

Antagonistic activity of selected fungi occurring in the soil after root chicory cultivation

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ABSTRACT

The present studies determined the antagonistic activity of selected fungi of the soil environment of root chicory – a high-inulin plant, with considerable pro-health values – towards such fungi pathogenic towards this plant as: *Alternaria alternata*, *Botrytis cinerea*, *Fusarium culmorum*, *F. oxysporum*, *Thanatephorus cucumeris* and *Sclerotinia sclerotiorum*. The cultivation of root chicory took into consideration soil mulching with cover crops (oats, tansy phacelia and common vetch) as well as the conventional cultivation, i.e. without any cover crops. The total population of fungi after soil mulching with common vetch was almost twice as small as in the control and smaller than with phacelia as a cover plant. Antagonistic *Clonostachys* spp., *Myrothecium* spp., *Penicillium* spp. and *Trichoderma* spp. displayed differentiated activity towards the studied fungi. The greatest antagonistic effect was observed after the mulch of oats. Besides, oats and common vetch the most positive effect on the antagonistic activity was that of *Clonostachys* spp., *Myrothecium* spp., *Penicillium* spp. and *Trichoderma* spp. Those fungi were most effective in inhibiting the growth and development of *F. oxysporum*, *T. cucumeris* and *S. sclerotiorum*.

Keywords: *Cichorium intybus*; plant pathogens; soil-borne fungi; antagonistic fungi

The world agriculture has been using different manners of cultivation for years with the aim of creating good conditions for the seed sprouting and the growth and development of plants. The latter uses intercrop cover plants that can be introduced into the soil or left on its surface in the form of so-called mulch (Borowy 2013). Plant mulch affects the physical properties of the soil, the management of the organic substance and mineral elements, weed infestation and the microbiological activity of the soil (Kołota and Adamczewska-Sowińska 2013). Of special importance is the effect of the plant mulch on the phytosanitary condition of the soil and the formation of the populations of microorganisms antagonistic towards plant pathogens (Patkowska and Konopiński 2013, 2014).

Mulching plants can inhibit the development of plant pathogens and stimulate the growth and

development of antagonistic microorganisms. The greatest antagonistic activity is found for fungi *Clonostachys* spp., *Myrothecium* spp., *Penicillium* spp. and *Trichoderma* spp. (Dipali et al. 2007, AyObi et al. 2010, Patkowska and Konopiński 2013, 2014).

The purpose of the studies was to determine the antagonistic activity of selected fungi of the soil environment of root chicory cultivated with using cover crops.

MATERIAL AND METHODS

Fieldwork. The field experiment was conducted in the years 2007–2008 in the Felin Experimental Station belonging to the University of Life Sciences in Lublin, district of Lublin (22°56'E, 51°23'N, Central Eastern Poland, 200 m a.s.l.), on a grey-

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brown podzolic soil. The object of the studies were selected fungi isolated from the soil sampled (every year during the first 10 days of June) from the depth of 5–6 cm of the plough layer of the field where root chicory (*Cichorium intybus* L. var. *sativum* Bisch.) cv. Polanowicka was cultivated. The cultivation of this plant took into consideration soil mulching with intercrop cover plants such as *Avena sativa* L., *Phacelia tanacetifolia* L. and *Vicia sativa* L. Chicory was sown during the first 10 days of May, whereas cover plants were sown in the first half of August of each year preceding the setting up of the experiment. Before winter, oats, phacelia and common vetch created an abundant green mass, constituting the natural mulch on the surface of the land. The conventional cultivation, i.e. without any cover crops, was the control. Completely randomized blocks method at four replications was used in the experiment.

