

Reaction of Pea Selections to *Fusarium oxysporum* f.sp. *pisi* (Races 1, 2, 5, 6) and *Fusarium solani*

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Abstract

Forma specialis (f.sp.) *pisi* of *Fusarium oxysporum* infestates pea but on other crop does not cause. Physiological races of this fungus (races 1, 2, 3 and 6) are tested on differentiation selections of pea. In the mixture with *Fusarium solani* there was better differentiated the reaction of single races. Own method is based on the artificial infection of the pea with the mixture of conidia and mycelial fragments of the race of *Fusarium oxysporum* f.sp. *pisi* and *Fusarium solani*. The roots of germinating pea are partially shortened on the top and poured with the suspension of fungi. After the germinating the infected pea is placed into Perlit (the substance for the cultivation). The pea was watered after that. During 3 years of our work we tested namely many new selections from the breeding station in Lužany. It seems that some of them should be used for new selection of really resistant varieties. In the year 1999 there were new selection relatively resistant: 595/32, 682/37, 238/847, in the year 2000 no resistant selections were gained and in the year 2001 there were like resistant evaluated these selections: 633/1409 and 1456/1919.

Keywords: *Fusarium oxysporum* f.sp. *pisi*; *Fusarium solani*; physiological races; varieties of pea; resistance test; evaluation of resistance

INTRODUCTION

Basic question is how evaluate the resistance of single pea plants to *Fusarium oxysporum* f.sp. *pisi*:

1. like the resistance of all plant,
2. like the partial resistance (the average of all partial resistances – the root, the stem, root collar, leaves, nodes, the bloom, shoot apex etc.).

All infections of pathogen agents are specific to different plant organ. Therefore *Fusarium oxysporum* f.sp. *pisi* cause namely the damage in the tracheary elements (vessels) and tracheomycosis. Basic moment of the infection is settling down the tracheary elements. Following moment is the infestation of surface tissues of plants and the ability to invade the plants of pea. From the point of view of the surviving on plant residues is this fungus saprophytic but before the infestation of the pea with high invasivity it is polyfageous pathogenic fungus. But it depends also on the virulence of single races. This fungus is studied many years in many countries of the world. The ability of this fungus to adaptate on the inner factors

described BROWN (1926). The conclusions of this paper was confirmed by other authors like ARMSTRONG and ARMSTRONG (1968) and VALÁŠKOVÁ (1975). The confirmation and maintaining of the pathogen is possible by the comparing of monosporic isolate and the isolate from ill plant (KRAFT & HAGLUND 1978). Some authors that higher temperature during the infestation increased the activity of biochemical reaction (SCHROEDER & WALKER 1942; KRÁTKÁ & KOVÁČIKOVÁ 1981). This temperature was 25–32°C. *Fusarium oxysporum* f.sp. *pisi* causes the vascular wilt on pea and for the objectivisation of this conclusion they were proved many methods. The basic method is the soaking in spore suspension (ARMSTRONG & ARMSTRONG 1948). The study of vascular wilt begun in 1928 when vascular wilt was described on pea in Wisconsin (LINFORT 1928). This race was signified as the race 1. The race which caused near wilt described by SNYDER and WALKER (1935) was signified as the race 2. SCHREUDER (1951) discovered next race for Holland, causing the wilt of cultivars Delwiche Com-mando and New Era which were resistant to the races

1 and 2. This race was indicated like race 3 (BUXTON 1957). BOLTON *et al.* (1966) described the new race 4 in Canada. It caused the wilt of varieties: WR Perfection, New Era and Wisconsin C 183, resistant to races 1 and 2, but it was not pathogenic for the variety of New Wales. The race 5 was described by HAGLUND and KRAFT (1978). All varieties resistant to races 1 and 2 were susceptible to this race. ARMSTRONG and ARMSTRONG (1974) described following 6 races of *Fusarium oxysporum* f.sp. *pisi*: race 6 which caused the wilt of the variety New Era, but the variety New Wales was resistant. The race 7 was described as resistant for varieties WR Alaska and Double One. The race 8 was pathogenic to varieties New Wales, WR Lincoln and Rovar, the cultivars Carter's Daisy and New Era were resistant. Next race 9 was pathogenic to cultivars Carter's Daisy, WR Greenfeast and New Wales, nepathogenic for New Era. The race 10 was pathogenic for the varieties Carter's Daisy and New Wales, but non-pathogenic for WR Greanfeast and New Era. For the race 11 the variety WR Alaska was susceptible, the varieties WR Lincoln, New Era and New Wales were resistant.

Fusarium wilt and near wilt are two major wilt diseases caused by the fungus *Fusarium oxysporum* f.sp. *pisi* (different races). The fungus can be introduced with seed. Once present, this fungus persists indefinitely in soil. *Fusarium* wilt often detected by appearance by yellowing of lower leaves and stunting of plants. Leaflet margins curl downward and inward. The stem may be slightly swollen and brittle near the soil. Internal woody stem tissue often is discolored, turning lemon brown to orange brown. Externally, the root system appears healthy, however, secondary root rots are likely to occur on plants wilted for long periods. Eventually, wilted plants may die. Near-wilt symptoms are similar to *Fusarium* wilt symptoms. However, near wilt plants die more slowly than do *Fusarium* wilt plants. Also, internal woody stem tissue usually is brick-red in near wilt plants. This discoloration extends throughout the plant.

