Impact of Microbiological Fertilizer Baikal EM-1 on Onion Growth in Greenhouse Conditions

Lelde Grantina-Ievina, Lasma Stanke, Guna Ergle

Latvian Plant Protection Research Centre, Struktoru 14A, Riga, LV-1039, Latvia; E-mail: lelde.grantina-ievina@laapc.lv

Abstract. The microbiological fertilizer Baikal EM-1 contains lactic acid bacteria, photosynthetic bacteria, nitrogen fixing bacteria, *Saccharomyces* yeasts and microbial cultivation media. The aim of the present study was to evaluate the impact of it on onion grown in controlled greenhouse conditions.

In summer 2014 two trials with this product were carried out on onions in greenhouse conditions. Before planting onion bulbs were soaked in water with added fertilizer and growth substrate was watered with the fertilizer according to the instructions of manufacturer. During the vegetation period plants were watered with the fertilizer several times. Identical treatment was performed with water in the control plots. Different treatment schemes and growth substrates (neutralized peat and substrate for vegetables) were used in both trials. The main difference of the used substrates was in the content of mineral nutrients that was higher in the substrate for vegetables. In the second trial additional fertilization with ammonium nitrate was applied three times during the vegetation period. At the end of the trials the yield of onion leaves was estimated, as well as the length of leaves and their chlorophyll content was measured.

At the first trial the obtained yield of onion leaves was increased by 6.4 % and in the second by 8.2 % due to treatment with Baikal EM-1 but these differences were not statistically significant in comparison to the control plots. In general, the fertilizer increased all other measured parameters as well, including average length of leaves by 4.6 % and 1.3 %, the chlorophyll content by 5.0 % and 1.5 %, in the first and second trial respectively, and percentage of onions developing leaves increased by 13.2 % in the second trial. Only the last parameter showed statistically significant differences in comparison to the control. Additionally the growth of the onion was more even in the control treatment. In conclusion, the microbiological fertilizer Baikal EM-1 gave positive impact on onions in greenhouse conditions.

Keywords: chlorophyll content, microbiological fertilizer, onions, yield.

I INTRODUCTION

Microorganisms and their populations in soil and rhizosphere are important players for plant nutrition and health. Beneficial soil microorganisms are responsible for soil born disease suppressiveness [1]. Various soils contain diverse microbial populations. The most important for nitrogen nutrition are nitrogen fixing bacteria, nitrifiers and denitrifiers [2].

Several studies have shown positive impact of the microbiological fertilizer Baikal EM-10n plants. This fertilizer contains among nitrogen fixing bacteria also lactic acid bacteria, photosynthetic bacteria, *Saccharomyces* yeasts and microbial cultivation media. Lactic acid bacteria have been isolated from rhizosphere of fruit trees [3]. Application of photosynthetic bacteria has shown potential for biocomposting [5]. Baikal EM-1, containing broad spectrum of microorganisms, has reduced the inhibition of photosystem II activity of tree seedlings

in the conditions of salt stress [6]. In field conditions Baikal EM-1 (1 and 2 %) has increased yield of maize by 1.85 and 2.65 t h⁻¹, respectively, chlorophyll content in leaves of maize [7] and yield of sugarbeet [8]. A bio-fertilizer, containing *Lactobacillus casei*, *Lactobacillus lactis, Rhodopseudomonas palustris, Saccharomyces cerevisiae*, has improved yield of tomato in greenhouse conditions by 13 and 19 % if used in soil or foliar application [9]. Since scientific information about the microbiological fertilizer Baikal EM-1 is limited, the aim of the present study was to evaluate the impact of it on onion grown in controlled greenhouse conditions.

II MATERIALS AND METHODS

In summer 2014 two trials with Baikal EM-1 (EM Technology, Russia) were conducted on onion (*Allium cepa* L.), cultivar 'Stuttgarter Riesen' (distributed by Latvijas Skirnes Seklas Ltd, Latvia) in greenhouse conditions in Kekava region (Latvia).

