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EFFECT OF FUNGAL INFECTION AND THE APPLICATION OF THE BIOLOGICAL AGENT EM 1™ ON THE RATE OF PHOTOSYNTHESIS AND TRANSPIRATION IN PEA (*PISUM SATIVUM* L.) LEAVES

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A b s t r a c t

Field experiments conducted during the years 2003-2005 showed that the rate of photosynthesis and transpiration decreased as a result of pea infection by *Peronospora viciae*. Foliar application of effective microorganisms (EM) combined with chemical control increased the rate of photosynthesis in pea, while other methods of EM application reduced net photosynthesis values (*An*). Chemical control and seed dressing with the tested biological agent caused a significant decrease in molar transpiration (*E*) values, compared to the control treatment. Soil application of EM contributed to inhibiting fungal pathogen infestation on pea plants.

K e y w o r d s: rate of photosynthesis, rate of transpiration, fungal pathogens, biological control.

WPŁYW INFEKCJI GRZYBOWEJ ORAZ APLIKACJI BIOLOGICZNEGO PREPARATU EM 1™ NA INTENSYWNOŚĆ FOTOSYNTEZY I TRANSPIRACJI LIŚCI GROCHU SIEWNEGO (*PISUM SATIVUM* L.)

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S ł o w a k l u c z o w e: fotosynteza, transpiracja, patogeny grzybowe, biologiczna ochrona roślin.

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Abstrakt

W badaniach polowych, przeprowadzonych w latach 2003-2005, stwierdzono zmniejszenie tempa fotosyntezy i transpiracji na skutek infekcji grochu patogenem *Peronospora viciae*. Stosowanie nalistne EM w połączeniu z ochroną chemiczną zwiększało intensywność fotosyntezy grochu, natomiast inne sposoby aplikacji tego preparatu ograniczały jej wartość netto (*An*). Wartości wskaźnika transpiracji molowej (*E*) po stosowaniu ochrony chemicznej oraz zaprawianiu nasion preparatem biologicznym były istotnie niższe niż w obiekcie kontrolnym. Stosowanie doglebowe preparatu biologicznego EM ograniczało występowanie patogenów grzybowych na roślinach.

Introduction

The photosynthetic capacity of plants is affected by abiotic and biotic stress factors. Infections cause leaf blade damage which leads to a decrease in the rate of photosynthesis, disappearance of chlorophyll and an increase in the rate of respiration (SCHOLES 1992). It is assumed that at an advanced stage of a disease the rate of photosynthesis may be reduced by 75% (GRZESIUK et al. 1999). A decrease in the rate of photosynthesis results from water unbalance in the plant, changes in the structure of mesophyll cells, a reduction in the number of chloroplasts and changes in their structure, an increase in the concentration of carbohydrates, and a decrease in the concentration of CO_2 in leaf tissues around the lesion (PINKARD, MOHAMMED 2006). The process of photosynthesis may also undergo certain modifications in consequence of reduced activity of ribulose 1.5-biphosphate (RuBP) and ribulose 1.5-biphosphate carboxylase/oxygenase (RubisCO) (MCELRONE, FORSETH 2004). The rate of photosynthesis usually increases in the first phase of pathogenesis, but then the rate of biochemical reactions involved in photosynthesis decreases rapidly. The inhibition of photosynthesis is directly proportional to pathogen virulence (PINKARD, MOHAMMED 2006).

Apart from photosynthesis, infections may also modify other physiological processes taking place in the plant. The metabolic activity of leaf cells increases from the moment of infection, which is usually followed by an increase in respiratory rate (BASSANEZI et al. 2001). The rate of transpiration changes as well – it may increase or decrease, depending on the mode of infection (SHTIENBERG 1992, BASSANEZI et al. 1997). An increase in the rate of transpiration is caused by cuticle damage in infected leaves and damage to stomata due to which they remain open, as well as by increased permeability of cell membranes (BASSANEZI et al. 2002). A decrease in the rate of transpiration may be a consequence of stomatal closure and a reduction in air volume contained in plant tissues (resulting from the presence of hyphae), as well as of hypertrophy of the mesophyll tissue, clogging of conducting tissues and defoliation (BASSANEZI et al. 2002).

