EFFECTS OF THE TREATMENT WITH FUNGICIDES ON THE INTENSITY OF THE PHYSIOLOGICAL PROCESSES IN *Cucumis sativus* L. PLANTS ATTACKED BY *Pseudoperonospora cubensis* (Berk. et Curt.) Rostov

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Abstract. The physiological research regarding the effects of the treatment with fungicides on the intensity of the physiological processes has been carried in cucumber plants (*Cucumis sativus* L.) cultivated in the Oltenia region. After applying the treatment with fungicides, there were carried out physiological analysis, on the 10th of July 2012, in the attacked plants and also in the plants treated with fungicides. The research carried out on the plants attacked by the pathogen, in comparison to the plants treated with fungicides, emphasized that these presents lower values of photosynthesis and transpiration intensity due to the reduction of the assimilation surface and the malfunctioning of the closing and opening mechanisms of stomata, as a result of the formation of the spots, which, at the beginning, are yellow, then brown, and to the necrosis of the tissue corresponding to the spots. In the plants attacked by the pathogen, it was observed a decrease of the chlorophyll content and the decrease of the water content, which caused hydric and metabolic imbalances in the attacked plants, in comparison with the plants analysed after the treatment with fungicides.

Keywords: cucumber plants, fungicide, pathogen, photosynthesis, transpiration.

Rezumat. Efectele tratamentului cu fungicide asupra intensităţii proceselor fiziologice la plantele *Cucumis sativus* L. atacate de *Pseudoperonospora cubensis* (Berk. et Curt.) Rostov. Cercetările fiziologice privind efectele tratamentului cu fungicide s-au efectuat la plantele de castraveţi (*Cucumis sativus* L.) cultivate în zona Olteniei. După aplicarea tratamentului cu fungicide s-au efectuat analize fiziologice, la data de 10 iulie 2012, la plantele atacate de patogen, dar şi la plantele tratate cu fungicide. În urma cercetărilor efectuate la plantele atacate de patogen, în comparaţie cu plante tratate cu fungicide, se observă că acestea prezintă valori mai mici ale intensităţii fotosintezei şi transpiraţiei datorită reducerii suprafeţei de asimilare şi dereglării mecanismelor de închidere şi deschidere a stomatelor, ca rezultat al formării petelor la început de culoare gălbuie, apoi brune şi necrozării tesuturilor corespunzătoare petelor. În plantele atacate de patogen s-a observat scăderea conţinutului de clorofilă şi scăderea conţinutului de apă, fapt care a determinat dezechilibre hidrice şi metabolice la plantele atacate, în comparaţie cu plantele analizate după tratamentul cu fungicide.

Cuvinte cheie: plante de castraveţi, fungicid, patogen, fotosinteză, transpiraţie.

INTRODUCTION

The genus *Cucumis*, which includes cucumber (*Cucumis sativus*) and melon (*Cucumis melo*), has numerous wild African species, and it has therefore been assumed that melon originated in Africa. For cucumber, this seemed less likely because wild cucumbers exist in India and a closely related species lives in the Eastern Himalayas. Using DNA sequences from plastid and nuclear markers for some 100 *Cucumis* accessions from Africa, Australia, and Asia, we show here that melon and cucumber are of Asian origin and have numerous previously overlooked species-level relatives in Australia and around the Indian Ocean (SEBASTIAN et al., 2010).

Vegetables are plants known for their nutritional value and therapeutic effects. Vegetable consumption stimulates body hydration, muscle activity, appetite, calcification, enzyme activity, defense ability, inhibits bacterial fermentation, and supplies essential amino acids (SUCIU et al., 1987).

Pseudoperonospora cubensis is a devastating, worldwide-distributed disease of cucurbit crops in the open field and under cover (LEBEDA & COHEN, 2011). Symptoms vary on different cucurbit species and varieties, specifically in terms of lesion development, shape and size. Infection of cucurbits by *Pseudoperonospora cubensis* impacts fruit yield and overall plant health (SAVORY et al., 2011).

