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Pathogenicity of fungi colonising the soil after the cultivation of cover crops towards the seedlings of salsify *Tragopogon porrifolius* var. *sativus* (Gaterau) Br.

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ABSTRACT

The purpose of the present study was to conduct pathogenicity tests for salsify seedlings in a soil environment colonised by selected fungi species, after the cultivation of cover crops such as oats, common vetch and tancy phacelia. A growth chamber experiment used the seeds of 'Mamut' salsify and soil overgrown with *Altenaria alternata, Fusarium culmorum, F. oxysporum, Pythium* *irregulare, Rhizoctonia solani* and *Sclerotinia sclerotiorum.* The pathogenicity of these microorganisms towards salsify seedlings was established on the basis of the grown plants, their health and a disease index calculated on the basis of a five-degree scale. The examined isolates considerably weakened the emergence and health of salsify. Based on pathogenicity tests, such species as *R. solani, S. sclerotiorum* and *P. irregulare* were considered as the major cause of seedling necrosis and root rot of the plants of the *Tragopogon porrifolius* var. *sativus.* Among the studied soil-borne plant pathogens, the most pathogenic ones towards salsify seedlings proved to be the isolates of *R. solani, S. sclerotiorum* and *P. irregulare*, whereas the least harmful included *A. alternata* and *F. culmorum.*

INTRODUCTION

Salsify (*Tragopogon porrifolius* var. *sativus*), also called Oyster Plant, is a vegetable closely related to scorzonera. It comes from the Mediterranean area, where it was known as early as ancient times. In Europe it gained the greatest popularity in the 15th and 16th centuries, while in Poland it is still a vegetable that is little known and rarely cultivated. It belongs to the family of *Asteraceae* and is an annual or biennial plant (Hardenburg et al. 1986, Muller-Lemans 1991). The edible part of this plant is the yellowish or grayish thin taproot with white flesh and a taste similar to that of parsley root (Mencarelli 2007). Salsify is a valuable vegetable, considering both its taste and nutritious value. It contains large quantities of calcium, phosphorus, iron, carotene, vitamins (B_1, B_2, PP and C) as well as inulin – a glycoside that has a positive effect on human and animal organisms (Kierstan 1978, Mencarelli 2007). Inulin reduces the content of fats and the energetic value of food, at the same time improving the content of intestinal bacteria (Teeuwen et al. 1992, Lutomski 2001).

Due to the rather rare cultivation of salsify plants, the literature provides only scarce information concerning pathogens dangerous to this plant. As reported by Dellavalle et al. (1999), TSWV (Tomato spotted wilt virus) plays a considerable role in infecting the aboveground organs of salsify. In addition, the health of the aboveground parts of this vegetable is decreased by such fungi as *Albugo tragopogonis*, causing the blight of salsify (Mencarelli 2007), *Erysiphe cichoracearum*, causing powdery mildew (Namesny-Vellespir 1996), and *Sclerotinia sclerotiorum*, causing sclerotinia rot (Biller and Draper 2001). On the other hand, there is no information on the role of soil-borne fungi in infecting the underground parts of salsify. It is known that fungi colonizing the soil environment may constitute a considerable danger to different cultivated plants, including salsify, throughout the period of vegetation. These fungi as facultative pathogens inhibit the germination of seeds and cause the necrosis of seedlings and the root rot of older plants. Cover crops used for soil mulching may have an inhibiting effect on the formation of soil pathogen communities (Patkowska and Konopiński, unpublished data). Studies conducted earlier (Patkowska and Konopiński 2008) revealed that using oats, common vetch and tancy phacelia as cover crops considerably reduced the population of fungi pathogenic towards scorzonera.

The purpose of the present study was to conduct pathogenicity tests for salsify seedlings referring to different microorganism species colonizing the soil environment of this vegetable after the cultivation of cover crops.