ISSN 1691-5402 © Rezekne Higher Education Institution (Rēzeknes Augstskola), Rezekne 2015 DOI: http://dx.doi.org/10.17770/etr2015vol2.273

Before planting onion bulbs were soaked in water with the added fertilizer (0.1 %) and growth substrate was treated with the fertilizer (1 %) according to the instructions of the manufacturer. During the vegetation period plants were watered with the fertilizer several times. Identical treatment was performed with water in the control plots. Different treatment schemes and growth substrates (neutralized peat and substrate for vegetables) were used in the trials (Table 1). The main difference of the used substrates was in the content of phosphorus that was two times higher in the substrate for vegetables. In the second trial additional fertilization was performed three times during the vegetation period with ammonium nitrate (Agrochema Latvia Ltd, content: total nitrogen 34.4 %; ammonium N-NH₄ 17.2 %; nitrates N-NO₃ 17.2 %) and malt extract was added to the fertilizer solution as suggested by the Baikal EM-1 manufacturer in order to activate the microorganisms present in the preparation. The trials were carried out in greenhouse conditions, and onions were planted in eight plastic boxes per treatment. Boxes were regularly watered manually.

In the first trial all onions were able to develop leaves but in the second trial the sprouting capacity was reduced and therefore the number of onions developing leaves was counted at the end of the trial.

At every treatment and assessment, the average air temperature and air humidity in the greenhouse was measured.



Fig. 1. Sorting of onion in groups depending from the length of the leaves in the second trial.

At the end of the trials, at four- to five-leaf stage, the yield of onion leaves per box was estimated (fresh weight), as well as the average length of leaves and their chlorophyll content was measured (five measurements per box) using a hand-held chlorophyll meter SPAD-502 (Konica-Minolta, Japan). At the end of the second trial all onions from each box were sorted in groups depending from the length of the leaves (Fig. 1) and respective number of onion and weight of each group (0-10 cm, 11-20 cm, 21-30 cm, 31-40 cm, 41-50 cm, 51-60 cm, 61-70 cm) was estimated.

Significance of differences between means was determined by the t-test at the $\alpha = 0.05$ level with *Excel* (Microsoft, USA). Significance was evaluated at p < 0.05 level.

	TREATMENT SCHEMES			
Process	First trial	Second trial		
Size of plastic boxes, m ²	0.24	0.10		
Substrate treatment two weeks before planting with 1 % Baikal EM-1 solution	yes	no		
Soaking of onion bulbs before planting in 0.1 % Baikal EM-1 solution	20 min	12 h		
Planting, 1500/m ²	07.05.2014.	25.07.2014.		
Spraying with 0.001 % Baikal EM-1 solution	one time	none		
Watering with 0.001 % Baikal EM-1 solution	two times	three times		
Addition of malt extract (0.001 %) to Baikal EM-1 solution	no	in all treatments		
Fertilizing with ammonium nitrate, 10 g m ⁻²	no	three times with 7-8 day interval		
Harvest	02.06.2014.	28.08.2014.		
Substrate and its content	Neutralized peat "Kasvuturvas", Estonian Peat Products Ltd., Estonia Added nutrients, g mg Γ^1 : Nitrogen (NH ₄ and NO ₃) – 120.00 Phosphorus (P ₂ O ₃) 140.00 Potassium – (K ₂ O) 240.00	Substrate for vegetables "Terra Vita", SAS "MNPP PHART", Russia. Content, mg I^{-1} : Nitrogen (NH ₄ and NO ₃) 150.00 Phosphorus (P ₂ O ₅) 270.00 Potassium (K ₂ O) 300.00.		

TABLE I TREATMENT SCHEMES

III RESULTS AND DISCUSSION

In the first trial the average length of onion leaves at the harvest in the control was 34.94 ± 3.25 cm. The average length of onion leaves in the treatment was

 36.55 ± 2.09 cm. The difference was 4.61 % but it was statistically not significant ($t_{Stat} = 1.17 < t_{Critical two-tail} = 2.15$). At the first trial the obtained yield of onion leaves per box of the control was 585 ± 67.40 g that corresponds to 24.4 t ha⁻¹. The obtained onion yield in

the treatment was 622.5 ± 83.79 g per box (0.24 m^2) that corresponds to 25.9 t ha⁻¹. The difference was 6.41 % but it was statistically not significant ($t_{Stat} = 0.99 < t_{Critical two-tail} = 2.15$). The average chlorophyll content of onion leaves in the control was 17.08 \pm 1.61 SPAD units. The average chlorophyll content of onion leaves in the treatment was 17.92 \pm 1.82 SPAD units that was increased by 4.92 % in comparison to the control. This difference was statistically not significant ($t_{Stat} = 0.97 t_{Critical two-tail} = 2.15$).

In the first trial all onions were able to develop leaves but in the second trial only 70.83 ± 12.36 % of bulbs developed leaves in the control but 84.00 ± 2.89 % bulbs developed leaves in the treatment. The difference was 13.17 % and it was statistically significant (p = 0.02; $t_{Stat} = 2.93 > t_{Critical two-tail} = 2.31$).