Under natural conditions, 12 to 54% of carbon taken in by plants during photosynthesis is released by the root system, which stimulates the activity of soil microbes (LYNCH, WHIPPS 1990). The activity of rhizosphere microorganisms is closely related to plant metabolism. They affect nutrient metabolism in the soil and root formation. Increased availability and uptake of minerals have a direct impact on photosynthesis. Rhizosphere microbes promote and stimulate nutrient uptake and transport in the plant (EL-SHATNAWI, MAKHADMEH 2001). Increasing microbial diversity of soils improves the overall health and productivity of plants. The lack of chemical control may result in excessive growth of pathogenic fungi, which in turn substantially decreases the yield and deteriorates its quality (SADOWSKI et al. 2006). Therefore, biological disease control is often applied as an alternative or in addition to chemical crop protection. The effectiveness of biological and microbiological agents used as biofertilizers could be increased by combining cultures of various specific antagonists (DAVELOS et al. 2004). Microbial inoculants (effective microorganisms) consist of around 70 species of microorganisms belonging to five groups, namely lactic acid bacteria, photosynthetic bacteria, actinomycetes, yeast fungi and filamentous fungi (VALARINI et al. 2003). The application of EM has a beneficial effect on soil texture and quality (KHALIQ et al. 2006). In Poland the biopreparation EM^{TM} is registered as a soil enhancer recommended for use in ecological farming (Certificate of Conformity no. Z/13/PR-20001/03/BP). It has been approved for use by the National Institute of Hygiene, and proven safe in terms of human and environmental health (Certificate no. PZH/HT-1448/2002).

The aim of the present study was to determine the effect of infections by fungal pathogens and different methods of EM application on the rate of photosynthesis and transpiration in pea under field conditions. The effectiveness of EM in controlling pea diseases was also estimated.

Materials and Methods

Field investigations were conducted during the years 2003-2005 at the Research and Experimental Station in Tomaszkowo near Olsztyn (NE Poland). An exact experiment was established on brown soil of quality class IVa and good rye complex (2003) developed from silt, brown soil of quality class IVa and good rye complex (2004) developed from medium silty loam, and brown soil of quality class IIIb and very good rye complex (2005) developed from light loam. Pea was grown with winter triticale (first and third year of the study) and spring barley (second year) as a fore crop. The experiment was performed in a randomized split-plot design, in four replications. Plot area was 16 m^2. The experimental factor was the method of EM application, i.e.:

1. control treatment (no effective microorganisms or crop protection chemicals);

2. chemical control (seed dressing T, fungicide Rovral Flo 250 SC, insecticide Owadofos 540 EC, herbicide Basagran 480 SL);

3. soil application of EM combined with chemical control;

4. seed dressing with EM combined with chemical control;

5. foliar application of EM combined with chemical control;

6. soil application of EM, seed dressing with EM and foliar application of EM.

Prior to soil application, effective microorganisms were proliferated as recommended by the manufacturer (Greenland). A 0.1% solution of effective microorganisms (1 dm^3 water: 1 cm^3 EM: 1 g saccharose) was stored in the dark at a temperature of around 20°C for 14 days. Wet seed dressing with a 0.2% EM solution was carried out for 30 min. The dose of EM solution used for soil and foliar application was 200 dm^3 ha^{-1}.

Gas exchange parameters

The rate of net photosynthesis *An* (μmol CO_2 m^{-2} s^{-1}) and the rate of molar transpiration *E* (mmol H_2Om^{-2} s^{-1}) were measured over the growing season, from the beginning of flowering to pod setting, using a Li-Cor 6400 gas analyzer (Portable Photosynthesis System, Licor, Lincoln, NB, USA). Readings were taken at several-day intervals on leaves of the medium storey. Each measurement was repeated 10 times.