MATERIAL AND METHODS

The experiment on the pathogenicity of soil-borne microorganisms towards salsify seedlings was conducted in a growth chamber. The seeds of 'Mamut' cultivar and soil overgrown with soil-borne plant pathogens, such as *Altenaria alternata, Fusarium culmorum, F. oxysporum, Pythium irregulare, Rhizoctonia solani* and *Sclerotinia sclerotiorum,* were used to establish the experiment. Those isolates came from the microbiological analysis of the soil conducted according to the method described by Martyniuk et al. (1991). The soil for the analysis was taken from the plough layer of the field where sasify was cultivated in the second 10-day period of June. The field experiment took into consideration soil mulching with intercrop cover crops such as oats, common vetch and tancy phacelia, which were ploughed over in autumn or in spring. A conventional cultivation of salsify, i.e. without any cover crops, was the control (Table 1).

The growth chamber experiment considered five randomly chosen isolates of each species of microorganisms, which were given the numbers from 1 to 5. Salsify seeds disinfected on the surface were sown into pots filled with soil overgrown with the mycelium of a single isolate of a given species. The control included salsify seedlings grown out of the seeds sown into sterile soil. 100 seeds were taken into consideration for each isolate and for the control. The infection mixture of plant pathogens used in the experiment was prepared according to Noll's method described in an earlier paper (Patkowska and Konopiński 2008). The conditions for seed germination and salsify growth in a growth chamber were similar to those described in an earlier experiment by Patkowska and Konopiński (2008). The temperature of the day, which lasted 16 hours, was 22-24°C (with irradiance of 27 μmol m^{-2} s^{-1}) and the temperature at night was 18-20°C. The relative humidity of the air ranged from 70 to 85%.

Four weeks after the seed sowing, the number of seedlings was determined and the percentage of plants with disease symptoms on the roots was established. The healthiness of the plants was estimated according to the five-degree scale given earlier for the seedlings of scorzonera: 0° – no disease symptoms, 1° – necrosis up to 10% of the root surface, 2° – necrosis up to 25% of the root surface, 3° – necrosis up to 50% of the root surface, 4° – necrosis over 50% of the root surface (Patkowska and Konopiński 2008). The disease index was calculated according to McKinney's formula provided by Łacicowa (1969) and Patkowska and Konopiński (2008). Next, the plant material was submitted to a mycological analysis according to Koch's postulates.

The obtained results were statistically analyzed, and the significance of differences was established on the basis of Tukey's confidence intervals (Oktaba 1987).

RESULTS AND DISCUSSION

The microbiological analysis of the soil taken from the plough layer of particular combinations of the field experiment pointed to differentiated numbers of plant pathogens whose isolates were used in pathogenicity tests in the conditions of a growth chamber (Table 1). *Fusarium oxysporum* and *Rhizoctonia solani* proved to be most frequently isolated, and their mean proportion was 20.7% (Fig. 1). *Fusarium culmorum* and *Pythium irregulare* had the smallest proportions among the tested microorganisms (respectively, 11.0% and 14.1%). The enumerated species are known for their ability to survive in the soil and for their polyphagous character, and hence, for being dangerous towards many vegetable species. As reported by Pięta and Kęsik (2006), those plant pathogens, and especially *F. culmorum,* constituted a serious threat towards onion plants. Studies conducted by Mazur et al. (2004) and Nawrocki (2005) also pointed to their harmfulness towards carrots and parsley. Blancard et al. (2005), on the other hand, provided information on the pathogenic character of the enumerated plant pathogens towards leaf chicory and endives.

