). The best emergencies in all the treatment combinations were characteristic of the species of *Ph. coccineus.* Nevertheless, the number of the diseased seedlings was great, ranging from 76% to 100%. The disease indices for this species were differentiated and ranged from 36 to 88 (Tab. 2).

The strains of *F. oxysporum studied* were virulent for all the plant species. It often happened that they were more dangerous for the non-host plant than for the host plant.

Tab. 2. Disease index of leguminous plants

Plant species / Experimental variant	*Pisum sativum*	*Phaseolus coccineus*	*Glycine max*	*Lens esculenta*	*Vigna radiata*
	Mean value of index				
Fusarium oxysporum from *P. sativum*	73.5^{c}	82.0^{c}	91.0^{b}	100.0^{c}	92.0^{ab}
Fusarium oxysporum from *Ph.coccineus*	56.0^{b}	77.5^{c}	49.5^{a}	87.0^{bc}	100.0^{b}
Fusarium oxysporum from *G. max*	50.5^{b}	36.0^{a}	82.5^{b}	76.5^{ab}	94.0^{b}
Fusairum oxysporum from *L.esculenta*	73.0^{c}	88.0^{c}	80.5^{b}	86.5^{bc}	76.0^{a}
Fusarium oxysporum from *V. radiata*	35.0^{a}	51.5^{b}	48.5^{a}	66.0^{a}	96.5^{b}

Mean figures differ significantly ($P \leq 0,05$), if they are not marked with the same letter

FIELD TRIAL

Six weeks after sowing, plant losses were observed on all plots, which resulted from the death of sprouts. The worst emergencies were characteristic of the species of *V. radiata* (75-87%), while the best were of *Ph. coccineus* (90-100%) (Fig. 2). Besides, all the treatments revealed plants showing inhibited growth and necrosis of roots and stem basis. In those plants at the turn of June and July, yellowing of the lower leaves was found and in the case of *P. sativum, V. radiata* and *L. esculenta* wilt and death of plants was observed. On the other hand, abscission of flowers and germ pods was observed in *Ph. coccineus* and *G. max.* There were from 55% to 87% of plants with the described disease symptoms on particular plots (Fig. 2).

As a result of a laboratory mycological analysis of the plants with the described disease symptoms, only the strains of *F. oxysporum* were obtained.

The seed yield was proportional to the number and healthiness of plants on a plot (Tab. 3). Independently of the kind of monoculture, the plants of *V. radiata* and *L. esculenta* gave a low yield, which did not differ from each other in any significant way (Tab. 3). The most differentiated mass of seed yield was obtained from the plants of *Ph. coccineus*, while on a monoculture field of that plant the yield was the smallest.The yield from the plants on a monoculture field of *G. max* (Tab. 3) was significantly higher.