Laboratory analyses. The microbiological analysis of the soil was made according to the method described by Czaban et al. (2007) and Patkowska (2009a,b). After preparing the proper dilutions of the soil, the total number of fungi was determined on Martin's medium. After the period of incubation, the number of fungi colonies was converted into 1 g dry weight (DW) of the soil, and the obtained isolates were determined to the species. In each studied year, the obtained isolates of *Clonostachys* spp., *Myrothecium* spp., *Penicillium* spp. and *Trichoderma* spp. were used to determine their antagonistic effect towards *Alternaria alternata*, *Botrytis cinerea*, *Fusarium culmorum*, *F. oxysporum*, *Thanatephorus cucumeris* and *Sclerotinia sclerotiorum* (isolated from the infected root chicory plants). Pathogenicity of those fungi towards the plants of root chicory was observed in earlier studies conducted in a growth chamber (Patkowska and Konopiński 2008). Estimation of the effect of *Clonostachys* spp., *Myrothecium* spp., *Penicillium* spp. and *Trichoderma* spp. on the studied pathogenic fungi was carried out using the method described by Mańka and Mańka (1992). Most frequently occurring species that build up over 75% of the fungal community were taken into consideration. Their phytopathological function is expressed by individual biotic effect (IBE) that is the effect of one isolate of the given species on the pathogens. The IBE multiplied by the species frequency results in the general biotic effects (GBE), treated as the effect of all the component's

isolates on the pathogen. After summarizing all the GBEs the summary biotic effect (SBE) will be obtained, providing the effect of the entire soil fungi community of the pathogen. The summary antagonistic effect of saprotrophic fungi from particular experimental combinations on the studied pathogenic fungi made it possible to determine their antagonistic activity in the soil environment of root chicory.

Statistical analysis. The total population of fungi was statistically analyzed, and the significance of differences was determined on the basis of the Tukey's confidence intervals ($P < 0.05$). Statistical calculations were carried out using Statistica program, version 6.0 (StatSoft, Krakow, Poland).

RESULTS AND DISCUSSION

The total population of fungi in the soil after using the mulch of oats was twice as low (28.19×10^3 CFU/g of soil DW – colony forming units/g dry weight of soil) as in the cultivation of root chicory without cover plants (57.50×10^3 CFU/g). The total population of fungi after soil mulching with common vetch was almost twice as small as in the control and smaller than with phacelia (Figure 1). The most of antagonistic fungi occurred in the

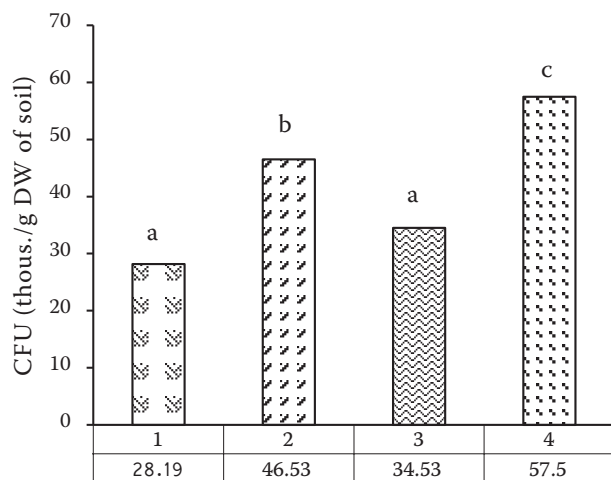


Figure 1. Total number of fungi isolated from the soil of particular experimental treatment (means from 2007–2008). 1 – soil after oat cultivation; 2 – soil after tansy phacelia cultivation; 3 – soil after common vetch cultivation; 4 – soil without cover crops cultivation. Means differ significantly ($P < 0.05$) if they are not marked with the same letter. CFU – colony forming units; DW – dry weight

Table 1. Activity of selected saprotrophic fungi isolated from soil after oat cultivation towards pathogenic fungi