Fungicides remain a vital solution to the effective control of plant diseases, which are estimated to cause yield reduction of almost 20 percent in major food and cash crop worldwide. Application of fungicides may affect crop physiology by various disruptions such as growth reduction, perturbation in the development of reproductive organs, alteration of nitrogen, and/or carbon metabolism leading to a lower nutrient availability for plant growth. The sensitivity of some plant species may depend on the developmental stage (more sensitive to the treatments at young stages or during critical events such as reproduction) or the type of pesticides used (PETIT et al., 2012).

It is expected that fungicides remain an essential tool for plant disease management and will continue to play a crucial role in optimizing yields from the world crops. Therefore, the development of new compounds with lower negative impact in plant physiology is a future challenge. This will provide benefits not only for plants yield but also for the environment and human health (DIAS, 2012).

The intensity of the process of photosynthesis varies depending on the species and age of the leaves. Thus, in the case of cucumber plants, basal leaves at senescence have reached a low photosynthetic intensity: 3.04 $\mu mol/m^2/s$. This process intensity increases in case of the leaves located in the upper third, at which, there has been determined a value of 7.10 $\mu mol/m^2/s$. Young leaves from the apical side of the plants have a very low photosynthetic intensity (1.62 $\mu mol/m^2/s$) - BURZO et al., 2000).

Positive correlations were established between the intensity of the physiological processes and the photosynthetic active radiation, the leaf temperature and stomatal conductance of CO_2 (NICOLAE, 2010).

Chlorophyll content is higher in the leaves analysed after the treatments with fungicides, compared to the leaves of the plants attacked by the pathogen; between chlorophyll content and photosynthesis intensity, there is a positive correlation (NICOLAE & BUŞE-DRAGOMIR, 2012).

The deployment of the normal physiological processes in the plant-host cells is disturbed by the attack produced by microorganism parasites and the presence of mycotoxins, which has consequently diminished the production and its quality. The primary effect of the pathogen attack occurs at the cellular level, but later, the whole plant metabolism changes; these changes manifest themselves in the transport of substances through the plant, in the process of raising, in the falling of the flowers and the fruits (BURZO et al., 1999).

MATERIAL AND METHODS

The physiological research on the effects of the treatment with fungicides in cucumber plants was performed at the *Mondial* variety cultivated in the climatic conditions of the Oltenia region.

Cucumis sativus L. is a widely cultivated plant in the family Cucurbitaceae. The plant is monoecious and the flowers are actinomorphic, corolla is yellow, campanulate, 5-parted with oblong to lanceolate lobes. The fruit are cylindrical usually with more than 90% water content. The cucumbers *Mondial* cultivar (early variety), produced by ICDLF Vidra, Romania, presents a vegetative apparatus, has medium to large and cylindrical fruits. The fruits are short, cornichon type (60-80 g) and have dark green colour.

After the identification of the attack produced by the pathogen, at the beginning of formation of the fruit, the cucumber culture has been divided into two groups. The first group of plants has treated twice with the fungicide *Shavit* F 72 WP at a period of 10 days between treatments (plants with treatment), and, in case of the second group, it has not been applied any treatment (plants without treatment). The physiological research has been carried out after a period of 12 days from the last treatment with fungicides (the 10^{th} of July 2012) and consisted in carrying out physiological analysis of pathogen attacked plants, compared to plants analysed after the treatment with fungicides.

The fungicide *Shavit* F 72 WP, produced by Makhteshim Agan-Israel, contains two active substances with systemic and contact action, which ensures a quick, preventative and curative of long duration effect (Triadimenol 20 g/kg + Folpet 700 g/kg). The protection period depends on the local conditions and varies from 7 to 14 days.

In case of the cucumber plants attacked by *Pseudoperonospora cubensis* (Berk. et Curt.) Rostov, the applied dose was 0.2% (20 g per 10 litres of water) and there were applied two treatments at an interval of 10 days.