Health of pea plants

The health status of the aboveground parts of pea plants was estimated at the flowering stage, using the modified Hillstrand and Auld scale (1982): 0 – no disease symptoms, 1 – infection rate of 1-10%, 2 – infection rate of 11-20%, 3 – infection rate of 21-30%, 4 – infection rate of 31-40%, 5 – infection rate of 41-60%, 6 – infection rate of 61-80%, 7 – infection rate of 81-90%, 8-9 – infection rate of 91-100%. The results provided a basis for calculating the infection index (*II*), as described by McKinney (ŁACICOWA 1969).

Statistical analysis

The results were processed statistically by analysis of variance (ANOVA) using STATISTICA software (Data Analysis Software System, ver. 6, StatSoft,

Inc. 2003 **www.statsoft.com**). The significance of differences between mean values was estimated by Duncan's test ($p = 0.05$ for infection index, $p = 0.01$ for gas exchange parameters). The strength of linear relationships between variables was determined by Pearson correlation analysis. The coefficient of Pearson correlation (R) ($p = 0.05$) was calculated with the use of STATISTICA 6.1.

Results

Pea Fusarium wilt was observed each year, but its severity was greater in 2003 and 2005, when the infection index reached 8.0% and 8.3% respectively (Table 1). In 2004 disease intensity was substantially lower (*II* = 4.8%). Pea plants were also attacked by pathogens causing ascochytosis (Table 2, Table 3). Its greater severity was observed in 2004, when infections rates reached 18.8% for leaves and 7.3% for pods. Symptoms of downy mildew (*Peronospora viciae*) were noted only in 2005 (Table 4). The incidence of all three diseases was significantly affected by the method of EM application. The highest intensity of Fusarium wilt was recorded in the control treatment (*II* = 11.1%) (Table 1), while the lowest in plots where effective microorganisms were applied without chemical control (*II* = 5.3%) and where spraying with EM was combined with chemical control (*II* = 5.4%). The disease was also effectively controlled in chemically-protected plots (*II* = 5.8%). In the case of downy mildew the highest infection rates were recorded in the control treatment (*II* = 5.7%) (Table 4), while the lowest in plots where soil application of EM was combined with chemical protection (*II* =4.3%), which enabled to reduce disease severity by 24.6%, compared to the check. The intensity of ascochytosis was also the highest in the control treatment, with regard to both pea leaves

Table 1

The occurrence of Fusarium root (Complex of *Fusarium* spp.) on pea (av. of *II* significant, %)

Experimental combination	Year			Average
	2003	2004	2005	
*K***	10.5	7.0	15.8	11.1
G	9.00	3.75*	6.75*	6.5*
Z	8.75	7.75	8.00*	8.2*
O	5.75*	4.50	6.00*	5.4*
GZO	7.00*	2.00*	6.75*	5.3*
FIH	7.00*	4.00*	6.50*	5.8*
Average	8.0	4.8*	6.5	

* average of *II* significant at $\alpha = 0.05$

*K*** – control, *G* – soil treated with Effective Microorganisms (EM), *Z* – seeds treated with EM, *O* – foliage treated with EM, *GZO* – soil, foliage and seeds treatment with EM, *FIH* – pesticide control

Table 2

The occurrence of Ascochyta Complex (*Ascochyta pisi*, *Mycosphaerella pinodes*, *Phoma medicaginis* var. *pinodella*) on leaf pea (av. of *II* significant, %)

Experimental combination	Year			Average
	2003	2004	2005	
*K***	0.0	26.5	16.8	14.4
G	0.0	12.75*	17.0	9.9*
Z	0.0	21.25*	13.8	11.7*
O	0.0	23*	13.0*	12.0*
GZO	0.0	11*	10.25*	7.1*
FIH	0.0	18.5*	15.8	11.4*
Average	0.0*	18.8	14.4*	

* average of *II* significant at $\alpha = 0.05$
** description under Table 1

Table 3

The occurrence of Ascochyta pod spot (*Ascochyta pisi*, *Mycosphaerella pinodes*, *Phoma medicaginis* var. *pinodella*) (av. of *II* significant, %)

