Biological control of potato against
*Rhizoctonia solani* (Kühn)

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The aim of three years field investigations was to evaluate the effect of bio-preparations Polyversum (B.A.S. *Pythium oligandrum*) and Biochikol 020 PC (B. A. S. chitosan) applied on infected tubers by *Rhizoctonia solani* sclerots during vegetation period. As a standard fungicide Vitavax 2000 FS (B.A.S. karboxin and thiuram) was used. The effect of the mentioned preparations applied at three different concentrations on *R. solani* mycelium linear growth was investigated under *in vitro* conditions. The examinations were made using Kowalik and Krechniak method (1961). During potato vegetation period all applied preparations affected both the lower tubers’ infestation degree by *R. solani* sclerots and the lower tuber infestation percent by these pathogens.

According to the results obtained from conducted *in vitro* examinations, it was found that Polyversum preparation, chemical preparation (Vitavax 2000 FS) and Biochikol 020 PC but applied only at the highest concentration (2.0 %) significantly reduced mycelium linear growth of *R. solani*.

**Key words:** *Rhizoctonia solani*, *Pythium oligandrum*, chitosan, karboxin and thiuram bioprotection, potato.

**Introduction.** In Poland potato belongs to economically important cultures. This plant is infected by many agrophags. Fungi *Rhizoctonia solani*, which causes black scurf of potato tubers, belong to commonly appearing potato pathogens. Sclerots of the mentioned fungi occurring on sets can be the source of infection for plants and descendant tubers. Moreover, it make their quality worsen (Ahrenniemi et al., 2005). Potato yield losses caused by this disease amounted even to 50 % (Hdni et al., 1998). Besides, rhizoctoniosis constitutes distinct aesthetic defect, which decreases market value of potatoes intendent for consumer purpose or for food industry (Lutomirska, 2007).

*R. solani* is soil born pathogens, which affects sets and contributes not only to yield losses but to fungi accumulation in soil (Bogucka, 1993). According to this author, interference with chemical preparations by sets dressing is still in use because of lack of discussed disease-resistant varieties. However, increasing environmental contamination through use pesticides among others things incline us for using alternative methods to fight with plant pathogens. In modern plant protection an interest of biological methods, which consist of replacing pesticides with bio-preparations based on plant’s extracts, organic compounds and antagonistic microorganisms to pathogens, is increasing. To
the mentioned antagonistic organisms belongs myco-parasite *Pythium oligandrum*, biologically active substance of bio-preparation Polyversum. Moreover, there are high possibilities of Biochikol 020 PC (B. A. S. chitosan) application in protection of many plants against diseases, especially as plant immunity inducer (Bell et al., 1998; Orlikowski et al., 2002, Pięta et al., 2006).

Therefore, the objective of the presented study conducted as field experiment was the test of reduction of *R. solani* occurrence by application of bio-preparation Polyversum (contained *Pythium oligandrum* oospors) and Biochikol 020 PC (B. A. S. chitosan). As standard fungicide Vitavax 2000 FS (B. A. S. karboxin and thiuram) was used. Besides, the effect of mentioned bio-preparations on the *R. solani* mycelium linear growth was investigated under *in vitro* conditions.

**Object, methods and conditions.** Field experiment was conducted at the Experimental Station at Mydlinki near Kraków owned by the Department of Plant Protection Academy of Agriculture, on brown soil, in 2005–2007. The winter wheat was the forecrop. During the investigations potatoes of mid-early cv. ‘Ibis’ cultivated according to recommendation of proper agro-technique was used. The experiment was established in third decade of April with the method of random squares in four replications (100 tubers on each plot) and in following combinations: 1 – control – plants derived from tubers without any protection treatment; 2 – plants derived from tubers dressed with Biochikol 020 PC (B. A. S. chitosan) at concentration of 2.5 %; 3 – dressed tubers + 4 times plants spraying with Biochikol 020 PC at concentration of 2.5 %; 4 – plants derived from tubers dressed with Polyversym (B. A. S. *Pythium oligandrum*) in dose 10 g/kg tubers; 5 – dressed tubers + 4 times plants spraying with Polyversum at concentration of 0.05 %; 6 – plants derived from tubers dressed with Vitavax 2000 FS (B. A. S. karboxin and thiuram) in dose 5 ml/kg tubers.