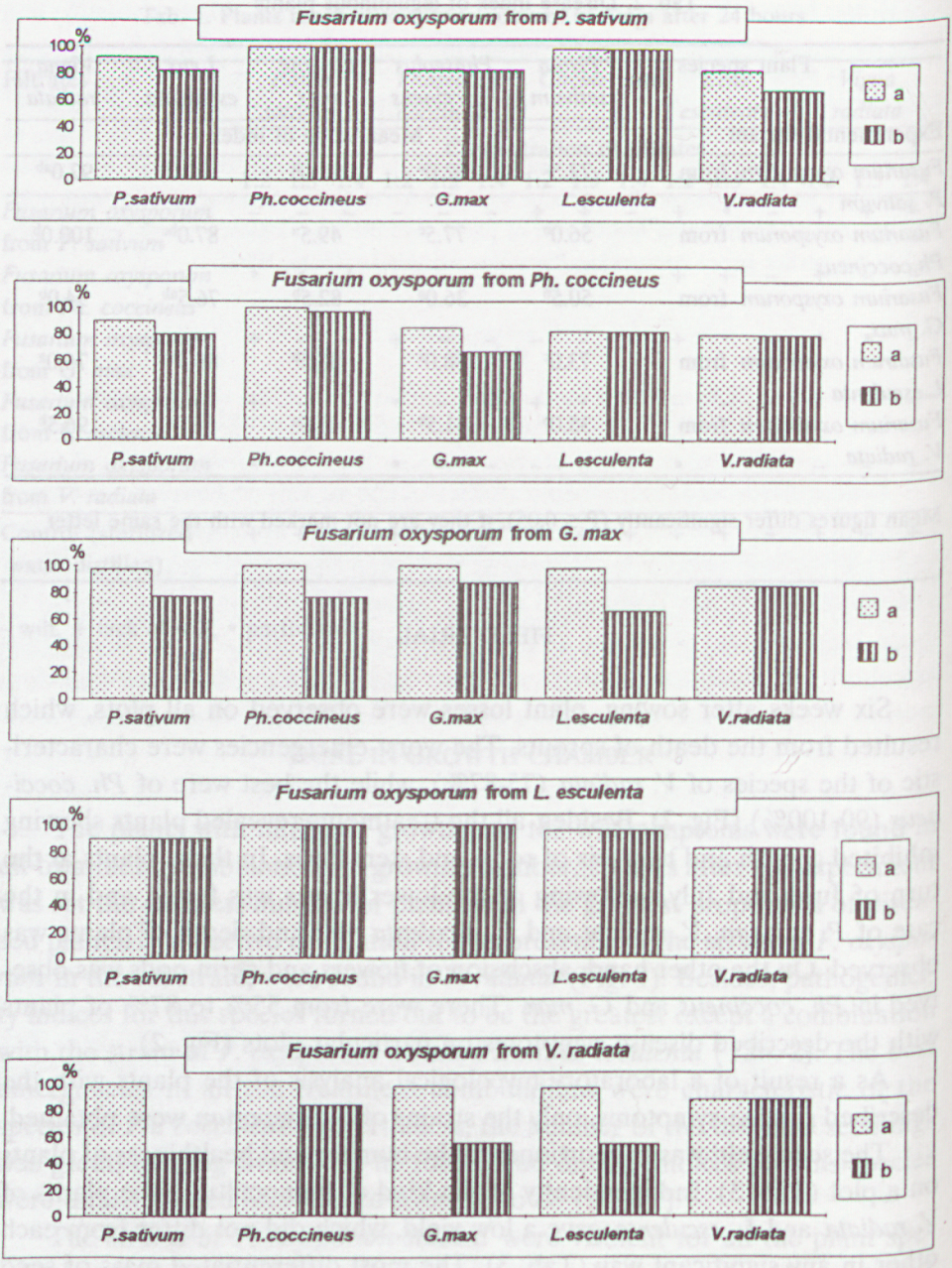

Fig. 1. The emergence (a) and proportion of diseased soybean seedlings (b) in chamber trial