Table 1. Microorganisms isolated from the soil

Fungus species	Experimental combination / Number of isolates							Total number
	1*	2	3	4	5	6	7	
Alternaria alternata (Fr.) Keissler	2	2	4	4	7	10	12	41
Fusarium culmorum (W.G.Sm.) Sacc.	1	1	2	3	4	5	9	25
Fusarium oxysporum Schl.	2	2	5	7	8	9	14	47
Pythium irregulare Baisman	1	2	3	4	5	6	11	32
Rhizoctonia solani Kühn	2	4	5	5	8	10	13	47
Sclerotinia sclerotiorum Lib. de Bary.	1	2	4	5	7	7	9	35
Total	9	13	23	28	39	47	68	227

*1 – oats mulch + spring ploughing, 2 – oats mulch + pre-winter ploughing, 3 – spring vetch mulch + spring ploughing, 4 – spring vetch mulch + pre-winter ploughing, 5 – tancy phacelia mulch + spring ploughing, 6 – tancy phacelia mulch + pre-winter ploughing, 7 – conventional cultivation

Fig. 2. The necrosis on roots of salsify seedlings (Phot. E. Patkowska)

The growth chamber experiment made it possible to determine the degree of pathogenicity of particular isolates of the studied microorganism species for salsify seedlings. This pathogenicity was established on the basis of the number of grown plants and their health (Tables 2 and 3). The poorest emergence was observed after inoculating the soil with the isolates of *P. irregulare, R. solani* and *S. sclerotiorum* (on average, 67.1, 60.1 and 61.6 seedlings). The best emergence occurred in the combinations with *A. alternata* and *F. culmorum* as respectively, from 72.5 to 97.0 and from 79.5 to 97.0 seedlings grew from the seeds, depending on the studied fungus isolate (Table 2). Slightly worse emergence of salsify was obtained after inoculating the soil with the species of *F. oxysporum* (on average, from 72.0 to 88.5 seedlings, depending on the studied fungus isolate). Earlier studies (Patkowska and Konopiński 2008) discovered that *P. irregulare* contributed to considerably worse emergence of scorzonera, and inoculation of the soil with the isolates of *S. sclerotiorum* caused the worst emergence of root chicory (Patkowska and Konopiński 2008, unpublished data). According to Biller and Draper (2001), fungus *S. sclerotiorum* also seriously threatened the field cultivation of salsify. On the other hand, the best emergence of scorzonera were also obtained after inoculating the soil with the isolates of *A. alternata* (Patkowska and Konopiński 2008).

Table 2. Mean number of salsify seedlings that emerged in particular experiment combinations

Isolate number	Pathogen species					
	A. alternata	*F. culmorum*	*F. oxysporum*	*P. irregulare*	*R. solani*	*S. sclerotiorum*
1	97.0	88.0	88.5	74.5	72.5	67.5
2	79.5	97.0	76.0	59.5	63.0	49.0
3	91.0	79.5	81.0	64.0	55.0	66.0
4	84.5	93.5	69.5	75.5	60.0	57.0
5	72.5	80.0	72.0	62.0	50.0	68.5
Mean	84.9	87.6	77.4	67.1	60.1	61.6
Control	100	100	100	100	100	100
$LSD_{0.05}$	9.87	7.49	8.91	9.17	10.93	12.01

Table 3. Participation of infected salsify seedlings in particular experiment combinations (%)

Isolate number	Pathogen species					
	A. alternata	*F. culmorum*	*F. oxysporum*	*P. irregulare*	*R. solani*	*S. sclerotiorum*
1	15.0	26.0	32.0	51.0	55.0	49.5
2	10.5	19.5	26.5	43.5	49.5	37.5
3	21.0	23.0	40.0	38.5	43.5	54.0
4	16.5	15.5	34.5	54.0	51.5	44.0
5	24.5	31.0	25.0	47.0	42.0	50.5
Mean	17.5	23.0	31.6	46.8	48.3	47.1
Control	0	0	0	0	0	0
$LSD_{0.05}$	4.51	5.01	6.32	7.86	4.76	4.63

