

## THE INFLUENCE OF BIO-ALGEEEN S90 ON THE GROWTH OF MULTIFLORA ROSE SEEDLINGS (*Rosa multiflora* Thunb.)

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### ABSTRACT

Striving to intensify horticultural production, new and more effective bio-preparations are being sought to stimulate plant growth and development. Bio-algeen S90 is a natural agent based on sea algae, the high biological activity of which results from the high content of natural growth regulators. The aim of the study was to verify the influence of Bio-algeen S90 on the growth, morphological characteristics and chlorophyll fluorescence of *Rosa multiflora* seedlings. The bio-preparation was applied one, two and three times at concentrations: 0.1, 0.2, 0.4 and 0.6 mg dm<sup>-3</sup>. Following parameters were measured to evaluate the response of plants to the bio-preparation:  $F_0$  – initial fluorescence,  $F_m$  – maximal fluorescence in the dark-adapted state,  $F_v/F_m$  – maximum photochemical efficiency of PSII. All concentrations of the bio-preparation and frequency of its application stimulated the number of shoots in a bush, the length of shoots and the diameter of the root crown of plants intended for budding. The most beneficial was the two-fold bio-preparation application at a concentration of 0.4 mg dm<sup>-3</sup>. Bio-algeen also positively influenced the chlorophyll fluorescence parameters. The highest mean  $F_0$  and  $F_m$  values were recorded with the two-fold preparation treatment. There was no significant effect of the bio-preparation on the  $F_v/F_m$  index, which was within the range of 0.75–0.66.

**Key words:** *Rosa*, rootstock, bio-preparation, fluorescence, chlorophyll

### INTRODUCTION

A rootstock significantly affects not only the resistance of plants to low temperatures, but also the size and quality of the flower yield, as well as the date and flowering of cultivated varieties [Włodarczyk and Bryzek 2008, Monder and Hetman 2011, Szmagara et al. 2016]. Selection of the rootstock also determines the viability of cultivated varieties grown on it, the inflorescence color, and the length of inflorescence shoots [Pudelska 2003, Hetman et al. 2007, Włodarczyk and Bryzek 2008, Balej and Zogaj 2011, Szmagara et al. 2016]. *Rosa multiflora* is the most commonly used rootstock in eastern Poland, especially in the Lublin region. As

shown by studies, cultivated varieties budded on *R. multiflora* are characterized by stronger growth of shrubs, higher flower yield compared to the inflorescence yield obtained from varieties budded on *R. canina*, both in the field cultivation and under cover [Pudelska 2003, Hetman and Szmagara 2013, Szmagara et al. 2016].

In recent years, there has been an increasing interest in bio-preparations that are safe for people and environment and allow to use the potential of arable crops. They stimulate the growth and development of species and varieties of various groups of arable crops, including potatoes, sugar beets, cereals, vegetables,

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herbs, ornamental plants: Norway spruce, cherry laurel and machaleb cherry [Grzeškiewicz and Trawczyński 1998, Dobromilska et al. 2008, Kiełtyka-Dadasiewicz and Król 2012, Truba et al. 2012, Dudaš et al. 2014, Hanzal et al. 2015, Lorenc et al. 2016, Szabó et al. 2016, Tobiasz-Salach et al. 2016].

Bio-algeen S90 is a natural preparation based on sea algae, the high biological activity of which results from high content of natural growth regulators (auxins, cytokinins and gibberellins) responsible for growth control and for plant resistance [Dobromilska et al. 2008, Mikiciuk and Dobromilska 2014]. Phytohormones contained in algae help plants adapt to stress conditions by maintaining constant hydration of cells and stimulation of the root system [Matysiak and Adamczewski 2009]. The preparation also includes microorganisms, and above all, macro- and microelements. Bio-algeen S90 contains 90 groups of organic compounds, including alginic acid, vitamins and amino acids [Sultana et al. 2011, Truba et al. 2012, Mikiciuk and Dobromilska 2014]. Bio-stimulants contained in it influence indirectly or directly on the uptake and transport of nutrients, water as well as photosynthesis [Dobromilska et al. 2008].

