PHYSIOLOGICAL RESPONSE AND YIELD OF PEPPER PLANTS (CAPSICUM ANNUM L.) TO ORGANIC FERTILIZATION

ФИЗИОЛОГИЧЕН ОТГОВОР И ПРОДУКТИВНОСТ НА ПИПЕРОВИ РАСТЕНИЯ (CAPSICUM ANNUM L.) ПРИ ОРГАНИЧНО НАТОРЯВАНЕ

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ABSTRACT

The experience was brought out to the experimental field of the Agroecological Centre at the Agricultural University of Plovdiv which owns a Certificate of ecological farm work. The research was carried out on two pepper cultivars: Gorogled 6 and Buketen 50 – intended for production of red pepper for grinding. For the purpose of the experiment a bio-fertilizer, produced by the Californian earthworm Lumbricus rubellus was used. Two levels of the bio-fertilizer were applied - 50 and 100 ml/per plant.

It was determined that the bio-fertilizer speeds up plant growth. It influences the growth rate of the roots and stems and affects the formation of the foliage. The bio-fertilizer has a positive effect upon the functional activity of the photosynthetic apparatus /increased content of photosynthetic pigments, improved leaf gas exchange/. The bio-fertilizer also influences the yield (raw material for drying). The highest effect has been achieved at application of 100 ml fertilizer to the Gorogled 6 cultivar.

KEY WORDS: leaf gas exchange, Capsicum annuum L., earthworm (Lumbricus rubellus), yield

РЕЗЮМЕ

Полският опит беше изведен на опитно поле в Агроекологичния център при АУ-Пловдив, който притежава сертификат за екологична ферма. В изследването бяха включени два сорта пипер: Гороглед 6 и Букетен 50 - предназначени за производство на червен пипер за мелене. За извеждане на опита беше използван биотор произведен от калифорнийския червей Lumbricus rubellus. В изследването бяха включени две нива от биотора – 50 и 100 ml/растение.

Въз основа на получените резултати беше установено, че подхранването с Лумбрикал ускорява растежа на растенията. Оказва влияние върху темпа на нарастване на корените и стъблото, въздейства върху формирането на листната площ. Установен е положителен ефект от биотора върху функционалната активност на фотосинтетичния апарат /повишено съдържание на фотосинтетичните пигменти, подобрен листен газообмен/. Лумбрикал влияе въху формирането на добива (суровина за изсушаване). Най- висок ефект е установен при подхранване на растенията с 100 ml биотор – при сорт Гороглед 6.

КЛЮЧОВИ ДУМИ: листен газообмен, Capsicum annuum L., калифорнийски червей (Lumbricus rubellus), добив

INTRODUCTION

In the past years it has been found out that the intensive agriculture aiming at high yields has lead to a breach in the ecological equilibrium in the agricultural ecosystems. This imposes development of new ecologically consistent technologies which would guarantee the steady growth of agriculture [7, 12, 23, 25].

Almost all European countries have introduced an entirely new type of production technology which does not cause harm to the environment. Most often this new type of technology (organic agriculture) is defined as a system for maintenance of the natural fertility of the soil, biological diversity of the species and the ecological balance of the environment. This type of agriculture avoids or totally excludes the use of synthetic fertilizers, pesticides, growth regulators and food additives. It counts mostly on the role of the sowing alternations, the use of plant waste, manure and green fertilization, as well as the aspects of biological protection. A number of authors consider that organic production gives lower yields [2, 31], however at the same time the harvest is characterized by better taste qualities compared to the one obtained through conventional production [1, 20, 27].

For the procurement of optimal conditions for growth and development of plants under the conditions of organic production a complex of agro-technical events must be applied and the specific features of the separate cultivars must be taken into consideration.

One of the agro-technical events related to organic production is the use of bio-fertilizers in the ecological farms [22, 28, 29, 30]. Recently bio-fertilizers obtained from various earthworms have been studied intensely [6, 21, 26] and methods of their application in organic fertilization are sought after.