The physiological processes intensity was carried out using ultra compact photosynthesis measurement system LCi (Ultra Compact Photosynthesis System - ADC BioScientific Ltd.). The obtained results were graphically represented and statistically interpreted using the Excel software.

The water contents and dry substance content were determined by the help of the drying stove - gravimetric method. The chlorophyll content was estimates by Minolta SPAD 502 chlorophyllmeter.

The estimate of the attack (frequency, intensity and degree of attack) produced by the pathogen was made using the calculation formulae elaborated by SĂVESCU & RAFAILĂ, 1978.

RESULTS AND DISCUSSIONS

In this paper we present the results of the research conducted on the physiological processes intensity of cucumber plants attacked by pathogens, in comparison with cucumber plants in case of which there were performed treatments with fungicide. After applying the treatments, the specific symptoms of the attack produced by pathogens were reduced and then stopped, which showed the effectiveness of the fungicide applied in the fight against the disease with positive effects on the production of cucumbers.

In recent years, downy mildew of cucumber has produced very large losses in different areas of the country, often compromising the crop.

The attack is manifested by the appearance of some yellowish spots, bounded by ribs on the upper epidermis of the leaves spots that later become brown. On the bottom of the leaf, in the spots, it can be observed the appearance of a greyish-purple fluff, consisting of sporangiophores with sporanges (MITREA, 2006) - Fig. 1.

In favourable conditions, the spots expand and join together, cover entirely the leaf surface, which dries and the plants lose their leaves.

Pseudoperonospora cubensis presents unicellular mycelium, branched intercellular. Sporangiophores are hyaline, in the upper third dichotomous branching. The terminal end presents sterigma with ellipsoidal or ovate sporanges, unicellular, grey-yellowish colour (TĂNASE & ŞESAN, 2006) - Fig. 2.

Through the application of the treatment with fungicides, the foliar surface faded or necrosis was significantly reduced, and the leaves with specific symptoms of the attack produced by the pathogen maintained green during the period of vegetation.

The physiological research was performed at the cucumber plants attacked by the pathogen, compared to plants that have been subjected to the fungicide treatments.

Figure 1. The *Cucumis sativus* L. attacked by *Pseudoperonospora cubensis* (original).

Figure 2. *Pseudoperonospora cubensis* - sporangiophores with sporanges (oc. 10 x ob. 20) (original).

The estimation of the attack produced by *Pseudoperonospora cubensis* (Berk. & Curt.) Rostov at the attacked cucumber plants, on the 10th of July 2012, is presented in figure 3.

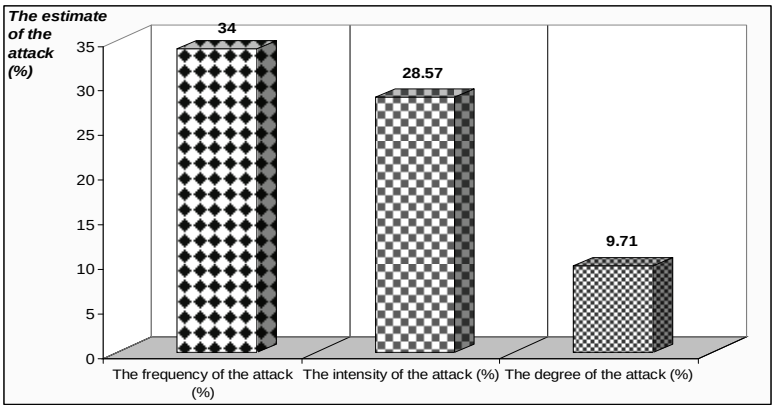

Figure 3. The estimate of the attack produced by *Pseudoperonospora cubensis* in *Cucumis sativus* L.