Table 4. Values of the disease index of salsify seedlings

Isolate number	Pathogen species					
	A. alternata	*F. culmorum*	*F. oxysporum*	*P. irregulare*	*R. solani*	*S. sclerotiorum*
1	5.8	11.4	16.4	24.5	26.5	30.3
2	2.7	10.0	12.2	19.4	35.0	36.0
3	8.5	9.5	20.8	29.6	40.2	25.2
4	4.2	6.6	16.4	32.8	37.1	39.4
5	6.5	12.4	21.6	34.4	34.0	24.8
Mean	5.5	9.9	17.5	28.1	34.5	31.1
Control	0	0	0	0	0	0
$LSD_{0.05}$	1.89	3.22	2.01	3.57	4.31	4.36

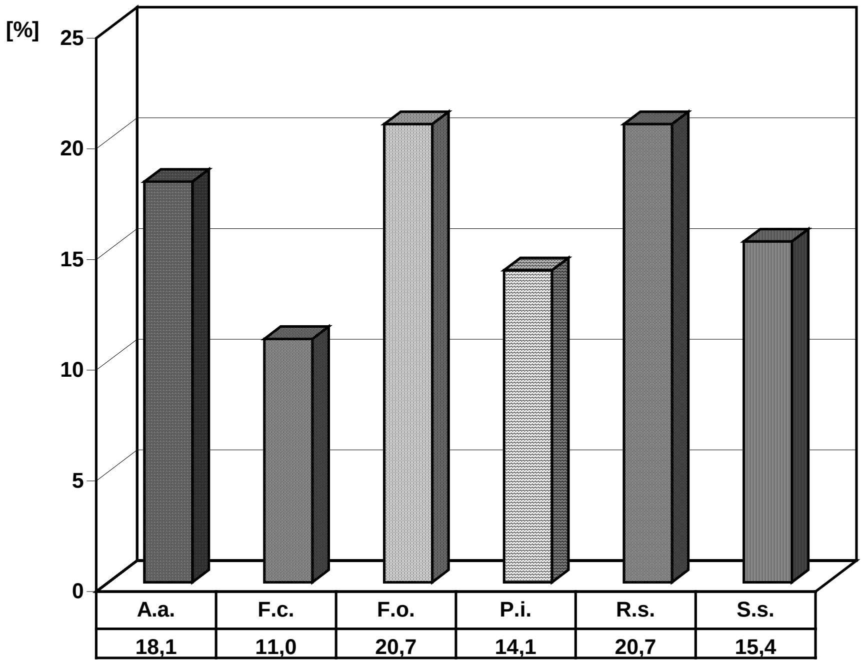

Fig. 1. Participation of selected phytopathogens isolated from the soil A.a. – *A. alternata*, F.c. – *F. culmorum*, F.o. – *F. oxysporum,* P.i. – *P. irregulare*, R.s. – *R. solani*, S.s. – *S. sclerotiorum*

Particular experimental combinations included the seedlings with inhibited growth, with necrotic symptoms on the roots (Fig. 2). The number of diseased seedlings was differentiated and found to be related to the studied isolate within a given plant pathogen species. The greatest number of diseased seedlings were obtained in combinations with *R. solani, S. sclerotiorum* and *P. irregulare,* as their proportion was, respectively, 48.3%, 47.1% and 46.8%, on average (Table 3). The smallest number of diseased salsify seedlings grew from the seeds sown into the soil infected with *A. alternata*, and their proportion ranged from 10.5% to 24.5%, depending on the examined isolate. The germinating salsify seeds and seedling roots were infected by the isolates of *F. culmorum* and *F. oxysporum* in a slightly greater degree. The proportion of diseased seedlings in those combinations of the experiment ranged, respectively, from 15.5 to 31.0% and from 25.0 to 40.0% (Table 3). In the control combination, healthy seedlings were obtained from all salsify seeds. As reported by Plentinger and Lamers (2000), infection of leaf chicory by *S. sclerotiorum, Pythium* sp. and *Phytophthora* sp. considerably decreased the health of this plant. According to Pięta and Kęsik (2006), fungi from the *Fusarium* genus also proved pathogenic towards onion seedlings. On the other hand, the isolates of *P. irregulare* proved to be the most pathogenic towards scorzonera seedlings, while *A. alternata* was the least pathogenic (Patkowska and Konopiński 2008).