Bio-fertilizers, obtained as a result of composting of earthworm organic waste (vermicomposts) have a number of advantages compared to the manure - they are richer in nutritive macro- and microelements, vitamins, enzymes, hormones, etc. [15]. It was found out that earthworms increase the populations of the useful microorganisms [10] which for their part synthesize biologically active substances. In the process of composting humates are obtained which adsorb the biologically active substances [5, 9]. The obtained product has a high content of stable humus substances which are close to the soil humic acids. Bio-fertilizers not only improve soil fertility with their content of growth regulators but also have a stimulating effect upon plant growth and development [3, 4, 8, 13].

Two types of earthworms have obtained practical utilization: the Red Californian Worm (Lumbricus rubellus) and the Red Tiger Worm (Eisenia foetida). They consume various types of organic fertilizers and other organic waste material which during digestion undergo physical and chemical changes. The end product consists of worm excrements and is known under various trade names (Lumbrical, Biohumus, etc.). Bio-fertilizers have good preservation abilities. They do not suffer putrefactive processes and preserve the properties of a “live” product [3].

In many countries the production and sale of bio-fertilizers in agriculture is well organized. The considerable interest towards them ensues from the fact that by influencing the soil fertility, they are also a naturally consistent solution of the problem for preservation of the environment and prevention of the plant production from contamination.

The practical application of this type of bio-fertilizers has not been supported enough by comprehensive scientific information. This exactly motivates our research work.

MATERIAL AND METHODS

The field experiment was carried out in the experimental field of the Agroecological centre, holder of a certificate for ecological farm. The research was carried out on two pepper cultivars: Gorogled 6 and Buketen 50 – intended for production of red pepper for grinding. The seedlings were grown in a tunnel-shaped plastic cover without pricking out, at sowing quota 12-15g m^{-2}.

The plants were bedded on a permanent place after increase in the soil temperature to over 16 °C, on a profiled surface (long furrows), with planting scheme 60x15 cm. Each experiment had four repetitions, including 24 plants in each repetition.

For the purpose of the experiment a bio-fertilizer, produced by the Californian earthworm Lumbricus rubellus was used (Table 1).

Lumbrical was placed at the bottom of previously formed furrows with 6-8 cm depth.

Table 1. Chemical specifications of the bio-fertilizer Lumbrical
Таблица 1. Химични характеристики на биотор Лумбрикал

Dry subst. [%]	pH	Hum. [%]	Total N [%]	NH_4N [ppm]	NO_3N [ppm]	Mov. min.N [ppm]	Total P [%]	Mov. P_2O_5 [ppm]	Total K [%]	Mov K_2O ppm
58.5	7.3	32.56	1.40	14.0	156.8	170.8	1.12	325	1.11	498

The experiment was carried out under the following scheme:

- Control – soil;
- Soil + biological fertilizer Lumbrical 50 ml per plant;
- Soil + biological fertilizer Lumbrical 100 ml/plant;

The harvest was carried out only once, when over 80% of the fruits have reached complete botanical ripeness. The unimpaired fruits were delivered for storage.

30 days after the nutrition with Lumbrical bio-fertilizer the biometrical indices of the plants were measured. The content of the plastid pigments and the leaf gas exchange were read.

The photosynthetic pigments were determined spectrophotometrically. The pigments were extracted with 85% water solution of acetone and centrifuged at 4500g for 10 min. The absorbance of the supernatant was measured using a spectrophotometer "Specol 11" (Carl Zeiss Jena, Germany) and calculated according to Lichtenthaler and Wellburn [14].

The leaf gas exchange (P_N – net photosynthetic rate, E – transpiration rate, and g_s - stomatal conductance) was determined using a portable infrared gas analyzer LCA-4 (ADC, England).

In the stage of complete ripeness the fruits were dried in a ventilator dryer at 50 °C. The grinded red pepper was prepared from the pericarp of the fruits. The total pigment content was determined under the ASTA – 19 method modified by Manuelyan [16], immediately after pepper grinding.

The results were statistically processed. The authenticity of the differences was determined under the criterion t of Student.