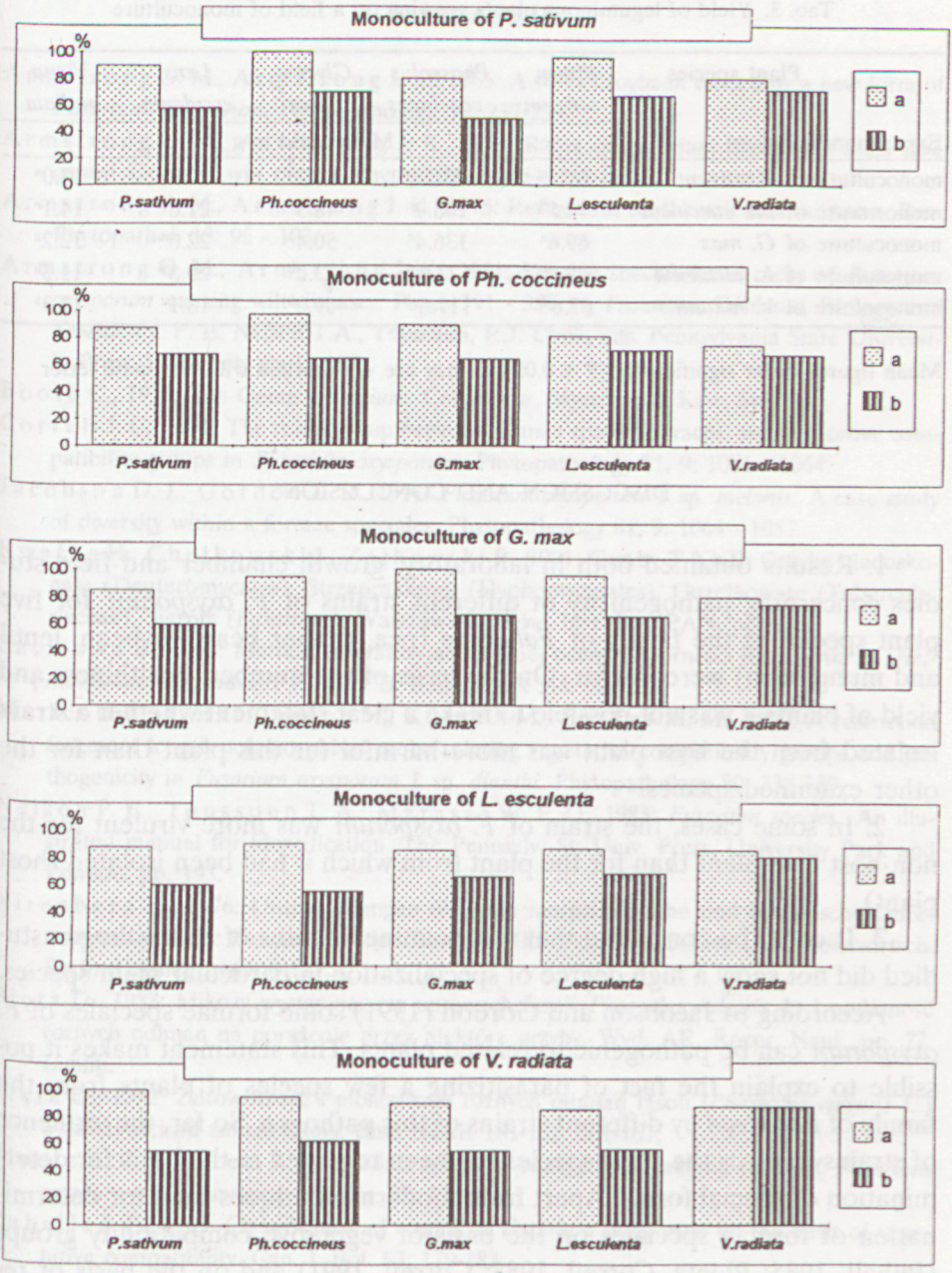

Fig. 2. The emergence (a) and proportion of diseased soybean plants (b) in field trial

Tab. 3. Yield of leguminous plants growing on a field of monoculture

Plant species / Experimental variant	*Pisum sativum*	*Phaseolus coccineus*	*Glycine max*	*Lens esculenta*	*Vigna radiata*
	Mean yield in g				
monoculture of *P.sativum*	61.5[a]	122.3[abc]	54.7[a]	19.9[a]	12.0[a]
monoculture of *Ph. coccineus*	75.5[b]	108.0[a]	48.3[a]	21.0[a]	14.1[a]
monoculture of *G. max*	69.6[a]	136.4[c]	50.4[a]	22.0[a]	12.2[a]
monoculture of *L. esculenta*	79.8[b]	127.0[bc]	73.2[b]	20.3[a]	11.5[a]
monoculture of *V. radiata*	67.6[ab]	117.6[ab]	69.0[b]	18.1[a]	11.3[a]

Mean figures differ significantly ($P \leq 0.05$), if they are not marked with the same letter

DISCUSSION AND CONCLUSIONS

1. Results obtained both in laboratory, growth chamber and field studies concerning pathogenicity of different strains of *F. oxysporum* for five plant species of the family of *Fabaceae* (pea, runner bean, soybean, lentil and mung bean) were similar. On the basis of the number, healthiness and yield of plants it was not possible to make a clear statement whether a strain isolated from the host plant was more harmful for this plant than for the other examined species.

2. In some cases, the strain of *F. oxysporum* was more virulent for the non-host host plant than for the plant from which it had been isolated (host plant).

3. It could be concluded that the examined strains of the pathogen studied did not show a high degree of specialization in particular plant species.

According to Jacobson and Gordon (1991) some formae speciales of *F. oxysporum* can be pathogenic to related plants. This statement makes it possible to explain the fact of parasitizing a few species of plants from the family of *Fabaceae* by different strains of this pathogen. So far, the virulence of strains towards the plant species has been regarded as the basis for determination of special forms. Apart from modern techniques used for determination of formae speciales on the basis of vegetative compatibility groups (Puhall, 1985; Ploetz, Correll, 1988; Correll, 1991) and on the basis of restriction fragment length polymorphism (Manicom et al. 1990; Correll, 1991; Jacobson, Gordon, 1991), the studies should also take into consideration in vitro and in vivo analyses on pathogenicity of those strains.

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STRESZCZENIE

Do badań użyto różne izolaty *Fusarium oxysporum* w celu określenia ich chorobotwórczości względem roślin *Pisum sativum, Phaseolus coccineus, Glycine max, Lens esculenta* i *Vigna radiata.* Na podstawie uzyskanych wyników doświadczeń, laboratoryjnego, fitotronowego i polowego, można stwierdzić, że badane izolaty powodowały objawy chorobowe u roślin wszystkich gatunków, lecz nie wykazywały wysokiego stopnia wyspecjalizowania do porażania rośliny-gospodarza. Spośród badanych gatunków roślin najbardziej podatne na porażenie przez testowane izolaty *F. oxysporum* okazały się rośliny *Vigna radiata,* a najmniej podatne *Phaseolus coccineus.*