After harvesting 100 tubers from each plot were randomly chosen, collected and put in storage (temperature 6–7 °C, relative air humidity 80–85 %). The analysis of degree and percent of tubers infested by *R. solani* sclerots were directly made after harvesting (in September) and later every 3 months (in December and March) during the storage period. The degree of infestation by above mentioned pathogens was defined according to following scale (Kapsa et al., 1998): 0 – lack of symptoms, 1 – disease symptoms founded on 5–25 % of tubers’ surface, 2 – disease symptoms on 26–50 % of tubers’ surface, 3 – disease symptoms appearing on 51–75 % of tubers’ surface, 4 – disease symptoms including over 75 % of surface. Received results were subjected to statistical analysis using analysis of variance. The multiple t-Duncan test was used to estimate the differences between mean values at significance level α = 0.05.

The Kowalik and Krechniak (1961) method was used to the *in vitro* studies. *R. solani* was isolated from potato tubers. Each of examined preparations were applied at three different concentrations: Biochikol 020 PC at concentrations: 0.5 %, 1.0 %, 2.0 %; Polyversum at concentrations: 0.05 %, 0.1 %, 0.2 %; Vitavax 2000 FS in doses: 0.025 %, 0.05 %, 0.1 %.

After pouring PDA agar to Petri dishes and after its silidified, the agar disk with mycelium of 14-days old culture of examined fungi was placed centrally into each dish. The control constituted Petrie dishes with medium PDA and agar disk with *R. solani* mycelium but without amendments. The test was carried out in five repetitions for each combination (10 Petri dishes for 1 repetition). Measurements
were started when first increase of mycelium in control dishes was observed and were conducted until the complete overgrowth of mentioned dishes was noted down. The crosswise measurement of colony diameter was made, and then the arithmetic mean for repetitions was calculated. The percent of inhibition of mycelium linear growth on medium with amendments of appropriate preparation compared to the growth of fungi on control Petri dish was used to measure the preparations activity (Kowalik and Krechniak, 1961). Received results were treated statistically using the analysis of variance followed by Duncan’s test (α = 0.05).

**Results.** According to the received results during three years of investigations the favourable effect of both sets dressing and potato plant spraying with tested preparations on the reduction of black scurf on tubers was observed (Table).

**Table.** The effect of bio-preparations on potato tuber *Rhizoctonia solani*, 2005–2007


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<th>Combination Derinys</th>
<th>Average degree of tuber infestation by <em>R. solani</em> sclerots</th>
<th>Percent of infected tubers</th>
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<td>Tubers dressed and plants sprayed with Biochikol 020 PC</td>
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* Means followed by the same letter do not differ at 5 % level of significance (Duncan’s multiple range test)

Application of chemical standard preparation Vitavax 2000 FS to sets dressing had the best influence on both inhibition of tubers’ infestation degree and tubers’ infestation percent by *R. solani* sclerots during the storage period. In all terms of analysis of tubers
from this combination, the percent of tubers’ infestation by *R. solani* sclerots was the
lowest and amounted from 1.0 (September) to 1.2 (March) – Table. Also the percent of
tubers infestation by discussed pathogen was the lowest and reached from 11.1 %
(September) to 16.8 in March (Table).

In comparison to the control Polyversum and Biochikol 020 PC preparations
significantly reduced *R. solani* growth on stored tubers of tested potato variety in all
terms of analysis (Table). Statistical calculations showed the important influence of
all preparations under consideration on the inhibition of both degree and percent of
infested tubers by *R. solani* sclerots (Table).

Figure represents the percent of *R. solani* mycelium linear growth inhibition.
Among examined preparations Polyversum applied at all concentrations (0.05 %,
0.1 %, 0.2 %) the most limited *R. solani* mycelium linear growth. The percent of
inhibition appropriately came to 89.5 %, 91.1 %, 92.0 % (Fig.). Also in combination
where Vitavax 2000 FS was used in all tested doses the important inhibition of above
mentioned pathogen mycelium linear growth was found but the best inhibition was
noted down at the highest concentration of this preparation, which is 0.1 % (87.5 %) –
Fig. The weakest *in vitro* effect of reduction *R. solani* mycelium linear growth was
found in combinations where Biochikol 020 PC was used. Mentioned preparation
limited *R. solani* mycelium linear growth applied only at the highest concentration
(2.0 %), whereas in doses 0.5 % and 1.0 % there was not any statistically important
effect of Biochikol 020 PC application on the discussed pathogen mycelium linear
growth (Fig.).