The diurnal dynamics of the physiological processes in the attacked plants is similar to the diurnal dynamics of the plants have been subjected to fungicide treatments, but presents lower values as a result of the reduction of the assimilation surface by the appearance of spots, yellowish at the beginning, then brown, by the necrosis of the tissues, malfunctioning of stomata closing and opening mechanisms under the action of the pathogen (Figs. 4; 5).

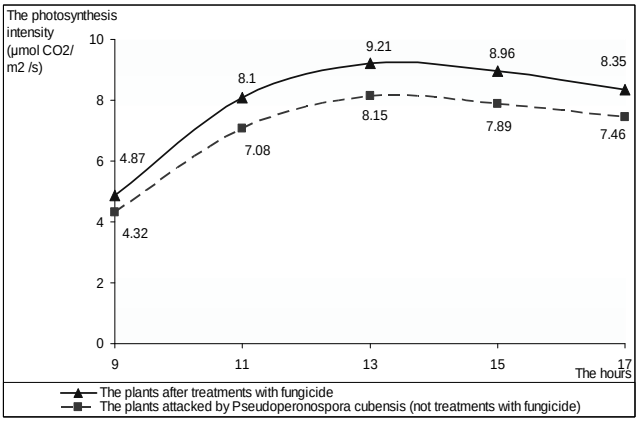

Figure 4. Diurnal dynamics of the photosynthesis intensity in cucumber plants (*Cucumis sativus* L.).

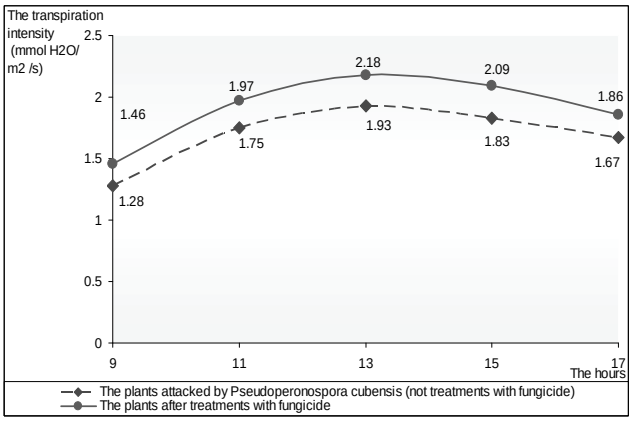

Figure 5. Diurnal dynamics of the transpiration intensity in cucumber plants (*Cucumis sativus* L.).

The physiological processes intensity is correlated with the physiological parameters; thus, it was established a strong association between these. It can be observed an increase of the physiological parameters, during the day, increase starting in the morning; they present higher values at lunch and a gradually decrease towards the evening (Table 1).

Table 1. Diurnal dynamics of the physiological parameters registered in cucumber plants.

The physiological parameters in the cucumber plants		**The hours of performed analyses**				
		9^{00}	**11^{00}**	**13^{00}**	**15^{00}**	**17^{00}**
The photosynthetic active radiation (µmol / m^2 / s)	The plants after the treatments with fungicide	1270	1459	1671	1592	1548
	The plants attacked by the pathogen	1260	1432	1665	1560	1535
The leaf temperature (^oC)	The plants after the treatments with fungicide	25.6	28.5	33.1	32	30.7
	The plants attacked by the pathogen	25.8	28.6	33.4	32.2	31
The stomatal conductance (mol / m^2 / s)	The plants after the treatments with fungicide	0.05	0.08	0.1	0.09	0.08
	The plants attacked by the pathogen	0.03	0.06	0.09	0.07	0.06

The linear regression performed between the photosynthesis intensity and photosynthetic active radiations present on the surface of the leaves shows a good positive correlation between these; the coefficient of determination (R^2) is 0.89 for the cucumber plants attacked by the pathogen and 0.92 for the cucumber plants analysed after the treatments with fungicide. The linear regression performed between the transpiration intensity and photosynthetic active radiations also shows a good positive correlation; the coefficient of determination (R^2) is 0.85 for the cucumber attacked plants and 0.87 for the cucumber plants analysed after the treatments with fungicide (Figs. 6; 7).