The value of the disease index for salsify seedlings, calculated on the basis of a five-degree scale, ranged from 2.7 to 40.2 in particular experimental combinations (Table 4). The smallest value of the disease index was characteristic of the examined seedlings grown in the combinations with *A. alternata* or *F. culmorum* (respectively, from 2.7 to 8.5 and from 6.6 to 12.4, depending on the examined isolate). Slightly higher values of the index of seedling infection were found after inoculating the soil with the isolates of *F. oxysporum* (from 12.2 to 21.6). On the other hand, the highest index of salsify seedling infection was observed in combinations with *R. solani, S. sclerotiorum* and *P. irregulare*, i.e. 34.5, 31.1 and 28.1, respectively (Table 4). Studies by Biller and Draper (2001) also pointed to the considerable pathogenicity of *S. sclerotiorum* towards salsify plants. On the other hand, Mazur et al. (2004) informed about the pathogenic character of *Altenaria* spp. towards carrots, parsley and celery. In addition, Nawrocki (2005) discovered a considerable pathogenicity of *F. oxysporum, P. irregulare* and *R. solani* towards the seedlings of parsley cultivated in field or greenhouse conditions.

Reisolation from the infected tissues of salsify carried out according to Koch's postulates confirmed the colonization of those plants by *Altenaria alternata, Fusarium culmorum, F. oxysporum, Pythium irregulare, Rhizoctonia solani* and *Sclerotinia sclerotiorum* with the morphological features identical to those of isolates used in inoculation.

CONCLUSIONS

1. It was discovered that soil-borne plant pathogens constitute a significant threat towards the seedlings and older plants of salsifies, as they considerably weaken the emergence and health of this plant.
2. Such species as *Rhizoctonia solani, Sclerotinia sclerotiorum* and *Pythium irregulare* could be considered the main cause of seedling blight and root rot of *Tragopogon porrifolius* var. *sativus.*
3. Among the studied species of soil microorganisms, the isolates of *R. solani, S. sclerotiorum* and *P. irregulare* proved to be the most pathogenic towards salsify seedlings, whereas *Altenaria alternata* and *Fusarium culmorum* were the least harmful.

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PATOGENICZNOŚĆ GRZYBÓW ZASIEDLAJĄCYCH GLEBĘ PO UPRAWIE ROŚLIN OKRYWOWYCH DLA SIEWEK SALSEFII *TRAGOPOGON PORRIFOLIUS* VAR. *SATIVUS* (GATERAU) BR.

Streszczenie: Celem prezentowanych badań było przeprowadzenie testów patogeniczności dla siewek salsefii wybranych gatunków grzybów zasiedlających środowisko glebowe tego warzywa po uprawie roślin okrywowych, takich jak: owies, wyka siewna i facelia. W doświadczeniu fitotronowym wykorzystano nasiona salsefii odm. 'Mamut' oraz ziemię ogrodową, przerośniętą izolatami *Alternaria alternata, Fusarium culmorum, F. oxysporum, Pythium irregulare, Rhizoctonia solani* i *Sclerotinia sclerotiorum.* Patogeniczność tych mikroorganizmów dla siewek salsefii określono na podstawie liczby wyrosłych roślin, ich zdrowotności oraz indeksu porażenia obliczonego na podstawie pięciostopniowej skali. Badane izolaty znacznie osłabiły wschody oraz zdrowotność salsefii. Na podstawie testów patogeniczności, gatunki takie jak *R. solani, S. sclerotiorum* i *P. irregulare* uznano za główną przyczynę zgorzeli siewek oraz zgniliznę korzeni roślin *Tragopogon porrifolius* var. *sativus.* Spośród badanych fitopatogenów odglebowych najbardziej patogenicznymi dla siewek salsefii okazały się izolaty *R. solani, S. sclerotiorum* i *P. irregulare,* a najmniej szkodliwymi *A. alternata* i *F. culmorum.*

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