MATERIAL AND METHODS

Methods of isolation. The isolates of pathogenic fungi were gained from infested plants. The plants were cleaned and washed, the root was divided from plant in the place of root collar. From the place where the symptoms were observed like discoloration of leading tissues were taken the segments of 5–10 mm which were disinfected by 5% of natrium chlorine (the preparation Savo diluted with distilled water).

The segments were dried and removed from cuticular tissues and the rests were placed on the surface of agar medium (potatoe agar) or malt agar with bengal red.

Cultivation. The segments on the media on Petri dishes were cultivated 5–6 days before the temperature 26°C, the hyphae growing out of the segment were transferred on the Petri dishes with potatoe agar and malt agar (pH 5.8) and they were cultivated 6 days before 26°C. Growing hyphae were studied under the microscope and determined by the morphological characteristics of micro- and macroconidia, the creation of chlamydospores and other systematic signs.

Laboratory method for testing of resistance. Test-tube method with liquid substrate was replaced on the method with solid substrate (KOVÁČIKOVÁ & KŮDELA 1987). The seeds of pea was superficially disinfected with natrium chlorate (5% of the preparation Savo). They were washed and let to germinate. After the creation of the roots, the top of the roots was removed and submerged into spore solution. The plants were aseptically inserted into test-tubes with the agar.

Methods of the evaluation. The inoculated and control plants was evaluated after 14 days after the infection. About every single plant the symptoms on upper part were evaluated and the symptoms on the roots. After the intensity of the infestation we distinguished 4 degrees:

- 0 – without symptoms
- 1 – the beginning of the browning in the roots, weak braking of the growth, little leaves, yellowing of the leaves
- 2 – more distinct symptoms like in the degree 1
- 3 – plants died.

Mean number of plant infestation was stated after the formula:

$$\text{MNPI} = \frac{n_1 \cdot G_1 + n_2 \cdot G_2 + n_3 \cdot G_3 + n_4 \cdot G_4}{N}$$

where: n_1, n_2, n_3, n_4 – number of the plants with the symptoms in single groups

G_1 – G_4 – number of plants in single groups

N – total number of plants

The criteria of the evaluation. After mean number of plant infestation (MNPI) the tested group were divided into 4 degrees:

		MNPI
R	resistant	0–0.49
MR	middle resistant	0.5–0.99
MS	middle susceptible	1.0–1.99
S	susceptible	2.0–3.00

After the change of weight of inoculated plants in the comparison with non-inoculated plants the plants were divided into 6 groups (% of the control = 100):

strong stimulation	126 and more
weak stimulation	106–125
no stimulation	91–105
weak inhibition	81–90
middle inhibition	71–80
strong inhibition	70 and less

RESULTS AND DISCUSSION

The new selections were evaluated on the infestation of *Fusarium solani* f.sp. *lisi*. The results are gathered in Tables 1 and 2.

From our tables there is visible that the infections in controlled conditions (by the control) are in single

years different. The interaction with *Fusarium solani* is evident. The symptoms in our climatic conditions occurred very rare. Into the vegetative top the fungus is widespread only before warm and sunny weather, before rain weather the atypical symptoms are developed (BUXTON & STOREY 1954). During vegetative development (parasexual recombination and adaptation on the environment) the variability of the fungus is reached. Parasexual recombination creates new populations which are different from original races (BUXTON 1957, 1960).

The adaptation of pathogenic fungus should effect on the pathogenicity of isolates. After the results from Tables 1 and 2 we see that our new selections are only partially resistant, they are to some races resistant, to other ones are susceptible. The differences between the reactions during different years should be influenced by the changes in the virulence of the isolate during its storage in culture collections.

Table 1. The susceptibility of the new selections of pea on different races of *Fusarium oxysporum* f.sp. *lisi* in 1999 and 2000

Selection/variety	Race I	Race II	Race V	Race VI	Selection/variety	Race I	Race II	Race V	Race VI
	+ <i>F. solani</i>	+ <i>F. solani</i>	+ <i>F. solani</i>	+ <i>F. solani</i>		+ <i>F. solani</i>	+ <i>F. solani</i>	+ <i>F. solani</i>	+ <i>F. solani</i>
2000					2001				
172/26	MR	MR	MS	MS	1604/34	R	R	MS	S
595/32	MR	MR	R	R	690/39	MR	R	MS	MS
855/34	MR	MR	MS	R	1740/40	R	MR	MS	MS
1343/1220	MR	MR	MS	MS	2026/505	S	MR	MS	MS
682/37	MR	MR	MS	R	1760/937	MR	MR	MS	S
359/820	MR	MS	R	R	2060/505	S	MS	S	S
238/847	MR	MR	MS	R	1340/672	S	MR	S	MS
378/906	MR	MS	MS	MS	1552/755	MS	MR	MR	MS
874/937	MR	MS	MS	MS	1529/905	MS	MR	MS	MS
1386/1222	MR	MR	S	S	472/946	S	MS	MS	MS
1790/1234	MR	MS	S	S	278/1066	MS	MS	MR	MR
1506/1268	MR	MR	S	S	356/1081	MR	MS	MR	MS
2563/2031	MR	MR	S	MR	1202/1220	MS	MS	MR	MS
P13 I-92084.10	MR	MR	S	S	778/1660	MR	MS	R	R
S-4582	MR	MR	S	S	59/1951	S	MS	MR	MS
C 7726	MR	MR	S	MR	S 61094	MS	R	MR	MS
Adept K	MR	MR	S	MR	Menhir K	S	MR	MS	MR
Menhir K	MR	MR	S	S	Adept K	R	MS	S	MR