Fungus species	Average number of isolates (2007–2008)	A.	B.	F.	F.	T.	S.
		<i>alternata</i>	<i>cinerea</i>	<i>culmorum</i>	<i>oxysporum</i>	<i>cucumeris</i>	<i>sclerotiorum</i>
average value of general biotic effect							
<i>Clonostachys rosea</i> (Link) Schroers, Samuels, Seifert & W. Gams.	59	237	265	325	296	325	295
<i>Myrothecium verrucaria</i> (Alb. et Schwein) Ditmar	26	104	78	104	104	130	156
<i>Penicillium aurantiogriseum</i> Dierckx	30	120	90	90	60	60	90
<i>Penicillium chrysogenum</i> Thom	3	–6	6	6	9	6	6
<i>Penicillium janczewskii</i> K.M. Zalessky	12	48	48	24	60	36	–12
<i>Penicillium simplicissimum</i> (Oudem.) Thom	10	30	20	10	20	20	30
<i>Penicillium verrucosum</i> Dierckx	10	30	40	20	30	20	20
<i>Trichoderma harzianum</i> Rifai	18	144	144	144	144	144	144
<i>Trichoderma koningii</i> Oudem	36	252	288	252	288	288	288
<i>Trichoderma pseudokoningii</i> Rifai	10	80	80	80	80	80	80
<i>Trichoderma viride</i> Pers.	48	384	384	384	384	384	384
Number of isolates	262						
Summary biotic effect		1423	1443	1439	1475	1493	1481

soil after using the mulch of oats (Tables 1–4). A decreased population of fungi in the soil was also found by Pięta and Kęsik (2007), who used common vetch and spring rye as mulch in the cultivation of onion and Patkowska and Konopiński (2013), who used oats in the cultivation of scorzoner.

The studied antagonistic fungi isolated from the soil after using the mulch of oats were the most effective in reducing the growth and development of *F. oxysporum*, *T. cucumeris* and *S. sclerotiorum*. The total value of their antagonistic effect was 1475, 1493 and 1481, respectively (Table 1).

Table 2. Activity of selected saprotrophic fungi isolated from soil after tansy phacelia cultivation towards pathogenic fungi

Fungus species	Average number of isolates (2007–2008)	A.	B.	F.	F.	T.	S.
		<i>alternata</i>	<i>cinerea</i>	<i>culmorum</i>	<i>oxysporum</i>	<i>cucumeris</i>	<i>sclerotiorum</i>
average value of general biotic effect							
<i>Clonostachys rosea</i> (Link) Schroers, Samuels, Seifert & W. Gams.	20	82	44	78	80	80	98
<i>Myrothecium verrucaria</i> (Alb. et Schwein) Ditmar	10	20	30	30	40	40	40
<i>Penicillium aurantiogriseum</i> Dierckx	16	48	32	16	16	16	32
<i>Penicillium chrysogenum</i> Thom	4	–48	4	4	8	4	4
<i>Penicillium simplicissimum</i> (Oudem.) Thom	5	5	5	5	5	5	10
<i>Penicillium verrucosum</i> Dierckx	11	22	33	22	11	11	11
<i>Trichoderma harzianum</i> Rifai	12	72	72	72	96	72	72
<i>Trichoderma koningii</i> Oudem	23	184	184	184	161	184	161
<i>Trichoderma pseudokoningii</i> Rifai	10	70	70	70	70	60	80
<i>Trichoderma viride</i> Pers.	11	88	88	88	88	88	88
Number of isolates	122						
Summary biotic effect		563	562	569	575	560	596

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Table 3. Activity of selected saprotrophic fungi isolated from soil after common vetch cultivation towards pathogenic fungi