The photosynthesis process is closely related to the plant species and even to the variety and is largely modified by environmental conditions, including the air temperature, amount of rainfalls and their distribution during the growing season [Niu and Rodriguez 2009, Olszewska et al. 2010]. Impact of these external factors may, to some extent, be alleviated by the use of certain bio-preparations to plants [Polanco et al. 2014]. Studies performed by Orlikowski et al. [2002], Szmagara [2007] and Polanco et al. [2014] reveal that bio-preparations based on organic compounds have the ability to limit the growth of many bacteria and fungi that cause plant diseases, but also strengthen and stimulate plant resistance mechanisms. Measurement of chlorophyll fluorescence – non-invasive method used to assess environmental factors using cultivated plants – gives the opportunity to assess their physiological status [Kalaji and Łoboda 2010]. It is a non-invasive and commonly used method for assessing the environmental factors on plants [Michalek and Sawicka 2005].

The aim of the study was to verify the effect of bio-preparation Bio-algeen S90, administered in the

form of spraying, on growth, morphological characteristics and chlorophyll fluorescence of multiflora rose seedlings (*R. multiflora* Thunb.).

## MATERIALS AND METHODS

The field experiments were conducted in 2013–2014 at the Experimental Farm Felin belonging to the University of Life Sciences in Lublin (Poland, 51°23'N, 22°56'E). The experimental material consisted of seedlings of a multiflora rose (*Rosa multiflora* Thunb.), which in the beginning of May, when they produced at least two true leaves, were pricked-out onto plots. They were planted in two rows separated from each other by 80 cm, and the distance of 15 cm was kept in rows. Then the belts were divided into plots with an area of 3.4 m<sup>2</sup>. Twenty-four plants grew on one plot, which was a single replicate. After the plant adaptation, top dressing fertilization with Azofoska at the dose of 200 g fertilizer on the plot, was applied. Systematic plant protection was carried out. From the end of June, sprays using Bio-algeen S90 at concentrations of 0.1, 0.2, 0.4 and 0.6 mg·dm<sup>-3</sup> were begun, which were performed one, two and three times a week. The control combination was made up of plants sprayed with water. The entire experiment included 15 treatments in 5 replications.

In each growing season, chlorophyll fluorescence measurements were carried out in the middle of August using a portable chlorophyll fluorescence meter OS30p (OptiScience Inc., USA). The chlorophyll fluorescence was measured on well-formed leaves, six measurements for each combination. To assess the response of plants to the bio-preparation used, the following parameters were measured:  $F_0$  – initial fluorescence,  $F_m$  – maximal fluorescence in the dark-adapted state,  $F_v/F_m$  – maximum photochemical efficiency of PSII.

At the end of August, young *R. multiflora* plants that were suitable for budding were dug out and their biometric measurements were carried out: height of the aboveground part, number of the first-order shoots and diameter of the root crown of the rootstock.

The results were subjected to statistical analysis using Tukey's analysis of variance and confidence intervals at the significance level  $\alpha = 0.05$ .

## RESULTS AND DISCUSSION

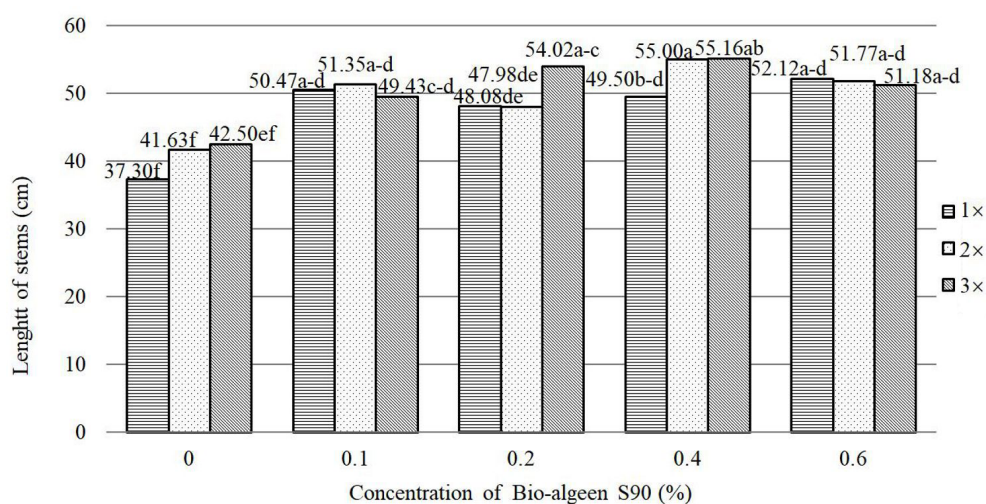
From the observations made over two years of research it appears that *R. multiflora* seedlings treated with Bio-algeen S90 tended to form longer shoots compared to the control plants. The mean longest shoots were developed by plants in the second year of the study after using Bio-algeen solution at the concentrations of 0.4 and 0.6 mg dm<sup>-3</sup>, i.e. 77.00 cm and 73.21 cm (Tab. 1). When analyzing the effect of concentration and frequency of the applied bio-preparation, the longest shoots were produced

