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BACTERIA AND FUNGI COMMUNITIES OCCURRING IN THE SOIL AFTER THE CULTIVATION OF CEREALS AND POTATO

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ABSTRACT

The studies conducted in the years 1997-1999 concerned the soil after cultivation of rye, winter wheat, triticale and potato. The purpose of the studies was to determine the composition of bacteria and fungi communities in the soil after the cultivation of these plant species.

As a result of the microbiological analysis of the soil after rye cultivation $3.08x10^6$ colonies of bacteria and 28.27×10^3 colonies of fungi were obtained. After triticale cultivation the soil contained 3.99×10^6 bacteria colonies and $38.74x10^3$ fungi colonies, whereas after winter wheat cultivation there were 4.89×10^6 bacteria colonies and 26.19×10^3 fungi colonies. The soil after potato cultivation contained the greatest numbers of

bacteria (5.01×10^6) and fungi (43.11×10^3) colonies. Among the obtained bacteria and fungi the greatest number of antagonistic microorganisms was found in the soil after winter wheat and rye cultivation, while the smallest number of antagonistic microorganisms was found in the soil after potato cultivation.

Key words: antagonism, *Bacillus* spp., *Pseudomonas* spp., *Trichoderma* spp., *Gliocladium* spp., winter wheat, triticale, rye, potato.

INTRODUCTION

The basic plant species cultivated in Poland include winter wheat, triticale, rye and potato. It follows from abundant information in literature that each of these plants exerts a remarkable influence on the quantitative and qualitative composition of fungi and bacteria in the soil environment [Huber, Watson, 1970; Księżak, Kobus, 1993]. Organic residue of particular plants plays a significant role in the formation of microorganisms [Huber, Watson, 1970]. On the other hand, in the vegetative period a special effect on the composition and numbers of bacteria and fungi populations is also exerted by root exudates, which are a rich source of free aminoacids and sugars [Funck-Jensen, Hockenhull, 1984; Pięta, 1981; Smith, Peterson, 1966; Rovira, 1965; 1969]. The chemical composition of organic residue and root exudates can have a stimulating or inhibiting effect on the growth and development of particular populations of microorganisms [Funck-Jensen, Hockenhull, 1984; Martyniuk et al., 1991; Pięta et al., 1999; Rovira, 1965; Schoruvitz, Zeigler, 1989; Sundin et al., 1990]. According to Solarska [1996], ploughing over the green mass of rye caused an increase of bacteria population by 50%, and this especially concerned the number of fluorescent *Pseudomonas* spp. characterised by antagonistic effect towards pathogenic fungi. Ploughing over post-harvest residue of barley caused an increase of the number of antagonistic isolates of *Bacillus subtilis* [Weinhold, Browman, 1968].

Determination of the composition of microorganisms populations formed in the soil under the effect of the cultivation of winter wheat, triticale, rye and potato was the **purpose** of the presented studies.

MATERIALS AND METHODS

The studies were carried out in the years 1997-1999 on an experimental field in Czesławice near Nałęczów. The plot was sown with winter wheat (Kobra cv.), triticale (Malno cv.), rye (Amilo cv.) and with cultivation of potato (Bronka cv.). The soil after the harvest of these plants was the subject of the studies. Soil sampling and then the manner of conducting the microbiological analysis in order to determine the population of microorganisms were true to the method described by Martyniuk et al. [1991]. Soil samples from particular plots were taken from the depth of 5 to 10 cm to sterile Petri dishes. Next, in the laboratory sterile conditions a soil solution was prepared in the dilutions ranging from 10^{-1} to 10^{-7}.

The total number of bacteria in 1 g of dry weight of the examined soil was determined on the medium "Nutrient agar" using soil solutions in the dilutions of 10^{-5}, 10^{-6}, 10^{-7}. In the case of *Bacillus* spp. "Tryptic soy agar" medium and dilution of 10^{-4}, 10^{-5}, 10^{-6} were used, while for *Pseudomonas* spp. the studies made use of "Pseudomonas agar F" medium and dilutions of 10^{-2}, 10^{-3}, 10^{-4}. The total number of fungi in 1g of dry weight of the same soil samples was determined on Martin's medium [1950], using the dilutions of 10^{-2}, 10^{-3}, and 10^{-4}.

The results concerning the number of bacteria and fungi were statistically analysed, and the significance of differences was determined on the basis of Tukey's confidence intervals.