RESULTS AND DISCUSSION

The data show (Table 2) that nutrition with Lubrical influences plant growth. Both levels of the bio-fertilizer accelerate the growth rate of the roots and stems. The root length in plants with added bio-fertilizer is bigger by 6% on average in Buketen 50 cultivar and by 17% in Gorogled 6. The fresh root mass in both cultivars is considerably higher compared to the control variant. The better developed root system is a precondition for better nutrition of the plants. The same tendency is observed with respect to the growth rate of the stem. These data correspond with the results obtained by other authors [18].

One of the main factors determining the quantity of absorbed sun energy and CO_2 from the plants is the size of the photosynthesizing leaves. Data show that nutrition with bio-fertilizer Lumbrical influences the leaf area. The

Table 2. Influence of bio-fertilizer Lumbrical on growth of young pepper plants, cv. *Buketen 50* and cv. *Gorogled 6*; 1. Control; 2. Nutrition with bio-fertilizer Lumbrical - 50 ml per plant; 3. Nutrition with bio-fertilizer Lumbrical - 100 ml per plant.

Таблица 2. Влияние на биотор Лумбрикал върху растежа на млади растения от пипер, *с. Букетен 50* и *с. Горогled 6*; 1. Контрола; 2. Подхранване с биологичен тор Лумбрикал 50 ml/растение; 3. Подхранване с биологичен тор Лумбрикал 100 ml/растение;

Var.	Root length [cm]	Stem length [cm]	Number of fully devel. leaves	Root fresh weight [g]	Stem fresh weight [g]	Leaf fresh weight [g]	Leaf area [cm^2]
			cv.Buketen 50				
1	25,4±1,2	20,0±0,9	7±1	2,7±0,1	2,0±0,1	7,0±0,1	189,2±10,0
2	26,8±0,9	27,3±0,9 *	9±1 *	5,3±0,1 **	3,8±0,2 **	11,8±0,1 **	362,1±12,0 **
3	27,0±0,8	27,9±1,0 *	10±1 *	4,5±0,1 **	4,4±0,2 **	12,5±0,2 **	357,0±18,2 **
			cv. Gorogled 6				
1	25,4±1,1	23,4±1,0	8±1	2,3±0,1	3,0±0,1	9,2±0,1	164,0±11,0
2	31,4±0,9 *	27,8±0,8 *	12±2 *	5,0±0,2 **	3,9±0,1 *	12,9±0,1 *	243,2±16,2 *
3	27,8±0,9 *	30,5±1,1 *	15±1 **	5,6±0,1 **	4,3±0,1 **	15,7±0,2 **	329,1±10,2 **

Measurements were made 30 days after the application of the bio-fertilizer. Each data point represents the average value of five plant samples ± standard error. Values with an asterisk are statistically significant towards the control at 5% level, and values with two asterisks are statistically significant towards the control at 1% level.

influence of the bio-fertilizer has been mathematically proven for both cultivars and for both nutrition levels.

Bio-fertilizer Lumbrical has a positive effect upon the content of photosynthetic pigments (Table 3). 30 days after the nutrition the content of chlorophyll a increases by 13% on average in Buketen 50 cultivar and by 29% in Gorogled 6, compared to the control variant. With respect to chlorophyll b, the same tendency is observed. The differences in chlorophyll content between the plants fed with bio-fertilizer and the control plants may result from the increased biosynthesis of the latter on the one part, and from the stabilizing effect of the bio-fertilizer upon the chlorophyll and protein complex.

Nutrition with Lumbrical also increases the content of carotenoids and its effect is more clearly defined in Gorogled 6 cv. No deviations from the standard in the ratios between the photosynthetic pigments are observed which shows that the separate pigments are equally

Table 3. Photosynthetic pigment content [mg g^{-1} fresh weight] in the fifth fully developed leaf of pepper plants cv. *Buketen 50* and cv. *Gorogled 6* after the application of the bio-fertilizer Lumbrical; 1. Control; 2. Nutrition with bio-fertilizer Lumbrical - 50 ml per plant; 3. Nutrition with bio-fertilizer Lumbrical - 100 ml per plant;
Таблица 3. Съдържание на фотосинтетични пигменти [mg g^{-1} свежа маса] в пети напълно развит лист на пиперови растения *с. Букетен 50* и *с. Гороглед 6* след подхранване с биотор Лумбрикал; 1. Контрола; 2. Подхранване с биологичен тор Лумбрикал 50 ml/растение; 3. Подхранване с биологичен тор Лумбрикал 100 ml/растение;