![Figure: Percent of inhibition of Rhizoctonia solani growth](image)

According to statistical calculations, the significant influence (in comparison to the
control) of preparations under examination (with the exception of Biochikol 020 PC

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*Fig. Percent of inhibition of *Rhizoctonia solani* growth

Pav. *Rhizoctonia solani* augimo inhibicijos procentas

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*Reikšmės, pažymėtos toms pažiūros raidemis, nesiskiria 5 % patikimumo lygje (Dunkano testas)*
at concentration 0.5 % and 1.0 %) on the percent of _R. solani_ mycelium linear growth inhibition was noted down.

**Discussion.** Applied preparations limited the black scurf of tubers of examined potato variety. Investigations conducted by Gajda and Kurzawińska (2004) confirmed favourable effect of sets dressing as well as Polyversum bio-preparation and chemical preparation on the inhibition of _R. solani_ sclerots occurrence on potato tubers. Similar results were obtained by Bernart and Osowski (2006). Findings from conducted experiments showed the possibility of the use of both preparations Polyversum and Biochikol 020 PC in protection of potatoes against _R. solani_. Previous test data demonstrated that application of Polyversum bio-preparation to sets dressing limited contamination of descended tubers by _R. solani_ sclerots. Under *in vitro* conditions above-mentioned preparation showed strong antifungal activity in relation to _R. solani_ (Gajda, Kurzawińska, 2004). It is believed that the application of micro-biological material on the seeds (tubers) surface is the most effective method of prevention through infestation. According to Martin and Hancock (1987), _Pythium oligandrum_ colonizes the ecological niche in the soil and successfully competes with plant pathogens. The authors showed that _Pythium oligandrum_ have an influence on pathogenic factors. It resulted that the great number of pathogenic factors in the soil, on seeds, tubers, bulbs and rootstocks may decrease to the degree when the significant increase of plant health is noted down. According to Deacon (1991), _Pythium oligandrum_ demonstrates destructive and parasitic character to many pathogenic fungi. Bell et al. (1998), Kurzawińska (2007), Nawrocki and Mazur (2007), Pięta et al. (2006) confirmed efficiency of chitosan in vegetable protection. The results of investigations conducted by above-mentioned authors showed that efficiency of protective chitosan activity applied in plant prevention is much better than applied in plant intervention. Effective influence of chitosan on phyto-pathogens depends on its concentration and the virulent of infectious factor. Followed to Pośpieszny (1997), chitosan induces immunity of many plants through cell wall lignification, phytoalexins production and synthesis of proteinase inhibitors. The treatment of the cells with chitosan stimulates their intensification by production of additional structures and accumulation of phenol substances, which are harmful for fungi. Chitosan applied together with fungicides may complete their activity reducing the doses and fungi immunization against them (Pośpieszny, 1997). Among the tested preparations under *in vitro* conditions bio-preparation Polyversum the most limited _R. solani_ mycelium linear growth. Chemical preparation Vitavax 2000 FS showed slightly weak effect. The least inhibition of _R. solani_ mycelium linear growth was found in combinations where Biochikol 020 PC was used. Followed to Orlikowski et al. (2002) most of studies on mechanism of chitosan activity to pathogenic fungi showed that mentioned substance do not inhibit the mycelium growth and spores’ germination under *in vitro* conditions. However, according to Pośpieszny (1997), under *in vitro* conditions chitosan inhibits growth of several fungi and microbes but not all of them. Exploitation of preparations based on natural substances, which can limit plant pathogens development comes into higher and higher prominence, especially restricting traditional chemical preparation application. It results from comparable efficiency of bio-preparations to pesticides.
Conclusions. 1. Polyversum and Biochikol 020 PC bio-preparations significantly (in comparison with control) reduced degree and percent of tuber infestation by R. solani.

2. Application of chemical standard preparation karboxin and thiuram mixture to sets dressing had the best influence on inhibition of tubers infestation by R. solani.

3. Among all the tested preparations under in vitro conditions the most effective in reduction R. solani mycelium linear growth turned out to be Polyversum bio-preparation. In vitro response of the tested pathogen depended on the type of preparation and its concentration.

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References


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**Bulvių biologinė kontrolė prieš *Rhizoctonia solani* (Kühn)**

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**Santrauka**


**Reikšminiai žodžiai:** bioapsauga, bulvės, chitosanas, karboksinas, *Pythium oligandrum*, *Rhizoctonia solani*, tiuramas.