by plants in the second year of the research with the application of Bio-algeen S90 solution twice and at the concentration of 0.4 mg dm<sup>-3</sup> (83.30 cm) and with triple spray (78.03 cm) (Tab. 1). The increase in the length of shoots in these treatments in 2014 relative to the control plants amounted to 24% and 17%, and in 2013 – 28% and 37%, respectively. Regardless the year of research, the longest shoots were formed by plants at two and three-fold application of bio-preparation and concentration of 0.4 mg dm<sup>-3</sup>, and the values were statistically different from control treatment (Fig. 1).

**Table 1.** Influence of Bio-algeen S90 on the length (cm) of *R. multiflora* shoots

Year	Number of sprays	Concentration of bio-preparation (mg dm <sup>-3</sup> )				
		0	0.1	0.2	0.4	0.6
2013	1×	20.07j	30.87hi	28.90hi	29.33hi	28.57hi
	2×	20.07j	29.67hi	30.33hi	28.00hi	30.90hi
	3×	20.1 j	31.20hi	30.63hi	32.28hi	31.03hi
2014	1×	54.30g	70.07b-f	67.27d-f	69.67c-f	75.67a-d
	2×	63.20ef	73.03b-e	65.63e-f	83.30a	72.63b-e
	3×	64.93ef	67.70d-f	77.40a-c	78.03ab	71.33bf
Mean	2013	20.07E	30.6 D	29.96D	29.87D	30.17D
	2014	60.89C	70.27B	70.10B	77.00A	73.21AB

Values marked with the same letter do not differ significantly at  $\alpha = 0.05$



**Fig. 1.** Average effects of Bio-algeen S90 sprays on the length (cm) of *R. multiflora* shoots

In the case of other species, such as common motherwort or ginseng, a positive effect of bio-stimulants on plant height was observed [Kiełtyka-Dadasiewicz and Król 2012]. Gruszczyk and Berbec [2004] reported a 21% increase in the height of feverfew after the use of Bio-algeen S90, while Kiełtyka-Dadasiewicz and Król [2012] recorded the increase in parameters such as plant height, shoot thickness and number of branches, inflorescence length and seed and herb yield in common motherwort. Thus, the foliar application of the bio-preparation significantly influenced

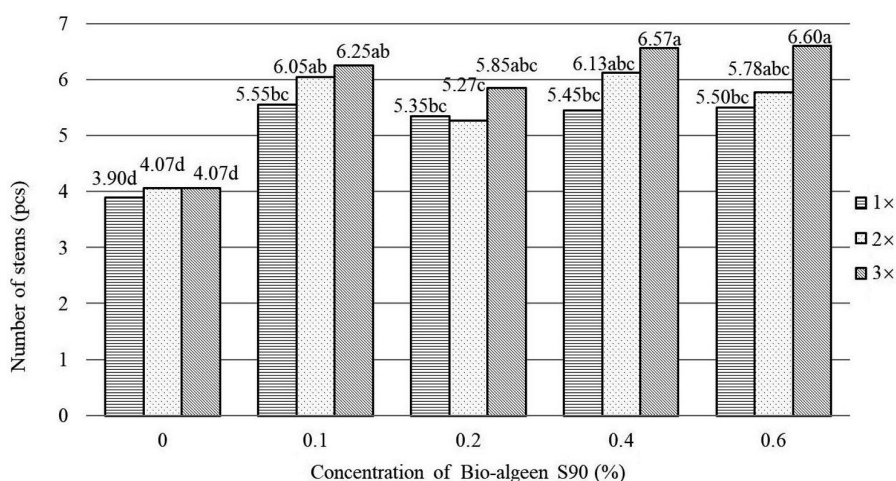
the improvement of qualitative and quantitative traits in herbal plants. The results of tests carried out on potatoes, sugar beets, oats or tomatoes also indicate the beneficial impact of this bio-preparation on the yield of plants [Grzeškiewicz and Trawczyński 1998, Dobromilska et al. 2008, Truba et al. 2012, Tobiasz-Salach et al. 2016].