Experimental combination	Year			Average
	2003	2004	2005	
*K***	0.0	11.75	7.3	6.3
G	0.0	2.50*	4.25*	2.3*
Z	0.0	9.25*	2.25*	3.8*
O	0.0	11.0	3.50*	4.8*
GZO	0.0	3.00*	2.50*	1.8*
FIH	0.0	6.50*	3.00*	3.2*
Average	0.0*	7.3	3.8*	7

* average of *II* significant at $\alpha = 0.05$
** description under Table 1

Table 4

The occurrence of downy mildew (*Peronospora viciae*) on pea leaf (av. of *II* significant, %)

Obiekt	Year			Average
	2003	2004	2005	
*K***	0.0	0.0	17.0	5.7
G	0.0	0.0	12.75*	4.3*
Z	0.0	0.0	14.50*	4.8
O	0.0	0.0	16.25	5.4
GZO	0.0	0.0	13.50*	4.5*
FIH	0.0	0.0	15.25*	5.1
Average	0.0*	0.0*	14.8	

* average of *II* significant at $\alpha = 0.05$
** description under Table 1

(*II* =14.4%) and pods (*II* = 6.3%) (Table 2, Table 3). The lowest infection rates were noted when effective microorganisms were applied without chemical control (7.1% and 1.8% for leaves and pods respectively).

The rate of photosynthesis (*An*) was significantly different in particular years of the study, reaching the highest level (16.7 μmol CO_2 m^{-2} s^{-1}) in 2004 and the lowest (14.1 μmol CO_2 m^{-2} s^{-1}) in 2005 (Table 5). Net photosynthesis values were significantly affected by the method of EM application. The highest rate of photosynthesis (16.7 μmol CO_2 m^{-2} s^{-1}) was recorded in plots where foliar application of EM was combined with chemical protection as well as in those where EM were applied without chemical control (16.5 μmol CO_2 m^{-2} s^{-1}) (Table 5). The lowest rate of photosynthesis (13.7 μmol CO_2 m^{-2} s^{-1}) was observed in plots where seed dressing with EM was combined with chemical protection.

Table 5

Value of net photosynthetic rate *An* (μmol CO_2 m^{-2} s^{-1}) after aplication of EM

Obiekt	Year			Average
	2003	2004	2005	
*K****	14.8	20.2	13.0	16.0
G	16.78**	14.85**	14.59**	15.40**
Z	13.8	15.13**	12.3	13.71**
O	16.3	18.97**	14.84**	16.68**
GZO	17.62**	16.98**	14.93**	16.5
FIH	15.1	13.99**	14.91**	14.67**
Average	15.7*	16.7	14.1**	

* average of *II* significant at α = 0.05
** average of *E* significant at α = 0.01
*** description under Table 1

Table 6

Value of transpiration rate *E* (mm H_2O m^{-2} s^{-1}) after aplication of EM

Obiekt	Year			Average
	2003	2004	2005	
*K****	10.45	6.53	2.88	6.62
G	11.58**	5.91	2.99	6.83
Z	9.58	5.23**	2.46*	5.77**
O	11.03	4.85**	2.87	6.25
GZO	12.08**	5.94	2.82	6.95
FIH	10.01	4.70**	3.21*	5.97**
Average	10.27	6.13**	2.89**	

* average of *II* significant at α = 0.05
** average of *E* significant at α = 0.01
*** description under Table 1

Table 7

Simple correlations between the gas exchange (*An* and *E*) and Infection Index (*II*) of diseases

Disease	Gas exchange	*R*	R^2
Fusarium root root	*An*	-0.20	0.04
	E	0.09	0.01
Downy mildew	*An*	-0.52*	0.27*
	E	-0.74**	0.54*
Acochyta complex (leaf)	*An*	-0.03	0.001
	E	-0.76**	0.58**
Acochyta complex (pod spot)	*An*	0.19	0.034
	E	-0.50*	0.25*