In the first trial at the harvest the average length of the onion leaves in the control was 40.88 ± 3.70 cm. The average length of the onion leaves in the treatment was 41.41 ± 3.91 cm. The difference was 1.31 % but it was statistically not significant ($t_{Stat} = 0.28 < t_{Critical two-tail} = 2.15$).

At the second trial the obtained yield of onion leaves per box of the control was 438.13 ± 44.60 g that corresponds to 43.8 t ha⁻¹. The obtained onion yield in the treatment was 474.00 ± 33.81 g per box (0.1 m²) that corresponds to 47.4 t ha⁻¹. The difference was 8.19 % but it was statistically not significant ($t_{Stat} = 1.81 < t_{Critical two-tail} = 2.15$).

The average chlorophyll content of onion leaves in the control was 25.80 ± 0.91 SPAD units. The average chlorophyll content of onion leaves in the treatment was 26.18 ± 1.18 SPAD units. The difference was 1.47 % but it was statistically not significant ($t_{Stat} = 0.32 < t_{Critical two-tail} = 2.15$).

The average chlorophyll content was statistically significantly higher in the second trial in the control $(p < 0.001; t_{Stat} = 8.65 > t_{Critical two-tail} = 2.15)$ as well as in the treatment (p < 0.001; $t_{Stat} = 7.77 > t_{Critical two-tail} =$ 2.15) in comparison to the first trial. Differences between trials can be explained by the higher level of nitrogen in the substrate used for the second trial and additional fertilization with ammonium nitrate. However the detected level of the chlorophyll has to be considered low in comparison to other investigations were onion has been grown on organic soils and the chlorophyll content was estimated with the same method. For example, in the study of Westerveld et al. [9] the chlorophyll content of yellow cooking onion cultivar 'Hamlet' leaves at five-leaf stage varied from 46.9 to 56.2 SPAD units on organic soil in field conditions depending from the amount of nitrogen rate in the fertilization $(0 - 200 \text{ kg ha}^{-1})$ and year of the trial. Such differences can be explained by different growth conditions (greenhouse vs. field) and different cultivars.

The average yield of fresh onion leaves was statistically significantly higher in the second trial in the control (p < 0.001; $t_{Stat} = 2.76 > t_{Critical two-tail} =$

2.15) as well as in the treatment (p < 0.001; $t_{Stat} = 2.79 > t_{Critical two-tail} = 2.15$) in comparison to the first trial. This can be explained by a higher amount of mineral nutrients in the substrate used in the second trial. Increased fresh weight of onion leaves followed by higher potassium and phosphorus concentrations, for example, has been reported in other investigations as well [11,12].

Sorting of onion in groups depending from the length of the leaves has shown that in the treatment higher biomass was obtained in the length groups up to 50 cm, especially in the length group 31-40 cm (Table 2) where this difference was statistically significant in comparison to the control (p = 0.01, $t_{Stat} = 3.00 > t_{Critical two-tail} = 2.15$). In the length groups 51-60 cm and 61-70 cm the highest biomass was observed in the control, especially in the length group 51-60 cm, where the difference in comparison to treatment was statistically significant (p = 0.01, $t_{Stat} = 2.78 > t_{Critical two-tail} = 2.15$).

Number of onion in the control boxes was higher in the smallest (0-20 cm) and largest length groups (51-70 cm) but in the treatment number of onion was higher in the length categories from 21 to 50 cm (Table 2) that is more suitable for the production quality.

Plant growth-promoting soil microorganisms improve plant nutrition and growth characteristics through various processes including nitrogen fixation, breakdown of organic substances, solubilization of minerals, release of chelating compounds and biologically active molecules such as phytohormones, vitamins and enzymes, and, as a result, improve the nutrient uptake by plant root system [13]. In the present study the tested fertilizer significantly increased the sprouting capacity of onion, slightly increased the length of the onion leaves, the fresh weight, the chlorophyll content, and the growth evenness. The growth period of the onion was remarkably shorter than in other studies where crops with longer vegetation period were used, such as tree seedlings, maize, sugarbeet and tomatoes [6,7,8,9]. Probably the time period in the present study was too short for the beneficial microorganisms of the fertilizer Baikal EM-1 to multiply up to effective level and to give the maximal potential.