In each year of the studies the obtained isolates (100 isolates of *Bacillus* spp. and 100 of *Pseudomonas* spp.) and all the isolates of saprophytic fungi of *Gliocladium* spp. and *Trichoderma* spp. were used to determine their antagonistic effect towards such pathogenic fungi as *Fusarium culmorum, F. oxysporum, F. solani, Pythium irregulare* and *Rhizoctonia solani.* In order to determine the antagonistic effect of the studied bacteria towards pathogenic fungi a 5-degree scale described by Martyniuk et al. [1991] and degrees of inhibition of the growth of pathogens provided by Pięta [1999] were used. An estimation of the effect of saprophytic fungi on the examined pathogenic fungi was performed by means of biotic rows [Mańka, 1974; Mańka, Mańka, 1992], while an individual antagonistic effect was determined on the basis of the scale provided by Mańka and Kowalski (1968).

RESULTS

A microbiological analysis of the soil after the cultivation of winter wheat, triticale, rye and potato found out differences in the number of particular populations of microorganisms (tab. 1). In particular years, the total number of bacteria in 1 g of dry weight of the examined soil samples ranged from 2.98×10^6 colonies (in 1999 after rye cultivation) to 5.01×10^6 colonies (in 1999 after potato cultivation). The lowest number of bacteria, i.e. 3.08×10^6 colonies, was found in the soil after rye cultivation, while the highest (5.01×10^6) after potato cultivation (tab. 1). The other two examined soil samples, that is after triticale cultivation and winter wheat cultivation, differed with the total number of bacteria. In each year there were significantly more bacteria colonies in 1 g of dry weight after winter wheat cultivation (4.89×10^6 on average) than in the soil after triticale cultivation (3.99×10^6 on average).

Table 1. Number of bacteria and fungi occurring in the soil after cultivation of particular plants

Type of soil	Total number of bacteria (mln/1g d. w. of soil)				Number of bacteria of *Bacillus* spp. (mln/1g d. w. of soil)				Number of bacteria of *Pseudomonas* spp. (mln/1g d. w. of soil)				Total number of fungi (thous./1g d. w. of soil)			
	1997	1998	1999	mean	1997	1998	1999	mean	1997	1998	1999	mean	1997	1998	1999	mean
rye	2.99^{a*}	3.27^{a}	2.98^{a}	3.08^{a}	0.71^{a}	0.89^{a}	0.98^{a}	0.86^{a}	2.01^{c}	2.12^{c}	2.08^{c}	2.07^{b}	26.12^{b}	28.61^{a}	30.08^{a}	28.27^{a}
triticale	3.75^{b}	4.02^{b}	4.20^{b}	3.99^{b}	0.89^{ab}	0.99^{a}	1.03^{a}	0.97^{ab}	1.56^{b}	1.68^{b}	1.65^{b}	1.63^{ab}	34.03^{c}	39.97^{b}	42.22^{b}	38.74^{b}
winter wheat	4.68^{c}	4.93^{c}	5.05^{c}	4.89^{c}	1.17^{b}	1.39^{b}	1.55^{b}	1.37^{b}	1.86^{bc}	1.99^{bc}	2.18^{c}	2.01^{b}	21.98^{a}	28.72^{a}	27.87^{a}	26.19^{a}
potato	4.96^{c}	4.99^{c}	5.08^{c}	5.01^{c}	1.08^{b}	1.19^{ab}	1.24^{b}	1.17^{ab}	0.89^{a}	1.08^{a}	1.12^{a}	1.03^{a}	39.73^{c}	44.53^{b}	45.07^{b}	43.11^{b}

*** means in columns differ significantly (P≤0.05), if they are not marked with the same letter**

In the case of *Bacillus* spp. bacteria the number of colonies in 1 g of dry weight of soil ranged from 0.71×10^6 (in 1997 after rye cultivation) to 1.55×10^6 (in 1999 after winter wheat cultivation). In each year the greatest number of *Bacillus* spp. colonies was found in the soil after winter wheat cultivation (1.37×10^6 colonies on average), while the smallest one in the soil after rye cultivation (0.86×10^6 colonies on average) (tab. 1). The mean number of *Bacillus* spp. bacteria occurring in soil samples after triticale cultivation (0.97×10^6 colonies) and potato cultivation (1.17×10^6 colonies) varied, but the differences were statistically insignificant.