Among the mean values of parameters in studied treatments measured in present experiment, the largest number of shoots (7.07 pcs.) was produced by shrubs of multiflora rose in 2014 using the solution of Bio-

**Table 2.** Influence of Bio-algeen S90 on the number (pcs) of *R. multiflora* shoots

Year	Number of sprays	Concentration of bio-preparation (mg dm <sup>-3</sup> )				
		0	0.1	0.2	0.4	0.6
2013	1×	2.90i	4.80 g-i	4.73hi	4.73 hi	4.17i
	2×	2.90i	4.60hi	4.50i	4.63hi	5.10f-i
	3×	2.90i	5.17e-i	5.27e-i	5.73d-i	5.77d-i
2014	1×	4.90f-i	6.30a-e	5.97d-h	6.17b-f	6.83a-d
	2×	5.23e-i	7.50ab	6.03c-g	7.63a	6.47a-e
	3×	5.23e-i	7.33abc	6.43 a-e	7.40ab	7.43ab
Mean	2013	2.90D	4.86C	4.83C	5.03C	5.01C
	2014	5.12C	7.04A	6.14B	7.07A	6.91A

Values marked with the same letter do not differ significantly at  $\alpha = 0.05$



**Fig. 2.** Average effects of Bio-algeen S90 sprays on the on the number (pcs) of *R. multiflora* shoots

algeen S90 at the concentration of 0.4 mg·dm<sup>-3</sup>, and their number significantly differed from the number of shoots (5.12 pcs) obtained from controls, but it did not differ significantly from the number of shoots (7.04 and 6.91 pcs) obtained from the treatments, in which the preparation was used at the concentration of 0.1 and 0.6 mg·dm<sup>-3</sup> (Tab. 2). Significantly less shoots were obtained in 2013 within all treatments, and their number did not differ significantly from the number of shoots in the control combination in 2014 (Tab. 2). The most beneficial was the bio-preparation concentration of 0.4 mg·dm<sup>-3</sup> with twice application; plants produced the largest number of shoots (7.63 pcs) and their number differed significantly from the number of shoots obtained from the control treatments throughout the entire study period, to which the bio-preparation was not used (Tab. 2). Therefore, the increase in the number of shoots in this most advantageous combination compared to the control was 31.5%. Analyzing the influence of the concentration of Bio-algeen S90 regardless of the years of research, the largest number of shoots (in the range 6.60–6.05 pcs) was obtained at three applications of the preparation within all concentrations and two applications at concentrations of 0.4 and 0.1 mg·dm<sup>-3</sup>. The least shoots were formed by control shrubs (in the range of 3.90–4.07 pcs), to which no bio-preparation was applied, and their length was significantly different from that of shoots within all treatments with Bio-algeen (Fig. 2).

According to Kiełtyka-Dadasiewicz and Król [2012], the double-spraying of common motherwort with Bio-algeen S90 increased the number of branching by up to 61%. The assessment of the influence of Bio-algeen S90 on the yield increase in vegetable cultivation shows that this product has a positive effect on such features as root thickness and size of pepper leaves as well as the number of fruits and flowering of tomatoes [Dobromilska and Guberewicz 2008, Jamiółkowska 2014]. Also in our research, this bio-stimulant positively affected the diameter of the root crown. The research showed that, on average, *R. multiflora* shrubs treated with Bio-algeen S90 within all concentrations of the bio-preparation, formed larger diameter of the root crown than in control ones. It differed significantly from the diameter within all treatments in 2013, but it did not differ from the diameter of the root crown in 2014 (Tab. 3). The largest diameter of root crown was found in *R. multiflora* shrubs treated twice with the bio-preparation at the concentration of 0.6 mg·dm<sup>-3</sup> and they were almost 20% larger in diameter as compared to that of the root crown of control shrubs. The smallest diameter of root crowns (5.2 mm) was found in control bushes in 2013 and it differed significantly from the crown diameter in all treatments from 2014 (Tab. 3). Similarly, the stimulating effect of the bio-preparation on the diameter of the root crown was observed regardless of the year of study. It was the highest with three applications

**Table 3.** Influence of Bio-algeen S90 on the diameter of root crown (mm) of *R. multiflora*

Year	Number of sprays	Concentration of bio-preparation (mg·dm <sup>-3</sup> )				
		0	0.1	0.2	0.4	0.6
2013	1×	5.22h	7.66fg	6.91fgh	7.03fg	6.50gh
	2×	5.22h	7.14fg	6.65gh	6.27gh	7.40fg
	3×	5.22h	6.98fg	7.66fg	7.78fg	7.90fg
2014	1×	8.55ef	10.14b-e	9.93cde	10.87a-d	11.31abc
	2×	9.61de	11.10a-d	10.72a-d	11.46ab	11.92a
	3×	9.81cde	11.38abc	10.96a-d	11.10a-d	10.87a-d
Mean	2013	5.22C	7.26B	7.07B	7.02B	7.26 B
	2014	9.32A	10.87A	10.53A	11.10A	11.37A