Fungus species	Average number of isolates (2007–2008)	A.	B.	F.	F.	T.	S.
		<i>alternata</i>	<i>cinerea</i>	<i>culmorum</i>	<i>oxysporum</i>	<i>cucumeris</i>	<i>sclerotiorum</i>
average value of general biotic effect							
<i>Clonostachys rosea</i> (Link) Schroers, Samuels, Seifert & W. Gams.	42	138	192	198	150	168	198
<i>Myrothecium verrucaria</i> (Alb. et Schwein) Ditmar	18	54	54	54	72	90	90
<i>Penicillium aurantiogriseum</i> Dierckx	10	20	30	20	20	20	10
<i>Penicillium chrysogenum</i> Thom	4	–8	4	4	4	4	8
<i>Penicillium expansum</i> Link	7	–14	–7	14	–7	–7	–14
<i>Penicillium janczewskii</i> K.M. Zalessky	11	22	22	11	33	33	22
<i>Penicillium simplicissimum</i> (Oudem.) Thom	1	3	1	1	2	2	2
<i>Penicillium verrucosum</i> Dierckx	22	66	44	44	44	44	22
<i>Trichoderma aureoviride</i> Rifai	10	80	70	60	70	70	80
<i>Trichoderma harzianum</i> Rifai	15	105	105	105	120	120	105
<i>Trichoderma koningii</i> Oudem	21	168	168	168	168	168	147
<i>Trichoderma pseudokoningii</i> Rifai	19	114	114	152	133	114	114
<i>Trichoderma viride</i> Pers.	20	160	160	160	160	160	160
Number of isolates	200						
Summary biotic effect		908	957	991	969	986	944

The tested antagonistic fungi occurring in the soil after the mulch of phacelia decreased the studied pathogenic fungi in a similar degree. The highest antagonistic activity was found, however, towards *S. sclerotiorum* and *F. oxysporum* since

the total value of antagonistic effect was 596 and 575, respectively (Table 2). The population of antagonistic *Clonostachys* spp., *Myrothecium* spp., *Penicillium* spp. and *Trichoderma* spp. occurring in the soil environment of root chicory cultivated

Table 4. Activity of selected saprotrophic fungi isolated from soil without cover crops cultivation towards pathogenic fungi

Fungus species	Average number of isolates (2007–2008)	A.	B.	F.	F.	T.	S.
		<i>alternata</i>	<i>cinerea</i>	<i>culmorum</i>	<i>oxysporum</i>	<i>cucumeris</i>	<i>sclerotiorum</i>
average value of general biotic effect							
<i>Clonostachys rosea</i> (Link) Schroers, Samuels, Seifert & W. Gams.	6	12	12	18	18	18	24
<i>Penicillium aurantiogriseum</i> Dierckx	2	4	4	2	2	2	2
<i>Penicillium janczewskii</i> K.M. Zalessky	2	4	4	4	6	4	–4
<i>Penicillium chrysogenum</i> Thom	1	–2	1	1	1	1	2
<i>Penicillium verrucosum</i> Dierckx	3	6	9	3	6	3	3
<i>Trichoderma harzianum</i> Rifai	6	36	36	36	36	36	36
<i>Trichoderma koningii</i> Oudem	4	32	24	28	28	32	32
<i>Trichoderma pseudokoningii</i> Rifai	1	6	8	6	7	7	7
<i>Trichoderma viride</i> Pers.	1	8	7	8	8	8	6
Number of isolates	26						
Summary biotic effect		106	105	106	112	111	108

after the mulch of common vetch was larger than after the mulch of phacelia. They reduced the growth and development of *F. culmorum* (991) and *T. cucumeris* (986) the most (Table 3). The fewest antagonistic fungi occurred in the soil with the conventional cultivation of root chicory, i.e. without any cover plants. Those fungi were also characterized with the lowest antagonistic activity towards the studied pathogenic fungi. The value of their antagonistic effect ranged from 105–112. The studied antagonistic fungi isolated from the soil without any cover plants were the most effective in reducing the growth and development of *F. oxysporum* (112) (Table 4).

The present studies showed differentiated antagonistic activity of fungi isolated from the soil environment of root chicory cultivated with the use of plant mulch. Oats and common vetch as cover plants had the best effect on this property. This could have resulted from the composition of the root exudates of the cover plant and from the ability of the enumerated fungi to mycoparasitism and antibiosis (Mukherjee et al. 2007). As reported by Dipali et al. (2007) and AyObi et al. (2010), antagonistic fungi – through cellulolytic and chitinolytic enzymes, cause the lysis of the mycelium hyphae of a number of plant pathogens.

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