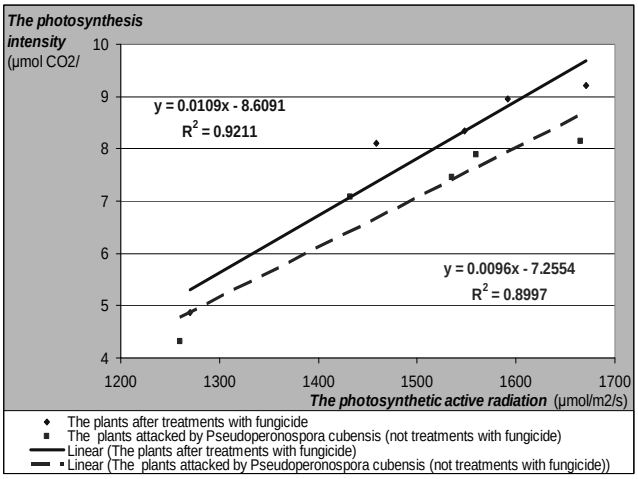

Figure 6. The correlation between the photosynthesis intensity and the photosynthetic active radiation *Cucumis sativus* L.

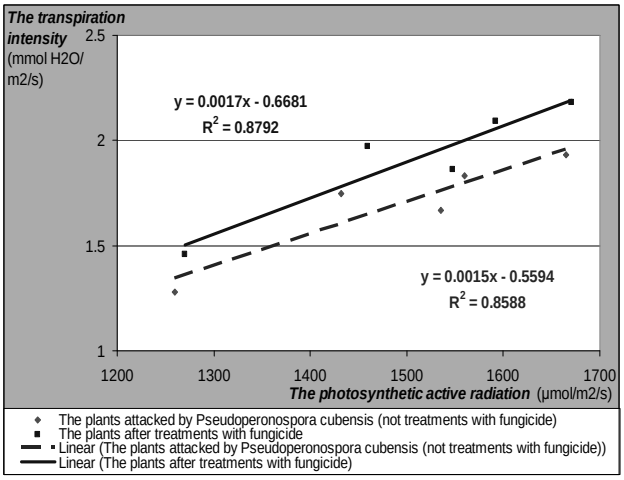

Figure 7. The correlation between the transpiration intensity and the photosynthetic active radiation in *Cucumis sativus* L.

The linear regression performed between the photosynthesis intensity and leaf temperature shows a positive correlation; the coefficient of determination (R^2) is 0.86 for the cucumber attacked plants and 0.87 for the cucumber plants after the treatments with fungicide. The linear regression performed between the transpiration intensity and leaf temperature shows a positive correlation between these; the coefficient of determination (R^2) is 0.80 for the cucumber attacked plants and 0.83 for the cucumber plants after the treatments with fungicide (Figs. 8; 9).

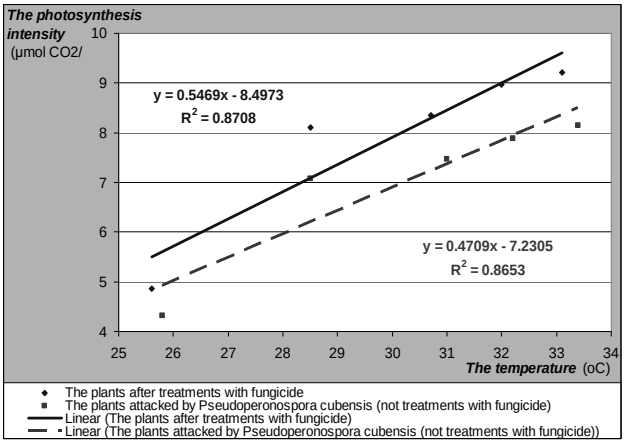

Figure 8. The correlation between the photosynthesis intensity and the leaf temperature in *Cucumis sativus* L.