Variant	Chlorophyll "a"	Chlorophyll "b"	Carotenoids	Chlorophyll (*a*+*b*) / Carotenoids
		cv. Buketen 50		
1	1,17±0.10	0,54±0.06	0,53±0.01	3,23±0.09
2	1,33±0.07*	0,65±0.05*	0,57±0.04	3,47±0.12
3	1,32±0.05*	0,63±0.06*	0,64±0.09*	3,04±0.06
		cv. Gorogled 6		
1	1,26±0.09	0,56±0,04	0,52±0,05	3,50±0.10
2	1,40±0.09	0,87±0,09*	0,73±0,07*	3,11±0.09
3	1,85±0.04*	0,92±0,08*	0,84±0,09*	3,30±0.09

Measurements were made 30 days after bio-fertilizer applied. Each data point represents the mean value of five plant samples ± standard error. Values with an asterisk are statistically significant towards the control at 5% level, and values with two asterisks are statistically significant towards the control at 1% level.

Table 4. Leaf gas exchange in the fifth fully developed leaf of pepper plants - cv. *Buketen 50* and cv. *Gorogled 6* after the application of bio-fertilizer Lumbrical; 1. Control; 2. Nutrition with bio-fertilizer Lumbrical - 50 ml per plant; 3. Nutrition with bio-fertilizer Lumbrical - 100 ml per plant; P_N – Net photosynthetic rate [µmol $m^{-2}s^{-1}$]; E – Transpiration rate [mmol $m^{-2}s^{-1}$]; g_s – Stomatal conductance [mol $m^{-2}s^{-1}$];
Таблица 4. Листен газообмен в пети напълно развит лист на пиперови растения *с. Букетен 50* и *с. Гороглед 6* след подхранване с биотор Лумбрикал; 1. Контрола; 2. Подхранване с биологичен тор Лумбрикал 50 ml/растение; 3. Подхранване с биологичен тор Лумбрикал 100 ml/растение; P_N – Скорост на нето фотосинтезата [µmol $m^{-2}s^{-1}$]; E – Интензивност на транспирация [mmol $m^{-2}s^{-1}$]; g_s – Устична проводимост [mol $m^{-2}s^{-1}$];

Variant	P_N	E	g_s
		cv. Buketen 50	
1	15,70±0.11	2,59±0.09	0,13±0.01
2	20,88±0.80*	2,67±0.06	0,17±0.02*
3	20,76±0.64*	2,69±0.02	0,17±0.01*
		cv. Gorogled 6	
1	22,73±0.39	1,12±0.07	0,05±0.01
2	23,20±0.52	1,58±0.05*	0,08±0.01*
3	26,23±0.15*	1,53±0.09*	0,08±0.01*

Each data point represents the mean value of five plant samples ± standard error.Measurements were made 30 days after bio-fertilizer applied. Values with an asterisk are statistically significant towards the control at 5% level, and values with two asterisks are statistically significant towards the control at 1% level.

influenced by the nutrition with the bio-fertilizer.

According to many authors [11, 24] the improved leaf gas exchange is a precondition for higher productivity of the plants.

Our research shows (Table 4) that bio-fertilizer Lumbrical improves the leaf gas exchange. In Buketen 50 cultivar the speed of the net photosynthesis is 33% higher compared to the control plants. The intensity of the transpiration does not differ considerably from that of the control variant. At the same time the stomatal conductance in the plants to which the bio-fertilizer was applied, is higher. In Gorogled 6 cultivar the accelerated photosynthesis (by 2-15%) is accompanied by increased intensity of the transpiration and higher stomatal conductance.

The data for the increased speed of the photosynthesis are unidirectional with those related to the content of photosynthetic pigments. This shows that along with the stomatal conductance the increased pigment content is one of the reasons for the higher photosynthetic speed in the plants to which the bio-fertilizer is applied.