Results of the microbiological analysis showed that in each year of the studies the highest number of *Pseudomonas* spp. colonies was found in the soil samples after rye cultivation (2.07×10^6 colonies on average) and after winter wheat cultivation (2.02×10^6 colonies in 1 g of

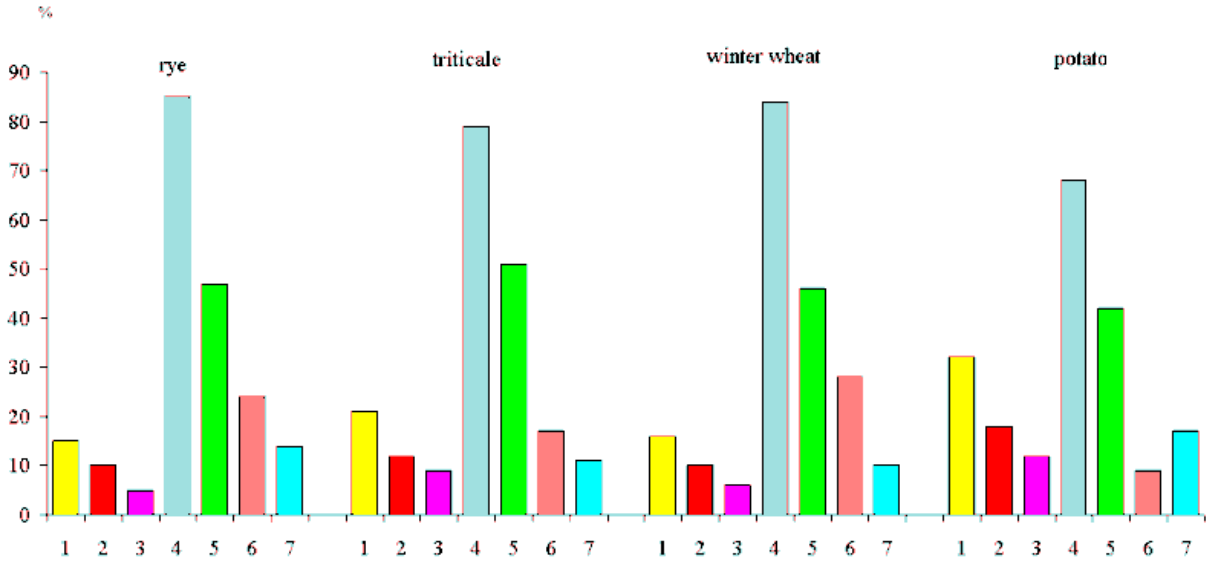
soil dry weight on average). The lowest number of colonies of this bacteria genus was found after potato cultivation (1.03×10^6 colonies in 1 g of soil dry weight on average), tab. 1.

The mycological analysis of the examined soil samples pointed at considerable differentiation in the number of fungi (tab. 1). The smallest number of fungi colonies was found in the soil after rye cultivation (28.27×10^3 colonies on average) and winter wheat (26.19×10^3 colonies on average), while the greatest after potato cultivation (4.11^3 colonies in 1 g of soil dry weight). A high number of fungi, namely 38.74×10^3 colonies, was found in 1 g of soil dry weight after triticale cultivation (tab. 1).

The mycological analysis of soil samples after the cultivation of particular plants isolated both pathogenic and saprophytic fungi. The proportion of those fungi differed with the species of the cultivated plant. The greatest number of pathogenic fungi, especially of *Fusarium* spp. and *Rhizoctonia solani* was found in the soil after potato cultivation, and the smallest number after the cultivation of rye and winter wheat (fig. 1). The isolated fungi of *Fusarium* spp. included *F. culmorum, F. solani* and *F. oxysporum.*

Within the saprophytic fungi the dominating species were of *Pseudomonas* spp. and *Trichoderma* spp., especially in the soil after the cultivation of winter wheat and rye. *Penicillium* spp. was represented by *P. brevi-compactum, P. decumbens, P. fellutanum, P. frequentans, P. funiculosum, P. janthinellum, P. lividum, P. meleagrinum, P. nigricans, P .paxilli, P. purpurescens, P. purpurogenum, P. roseo-purpureum, P. velutinum, P. verrucosum* var. *cyclopium, P. verrucosum* var. *verrusocum,* while *Trichoderma* spp.was represented by *T. hamatum, T. harzianum, T. koningii, T. viride.* Those fungi were a little less numerous in the soil after triticale cultivation, and the least numerous in the soil after potato cultivation (fig. 1). The saprophytic fungi marked as 'others' in the chart included the species from the genera of *Acremonium, Chaetomium, Cladosporium, Gliocladium, Mucor* and *Rhizopus.* The proportion of isolates of those fungi in particular soil samples ranged from 10.01% to 17.03% of the total number of fungi colonies (fig. 1).