* *R* significant at $\alpha = 0.05$
** *R* significant at $\alpha = 0.01$

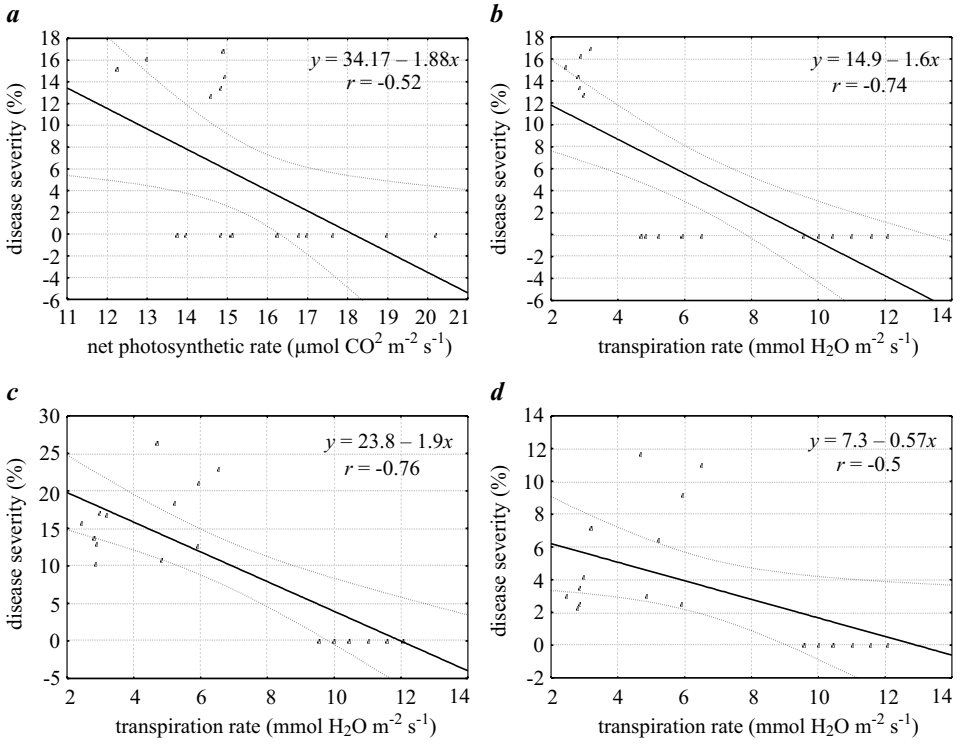

Fig. 1. Linear regression between: *a* – photosynthetic rate and disease severity of downy mildew (*II*); and transpiration rate and disease severity, *b* – downy mildew, *c* – Ascochyta leaf spot, *d* – Ascochyta pod spot

The rate of molar transpiration (*E*) also differed significantly in particular years, ranging from 10.79 (mmol H_2Om^{-2} s^{-1}) in 2003 to 2.87 (mmol H_2Om^{-2} s^{-1}) in 2005 (Table 6). Seed dressing with EM combined with chemical control substantially reduced transpiration values, to *E*=5.77 (mmol H_2Om^{-2} s^{-1}) and

E=5.97 (mmol H_2Om^{-2} s^{-1}), compared to the check. Over the experimental period pea plants responded to biological and chemical disease control by changes in the transpiration process.

The statistical analysis revealed that the rates of photosynthesis and transpiration were affected not only by the method of crop protection, but also by disease incidence. In 2005 the occurrence of downy mildew on pea leaves significantly decreased the rate of photosynthesis. The coefficient of Pearson correlation between photosynthesis rate and the severity of this disease was R = -0.51 (Table 7, Figure 1a). The values of *An* were also lower in Fusarium wilt-infected plants, but this trend was not confirmed by a statistical analysis (R = -0.20). There was no significant correlation between the values of *An* and ascochytosis severity (R = – 0.03). A strong correlation was recorded between the rate of molar transpiration E and infestation by fungal pathogens. A decrease in the values of E (R = -0.74) was noted in the case of infection by *Peronospora viciae* (Table 7, Figure 1b). The occurrence of ascochytosis on pea leaves and pods also reduced the rate of transpiration, which was reflected in the values of the coefficient of Pearson correlation: R = -0.74 and R = -0.50, respectively (Table 7, Figure. 1c, Figure 1d).