R – resistant; S – susceptible; MR – middle resistant; MS – middle susceptible

Table 2. The susceptibility of the new selections of pea on different races of *Fusarium oxysporum* f.sp. *pisi* in 2001

Selection/ variety	Race I + <i>F.</i> <i>solani</i>	Race II + <i>F.</i> <i>solani</i>	Race V + <i>F.</i> <i>solani</i>	Race VI + <i>F.</i> <i>solani</i>
558/33	R	R	MS	MS
1560/39	MR	R	S	MS
633/1409	R	MR	MS	MR
2728/1919	S	MR	MS	MR
1456/1919	MR	MR	MS	MR
59/1951	S	S	S	MS
246/1991	S	MR	MS	MS
957/2028	MS	MR	MS	MS
258/2179	MS	MR	MS	MR
336/2182	S	MS	MS	–
929/2213	MS	MS	MS	MS
S-6207	MR	MR	MR	MS
S-7206.6	MS	MS	R	–
Adept K	R	MS	S	MR
Menhir K	S	MR	R	MS

References

- ARMSTRONG G.M., ARMSTRONG J.K. (1948): Nonsusceptible hosts as carriers of wilt fusaria. *Phytopathology*, **38**: 808–826.
- ARMSTRONG G.M., ARMSTRONG J.K. (1968): Formae specialis and races of *Fusarium oxysporum* causing a tracheomycosis in the syndrome of disease. *Phytopathology*, **58**: 1242–1246.
- ARMSTRONG G.M., ARMSTRONG J.K. (1974): Races of *Fusarium oxysporum* f.sp. *pisi*, causal agent of wilt of pea. *Phytopathology*, **64**: 849–857.
- BOLTON A.T., NUTTALL V.W., LYALL L.H. (1966): A new race of *Fusarium oxysporum* f.sp. *pisi*. *Can. J. Plant Sci.*, **46**: 343–347.
- BUXTON E.W. (1957): Some effects of pea root exsudates on *Fusarium oxysporum* Fr. f.sp. *pisi* (Linf.) Snyder et Hansen. *Trans. Br. Mycol. Soc.*, **40**: 145–151.
- BUXTON E.W. (1960): Heterokaryosis, saltation and adaptation. In: HORSFALL J.G., DIMOND A.E.: *Plant Pathology, an Advanced Treatise*, 2. Acad. Press, New York: 259–405.
- BUXTON E.W., STOREY I.T. (1954): The occurrence of pea wilt in Britain. *Plant Path.*, **3**: 13–16.
- HAGLUND W.A., KRAFT J.M. (1979): *Fusarium oxysporum* f.sp. *pisi*, race 6: occurrence and distribution. *Phytopathology*, **69**: 818–820.
- Internet (2002): Peas: *Fusarium* Wilt and Near Wilt.
- KRAFT J.M., HAGLUND W.A. (1978): A reappraisal of the race classification of *Fusarium oxysporum* f.sp. *pisi*. *Phytopathology*, **68**: 273–275.
- KRÁTKÁ J., KOVÁČIKOVÁ E. (1981): Pathogenicity of *Fusarium oxysporum* f.sp. *pisi* at various temperatures as related to biochemical processes in peas. *Zbl. Bakt. II Abt.*, **136**: 648–655.
- KOVÁČIKOVÁ E., KŮDELA V. (1987): Metody testování jetelovin vůči patogenům způsobujícím choroby kořenového krčku a kořene rostlin. VÚRV, Praha.
- SCHREUDER J.C. (1951): Een onderzoek over de Amerikaanse vaatziekte van de erwten in Nederland. *Tijdschr. Plantenziekten*, **57**: 175–206. (English summary.)
- SCHROEDER W.T., WALKER J.C. (1942): Influence of garden pea to *Fusarium* wilt. *J. Agric. Res.*, **65**: 221–248.
- SNYDER J.C., WALKER J.C. (1935): *Fusarium* near – wilt of pea. *Zbl. Bakt. Parasitenk., Abt. II*, **91**: 355–378.
- VALÁŠKOVÁ E. (1975): Studium patogenních hub rodu *Fusarium*. [Závěrečná zpráva.] ÚZLK, Praha.