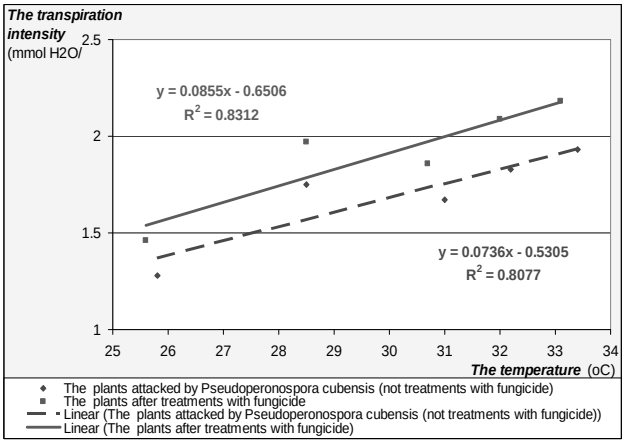

Figure 9. The correlation between the transpiration intensity and the leaf temperature in *Cucumis sativus* L.

The linear regression carried out between the photosynthesis intensity and stomatal conductance of CO_2 shows a positive correlation; the coefficient of determination (R^2) is 0.85 for the cucumber attacked plants and 0.94 for the cucumber plants after the treatments with fungicide. The linear regression carried out between the transpiration intensity and stomatal conductance of CO_2 shows a good positive correlation; the coefficient of determination (R^2) is 0.93 for the cucumber attacked plants and 0.97 for the cucumber plants after the treatments with fungicide (Figs. 10; 11).

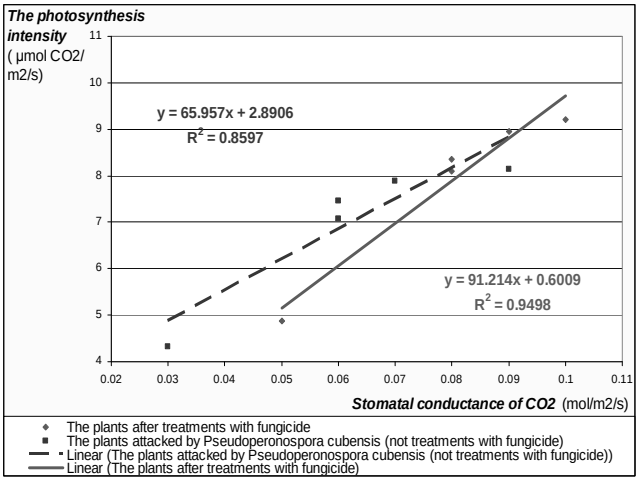

Figure 10. The correlation between the photosynthesis intensity and the stomatal conductance in *Cucumis sativus* L.

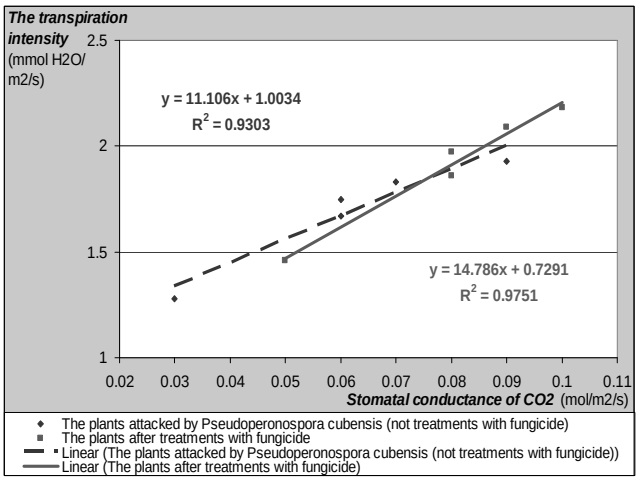

Figure 11. The correlation between the transpiration intensity and the stomatal conductance in *Cucumis sativus* L.