Discussion

The effect of plant diseases on the rate of photosynthesis and other parameters of gas exchange has been widely discussed by numerous authors. In the majority of experiments conducted to date plants were inoculated under controlled conditions. In such cases infection severity is dependent on the amount of inoculum applied to plants and on the pathogenicity of isolates. The present study was performed under conditions of natural infection, to determine the interactions between the experimental factors and the impact of the natural environment, considered important for agricultural practice. In this experiment the rate of photosynthesis was significantly affected by the degree of pea infestation by *Peronospora viciae* (coefficient of Pearson correlation R = -0.51). The phenomenon of photosynthesis inhibition by pathogens damaging leaf tissue has been already described in professional literature (GOICOECHEA et al. 2001, ROBERT et al. 2004).

It was found that other pathogenic factors had no significant influence on photosynthesis values. The results reflect the interactions between a variety of factors affecting the process of photosynthesis and environmental impacts on infection levels. Pea infestation by fungal pathogens was relatively low, due to weather conditions and the use of the tested biopreparation (Figure 2a, Figure 2b). According to MARCINKOWSKA (1997), the incidence

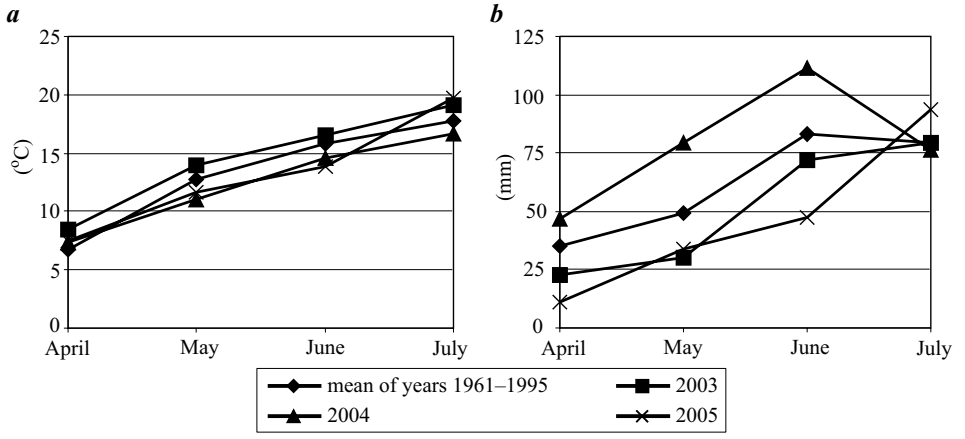

Fig. 2. Pattern of weather conditions in growing periods (data according to the Meteorolgical Station in Tomaszkowo)

of leaf-and-pod spot of peas is closely related to weather conditions during the growing season, since the spread of this disease is observed at relative air humidity of 95%.

The greatest severity of Fusarium wilt of peas is recorded during drought and at high temperatures, which accelerate the development of this disease (MAJCHRZAK 1998, PAŃKA, SADOWSKI 1999). In the current study the highest intensity of Fusarium wilt was noted in the first and third year. This period was characterized by high temperatures and low precipitation (Figure 2a, Figure 2b), which promoted the spread of the disease.

Downy mildew of peas is favored by cool, wet weather, and survives as oospores on infected seeds until the next year (FALLOON et al. 2000). In this experiment symptoms of downy mildew were observed in 2005, when precipitation rates were high (Figure 2b). Transpiration values *E* were considerably affected by fungal pathogens. The occurrence of all pathogens causing leaf tissue infections reduced the rate of transpiration in pea plants. It seems that infection resulted in the closure of stomata, to prevent water loss from the plant. Necrotrophic pathogenic factors may cause stomatal closure over the entire surface of leaf blades even if infection severity is low, which in turn decreases the rate of photosynthesis (MEYER et al. 2001) and affects transpiration. In the case of this group of pathogens a reduction in the rate of transpiration in infected plants is proportional to leaf lesion area (SHTIENBERG 1992). In the case of biotrophic pathogenic factors transpiration inhibition is related to the damaged leaf area to a lesser degree.