Fig.1. Participation of fungi in soil after cultivation of particular plants: 1 – total pathogenic fungi, 2 – *Fusarium* spp., 3 – *Rhizoctonia solani* 4 – total saprophitic fungi, 5 – *Penicillium* spp., 6 – *Trichoderma* spp., 7 – other saprophitic fungi



Among the bacteria *Bacillus* spp., *Pseudomonas* spp. and the fungi *Gliocladium* spp. and *Trichoderma* spp., laboratory studies *in vitro* separated the isolates characterised by antagonistic effect towards pathogenic fungi. The frequency of the occurrence of antagonistic microorganisms in particular soil samples varied (tab. 2). In the case of *Trichoderma* spp. all the isolates separated from the analysed soils turned out to be antagonistic. On the other hand, 70% isolates within *Gliocladium* spp. were distinguished by these properties, while antagonistic bacteria of *Bacillus* spp. constituted 23% on average, and those of *Pseudomonas* spp. made up 27% of all the obtained isolates. The highest number of antagonistic bacteria and fungi (388 isolates) was found in the soil after winter wheat cultivation, and the lowest in the soil after potato cultivation (105 isolates). In the other soils, that is those after rye and triticale cultivation, there were 327 and 286 isolates of antagonistic microorganisms, respectively (tab. 2).

Table 2. Antagonistic bacteria and fungi isolated from the soil after cultivation of particular plants (sum of isolates 1997-1999)

Bacteria and fungi	Number of isolates			
	rye	triticale	winter wheat	potato
Bacillus spp.	31	37	51	13
Pseudomonas spp.	117	86	105	18
Gliocladium catenulatum	12	17	32	7
Gliocladium roseum	3	1	3	1
Trichoderma hamatum	36	27	36	8
Trichoderma harzianum	43	39	56	-
Trichoderma koningii	29	31	43	26
Trichoderma viride	56	48	62	32
Total	327	286	388	105

DISCUSSION

Results of the studies pointed at varied composition of bacteria and fungi communities formed under the effect of cultivation of particular plant species. The soil after potato cultivation contained the greatest total number of bacteria and fungi colonies. On the other hand, the soil after winter wheat cultivation was characterised by the smallest number of fungi colonies. Small numbers of fungi colonies were also found in the soil after rye cultivation, and a big number of fungi was present in the soil after triticale cultivation. The quantitative and qualitative composition of microorganisms could have been affected by root exudates and post-harvest residue of the cultivated plants [Huber, Watson, 1970; Rovira, 1965; 1969; Schroth, Hildebrand, 1964]. According to Funck-Jensen and Hockenhull [1984], the chemical composition of root exudates and plant residue significantly affects the changes in the numbers within the populations of bacteria and fungi. Chemical compounds exudated by roots

to the soil or those occurring in post-harvest residue can stimulate or inhibit the growth and development of soil-borne microorganisms.

Results of the mycological analysis showed that the soil after rye and winter wheat cultivation contained twice a smaller number of pathogenic fungi than the soil after potato cultivation, while the soil after triticale cultivation contained 50% fewer pathogenic fungi as compared to the soil after potato cultivation, but 35% more of them in comparison with the soil after rye and winter wheat cultivation. A high number of isolates of pathogenic fungi in the soil after potato cultivation can be explained by the smallest proportion of bacteria and saprophytic fungi populations with an antagonistic effect. On the other hand, the smallest proportion of pathogenic fungi isolates was found in the soil after rye and winter wheat cultivation, which can be related to numerous occurrence of antagonistic bacteria of *Bacillus* spp. and especially *Pseudomonas* spp., and such antagonistic fungi as *Gliocladium* spp. and *Trichoderma* spp.

According to Keel [1992] and Weller [1988], bacteria of *Pseudomonas* spp. are capable of active colonisation of plant roots, owing to which they can effectively compete with pathogens for nutritive elements found in root exudates and thus become a factor of biological plant control. The species from the genera of *Gliocladium* and *Trichoderma*, which are found in big numbers in the soil after the cultivation of the studied cereals (rye, winter wheat, triticale) can significantly reduce the numbers of populations of soil-borne pathogens (Łacicowa, Pięta, 1985a; 1985b; 1989; Papavizas, 1985]

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