The cucumber plants attacked by *Pseudoperonospora cubensis* present a 2.63 % lower water content and a 22.31 % higher dry substance content in comparison with the cucumber plants analysed after the treatments with fungicide; this fact causes hydric and metabolic imbalances (Fig. 12).

In the plants attacked by the pathogen, there can be observed a decrease of the chlorophyll content by 20.35 % as a result of the deterioration of the chlorophyll, appearance of spots and necrosis on the leaves, this correlating with the decrease of the photosynthesis intensity (Fig. 13).

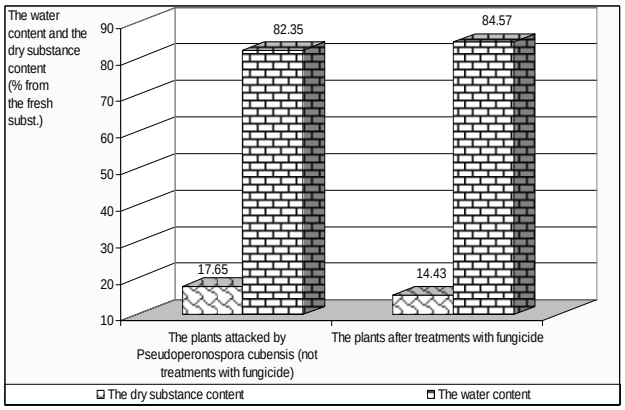

Figure 12. The water content and the dry substance content in the leaves of cucumber plants (*Cucumis sativus* L.).

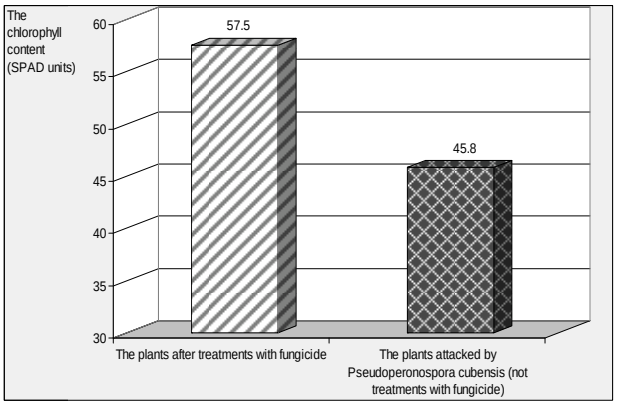

Figure 13. The chlorophyll content in the leaves of cucumber plants (*Cucumis sativus* L.).

CONCLUSIONS

In this paper we present the effects of the treatment with fungicides on some physiological processes of the cucumber plants, compared with the cucumber plants without treatment with fungicide, with implications in the processes of growth and development of plants.

In the cucumber plants attacked by the *Pseudoperonospora cubensis*, in comparison with the plants analysed after the treatments with fungicide, it was noticed that photosynthesis and transpiration intensity is lower as a result of the reduction of the assimilation surface induced by the appearance of spots, coloured yellowish at the beginning, then brown, of the necrosis of the tissues and also malfunctioning of stomata closing and opening mechanisms under the action of the pathogen. At the analysed plants, according to the climatic conditions, it has been established a positive correlations between the physiological processes intensity and photosynthetic active radiation, leaf temperature and stomatal conductance.

In the cucumber plants attacked by the pathogen it was observed a decrease of the chlorophyll content and the decrease of the water content, in comparison with the plants analysed after the treatments with fungicide; this is manifested through the hydric and metabolic imbalances.

At cucumber plants attacked by the pathogen, after the application of the treatments with fungicides, in comparison with the plants without treatments with fungicides, the surface of the leaf affected by the pathogen has been reduced, the damaging action of the agent being stopped, which influenced positively the growth and further development of the plants.

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Received: March 11, 2014
Accepted: May 10, 2014