SHTIENBERG (1992) demonstrated that transpiration values increased in plants attacked by rust caused by a biotrophic pathogen. In the present study

another biotrophic pathogen – *Peronospora viciae* – was identified only in 2005, when weather conditions favored disease development. Constant water deficits doubtlessly affected the defense strategy of plants, part of which was stomatal closure. This effect could be enhanced even by a low level of infection.

Literature data on the impact of biological disease control on gas exchange parameters are scant. In this experiment the highest rate of photosynthesis was recorded following foliar application of effective microorganisms. Similar results were reported by XU et al. (2000), who observed a higher rate of photosynthesis in treatments where inoculants containing effective microorganisms were applied, compared to the control treatment. Current results confirmed the impact of effective microorganisms on the occurrence of diseases associated with the soil environment, and leaf diseases. It was also found that all methods of EM application reduced disease incidence in pea plants. Good results were obtained in the case of biological disease control, applied alone or combined with chemical methods. ERRAMPALLI and BRUBACHER (2006) demonstrated that a combination of biological and chemical pathogen control allowed to increase the effectiveness of plant protection. However, it is important to properly select biological and chemical agent to be applied together, so that to make sure that the active substances contained in crop protection chemicals do not inhibit the growth of microbial antagonists introduced into the soil (FRAVEL et al. 2005).

Conclusions

1. The tested biological agent (EM) reduced the incidence of pea diseases.
2. Foliar application of EM significantly increased the rate of photosynthesis in pea.
3. Soil application of EM, seed dressing and chemical control decreased the rate of photosynthesis in pea.
4. Seed dressing with the tested biological agent (EM) and chemical control caused a significant decrease in molar transpiration values in pea.
5. The occurrence of downy mildew of peas significantly reduced the rate of photosynthesis.
6. The occurrence of downy mildew and ascochytosis of peas decreased the rate of molar transpiration.

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References

BASSANEZI R.B., AMORIM L., BERGAMIN FILHO A. 2001. *Eficiencia fotossintetica de folhas de feijoeiro infectadas com o virus do mosaico- em-desenho, Uromyces appendiculatus e Phaeoisariopsis griseola*. Summa Phytopatologica, 27: 5-11.

BASSANEZI R.B., AMORIM L., BERGAMIN FILHO A., BERGER R.D. 2002. *Gas exchange and emission of chlorophyll fluorescence during the monocycle of rust, angular leaf spot and anthracnose of bean leaves as a function of their trophic characteristics*. Journal of Phytopathology, 150: 37-47.

BASSANEZI R.B., MARTINS M.C., GODOY C.V., AMORIM L., BERGAMIN FILHO A. 1997. *Efeito da antracnose na eficiência fotossintética do feijoeiro*. Fitopatologia Brasileira, 22: 520-524.

DAVELOS A.L., KINKEL L.L., SAMAC D.A. 2004. *Spatial variation in frequency and intensity of antibiotic interactions among Streptomycetes from prairie soil*. Appl. Environ. Microbiol., 70: 1051-1058.

EL-SHATNAWI M.K.J., MAKHADMEH I.M. 2001. *Ecophysiology of the Plant-Rhizosphere System*. J. Agronomy & Crop Science 187: 1-9.

ERRAMPALLI D., BRUBACHER N.R. 2006. *Biological and integrated control of postharvest blue mold (Penicillium expansum) of apples by Pseudomonas syringae and cyprodinil*. Biological Control. 36: 49-56.

FALLOON R.E., FOLLAS G.B., BUTLER R.C., GOULDEN D.S. 2000. *Resistance in Peronospora viciae to phenylamide fungicides: reduced efficacy of seed treatments of pea (Pisum sativum) and assessment of alternatives*. Crop Protection, 19: 313-325.

FRAVEL D.R., DEAHL K.L., STOMMEL J.R. 2005. *Compatibility of the biocontrol fungus Fusarium oxysporum strain CS-20 with selected fungicides*. Biol. Control, 34: 165-169.