Contradictory data about the relationship between photosynthetic intensity and chlorophyll content of leaves have been reported. Along with the reports for the lack of linear relation between two indices there are other reports that higher total chlorophyll content is one of the main factors stimulating the rate of photosynthesis and biological productivity of organisms [17, 19]. Our results support this hypothesis.

One of the yield components which determine its quantity to a large extent is the fruit mass. The results show (Table

Table 5. Pepper yield (raw material for drying) after nutrition with bio-fertilizer Lumbrical; 1. Control; 2. Nutrition with bio-fertilizer Lumbrical - 50 ml per plant; 3. Nutrition with bio-fertilizer Lumbrical - 100 ml per plant;

Таблица 5. Добив от пипер (суровина за изсушаване) след подхранването с биотор Лумбрикал; 1. Контрола; 2. Подхранване с биологичен тор Лумбрикал 50 ml/растение; 3. Подхранване с биологичен тор Лумбрикал 100 ml/растение;

Variant	Number of fruits per plant	One fruit weight [g]	Total yield [kg da^{-1}]
	cv.Buketen 50		
1	9 ± 1	16,11 ± 0,45	811,2 ± 6,0
2	13 ± 1*	16,98 ± 0,39	914,0 ± 7,2
3	12 ± 1*	17,77 ± 0,78*	958,2 ± 8,0*
	cv. Gorogled 6		
1	9 ± 1	15,27 ± 0,47	847,0 ± 10,0
2	13 ± 1*	15,66 ± 0,56	910,2 ± 12,2
3	13 ± 1*	17,31 ± 0,72*	1033,6 ± 26,0**

Each data point represents the mean value of plots per variant ± standard error. Values with an asterisk are statistically significant towards the control at 5% level, and values with two asterisks are statistically significant towards the control at 1% level.

Table 6. Total pigment content /ASTA units/ in red pepper fruits - cv. *Buketen 50* and cv. *Gorogled 6*; yellow (I) and red pigments (II) ratio; 1. Control; 2. Nutrition with bio-fertilizer Lumbrical - 50 ml per plant. 3. Nutrition with bio-fertilizer Lumbrical - 100 ml per plant.

Таблица 6. Общо съдържание на багрила /в ASTA единици/ в червените плодове - с. *Букетен 50* и с. *Гороглед 6*; съотношение жълти (I) и червени (II) пигменти; 1. Контрола; 2. Подхранване с биологичен тор Лумбрикал 50 ml/растение; 3. Подхранване с биологичен тор Лумбрикал 100 ml/растение;

Variant	ASTA	Ratio I/II
	cv.Buketen 50	
1	144	1.021
2	152	1.021
3	158	1.021
	cv. Gorogled 6	
1	159	1.044
2	136	1.027
3	180	1.004

5) that nutrition with bio-fertilizer Lumbrical has a positive effect on this index. The fruit mass increases by 5-10% in Buketen 50 cultivar and by 3-13% in Gorogled 6 cultivar.

Bio-fertilizer Lumbrical influences the yield formation. The yield increases by 18% in Buketen 50 cultivar and by 22% in Gorogled 6 cultivar at higher level of nutrition with bio-fertilizer. The increased yield results from the increased mass as well as from the increased number of the fruits in plants to which the bio-fertilizer was applied.

The most important specification in the red pepper for grinding is the content of coloring substance.

The results show that nutrition with bio-fertilizer Lumbrical has a positive effect upon this index (Table 6). The values of the ASTA units in the grinded red pepper are higher by 8% on average in Buketen 50 cv, compared to the control variant. In Gorogled 6 cv., an increased pigment content is detected at higher level of nutrition with bio-fertilizer.

CONCLUSION

On the basis of the obtained results it was found out that nutrition with bio-fertilizer Lumbrical increases plant growth. It influences the growth rate of the roots and stem of the plant, as well as the leaves formation. There is a positive effect of the bio-fertilizer upon the functional activity of the photosynthetic apparatus /increased content of the photosynthetic pigments, improved leaf gas exchange/.

Lumbrical also influences the yield. The latter increases by 18% in Buketen 50 cv and by 22% in Gorogled 6 cv at higher level of nutrition with bio-fertilizer. Nutrition with Lumbrical has a positive effect upon the pigment content in the red pepper for grinding.

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