IV CONCLUSIONS

Although the level of mineral nutrients caused significant impact on the yield of onion leaves (fresh weight), as well as the average length of leaves and their chlorophyll content, the added fertilizer has shown positive impact of these parameters if comparing the control plots with treatment plots. Additionally Baikal EM-1 showed plant growth stimulating activity in the second trial when the sprouting capacity of onions was reduced. The percentage of onions developing leaves was increased by 13.2 % in comparison to the control plots, and the suitable for the production quality aspects. length of the onion leaves was more even that is

ONION FRESH MASS AND NUMBER OF PLANTS IN EVERY LENGTH GROUP								
Measured parameter	0-10 cm	11-20 cm	21-30 cm	31-40 cm	41-50 cm	51-60 cm	61-70 cm	
Fresh mass (g) (± S.D.) in the control	0.75±0.46	6.13±2.47	20.75±5.18	91.75±33.38	196.75±30.87	109.63±29.26	12.38±15.95	
Fresh mass (g) (± S.D.) in the treatment	0.88±0.35	6.38±3.46	26.75±9.97	141.50±32.91	225.75±56.70	67.38±31.37	5.38±11.70	
Number of plants (± S.D.) in the control	2.38±2.07	7.00±2.45	13.63±3.02	28.63±10.50	37.75±9.38	14.13±3.40	1.25±1.58	
Number of plants (± S.D.) in the treatment	2.50±1.85	6.50±2.33	16.00±5.58	42.25±8.97	44.25±10.94	9.88±3.83	0.75±1.75	

TABLE 2 ONION FRESH MASS AND NUMBER OF PLANTS IN EVERY LENGTH GROUI

V ACKNOWLEDGEMENTS

We are very thankful to EM Technologies for providing the fertilizer used in the study.

VI REFERENCES

- R. L. Berendsen, C. M. J. Pieterse, P. A. H. M. Bakker, "The rhizosphere microbiome and plant health," Trends Plant Sci., vol. 17, pp. 478-486, 2012.
- [2] T.-H. Anderson, "Microbial eco-physiological indicators to asses soil quality," Agric. Ecosyst. Environ., vol. 98, pp. 285-293, 2003.
- [3] Y.-S. Chen, F. Yanagida, T. Shinohara, "Isolation and identification of lactic acid bacteria from soil using an enrichment procedure," Lett. Appl. Microbiol., vol. 40, pp. 195-200, 2005.
- [4] M. Kobayashi, M. Z. Haque, "Contribution to nitrogen fixation and soil fertility by photosynthetic bacteria," Plant Soil, vol. 35, pp. 443-456, 1971.
- [5] M. M. Roper, J. K. Ladha, "Biological N₂ fixation by heterotrophic and phototrophic bacteria in association with straw," Dev. Plant Soil Sci., vol. 65, pp. 211-224, 1995.
- [6] S. Allahverdiyev, A. Atilla, B. S. Ismail, A. Sahmurova, "Response of photosystem II and photosynthetic pigments to

salt and Baikal EM1 in tree seedlings," Afr. J. Biotechnol., vol. 10, pp. 535-538, 2011.

- [7] Z. P. Okazova, M. V. Efakov, "Use of biological products in corn seed farming," Plant Protect., vol. 40, pp. 261-263, 2014.
- [8] A. K. Kulikova, O. S. Dronina, E. A. Nikiforov, "Effect of seed treatment by diatomite powder on soil nutrient status and sugarbeet productivity," Sakharnaya Svekla, vol. 1, pp. 24-26, 2009.
- [9] S. M. Westerveld, A. W. McKeown, M. R. McDonald, "Assessment of chlorophyll and nitrate meters as field tissue nitrogen tests for cabbage, onions, and carrots," HortTechnology, vol 14, pp. 179-188, Apr.-June. 2004.
 [10] I. Tringovska, "The effects of humic and bio-fertilizers on
- [10] I. Tringovska, "The effects of humic and bio-fertilizers on growth and yield of greenhouse tomatoes," Acta Hort., vol. 960, pp. 443-449, 2012.
- [11] A. M. El-Bassiony, "Effect of potassium fertilization on growth, yield and quality of onion plants," J. Appl. Sci. Res., vol. 2, pp. 780-785, 2006.
- [12] A. M. Shaheen, M. M. Abdel-Mouty, A. H. Ali, F. A. Rizk, "Natural and chemical phosphorus fertilizers as affected onion plant growth, bulbs yield and its some physical and chemical properties," Austr. J. Basic Appl. Sci., vol. 1, pp. 519-524, 2007.
- [13] C. Altomare, I. Tringovska, "Beneficial soil microorganisms, an ecological Aalternative for soil fertility management," Sust. Agric. Rev., vol. 7, pp 161-214, May 2011.