GOICOECHEA N., AGUIRREOLEA J.S., GARCIA-MINA J.M. 2001. *Gas exchange and flowering in Verticillium-wilted pepper plants*. J. Phytopathology, 149: 281-286 .

GRZESIUK S., KOCZOWSKA I., GÓRECKI R.J. 1999. *Fizjologiczne podstawy odporności roślin na choroby*. Wyd. UWM, Olsztyn.

HILLSTRAND D.S., AULD D.J. 1982. *Comparative evaluation of four techniques for screening winter peas for resistance to Phoma medicaginis var. pinodella*. Crop Sci., 22(2): 282-287.

KHALIQ A., KALEEM ABBASI M., HUSSAIN T. 2006. *Effects of integrated use of organic and inorganic nutrient sources with effective microorganisms (EM) on seed cotton yield in Pakistan*. Bioresource Technology, 97: 967-972.

LYNCH J.M., WHIPPS J.M. 1990. *Substrate flow in the rhizosphere*. Plant Soil, 129: 1-10.

ŁACICOWA B. 1969. *Metoda laboratoryjna szybkiej oceny odporności jęczmienia na Helminthosporium sativum*. Biul. IHAR, 3(4): 61-62.

MAJCHRZAK B. 1998. *Wpływ stresu chłodno-wodnego na kiełkowanie nasion i zdrowotność siewek wybranych roślin strączkowych*. Rozpr. Monogr. ART., Olsztyn, ss. 1-56.

MARCINKOWSKA J. 1997. *Zdrowotność grochu uprawianego na suche nasiona*. Biuletyn IHAR. 201: 279-287.

MCELRONE A.J., FORSETH I.N. 2004. *Photosynthetic Responses of a Temperate Liana to Xylella fastidiosa Infection and Water Stress*. J. Phytopathology, 152: 9-20.

MEYER S., SACCARDY-ADJI K., RIZZA F., GENTY B. 2001. *Inhibition of photosynthesis by Colletotrichum lindemuthianum in bean leaves determined by chlorophyll fluorescence imaging*. Plant Cell Environ., 24: 947-956.

PAŃKA D., SADOWSKI C. 1999. *Pathogenicity of Fusarium solani isolates excised from different plants infecting pea (Pisum sativm L.) and faba bean (Vicia faba ssp. minor Harz.).* Phytopathologia Polonica, 18: 81-93.

PINKARD E.A., MOHAMMED C.L. 2006. *Photosynthesis of Eucalyptus globules with Mycosphaerella leaf disease*. New Phytologist, 170: 119-127.

ROBERT C., BANCAL M.O., NICOLAS P., LANNOU C., NEY B. 2004. *Analysis and modelling of effects of leaf rust and Septoria tritici blotch on wheat growth*. Journal of Experimental Botany, 55: 1079-1094.

SADOWSKI C., LENC L., KORPAL W. 2006. *Out of investigations on vegetable seed coating with Trichoderma viride and plant health in organic system*. J. Research and Applications in Agricultural Engineering, 51(2): 150-153.

SCHOLES J.D., LEE P.J., HORTON P., LEWIS D.H. 1994. *Invertase: understanding changes in the photosynthetic and carbohydrate metabolism of barley leaves infected with powdery mildew*. New Phytologist, 126: 213-222.

SHTIENBERG D. 1992. *Effects of foliar diseases on gas exchange processes: a comparative study*. Phytopathology, 82: 760-765.

VALARINI P.J., ALVAREZ M.C.D., GASCO J.M., GUERRERO F., TOKESHI H. 2003. *Assessment of soil properties by organic matter and EM – microorganisms incorporation*. R. Bras. Ci. Solo, 27: 519-525.

XU H.L, WANG R, MRIDHA M.A.U., KATO S., KATASE K., UMEMURA H. 2000. *Effect of organic fertilization and EM inoculation on leaf photosynthesis and fruit yield and quality of tomato plants*. In Proceedings of the 6th International Conference on Kyusei Nature Farming, South Africa, 1999 Senanayake, Y.D.A. and Sangakkara U.R.