Values marked with the same letter do not differ significantly at  $\alpha = 0.05$



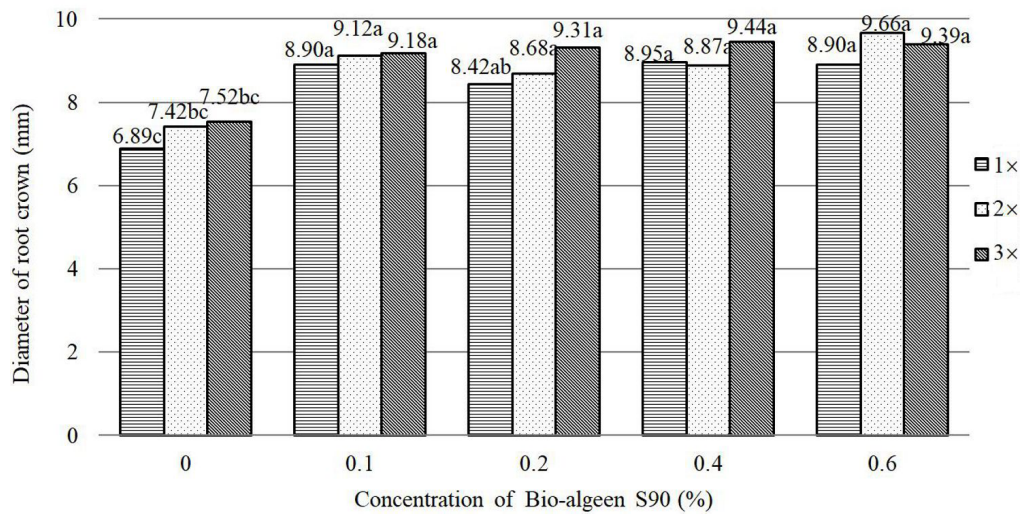


Fig. 3. Average effects of Bio-algeen S90 sprays on the diameter of root crown (mm) of *R. multiflora*

Table 4. Fluorescence parameters ( $F_0$ ,  $F_m$ ,  $F_v/F_m$ ) measured in *R. multiflora* leaves

Fluorescence parameters	Number of sprays	Concentration of bio-preparation ( $\text{mg dm}^{-3}$ )					Mean
		0	0.1	0.2	0.4	0.6	
$F_0$	1x	197.33bc	173.17c	196.33bc	192.50bc	225.17ab	196.9C
	2x	217.00ab	254.50a	206.33bc	220.67ab	226.00ab	224.90A
	3x	220.33ab	220.83ab	207.33bc	189.83bc	220.17ab	211.70B
	Mean	211.56ABC	216.17AB	203.33BC	201.00BC	223.78A	211.17
$F_m$	1x	646.00e	669.17de	685.17cde	786.33abc	780.17abc	713.37B
	2x	703.17a-e	755.50a-d	803.83ab	793.83ab	732.17a-e	757.70A
	3x	674.00de	810.33a	656.67de	733.67a-e	696.83b-e	714.30B
	Mean	674.39C	745.00AB	715.22B	771.28A	736.39AB	728.46
$F_v/F_m$	1x	0.67a	0.73a	0.67a	0.75a	0.71a	0.70A
	2x	0.67a	0.70a	0.75a	0.73a	0.68a	0.70A
	3x	0.71a	0.73a	0.67a	0.74a	0.66a	0.70A
	Mean	0.69A	0.72A	0.69A	0.74A	0.68A	0.70

$F_0$  – initial fluorescence;  $F_m$  – maximal fluorescence in the dark-adapted state;  $F_v/F_m$  – maximum photochemical efficiency of PSII

Values marked with the same letter do not differ significantly at  $\alpha = 0.05$

The values are means of two growing seasons

of the bio-preparation regardless of its concentration and with two applications at concentrations of 0.6 and 0.1 mg·dm<sup>-3</sup> and it was significantly different from the diameter in the control treatments. The diameter of the root crown in all treatments with Bio-algeen was significantly higher than in the control, apart from the crown diameter, when the bio-preparation was used once at the concentration of 0.2 mg·dm<sup>-3</sup> (Fig. 3).

The conducted studies have shown the improvement in the root crown parameters, which is very important in the case of the budding procedure. Its larger diameter is very desirable feature of a good rootstock, that ensures better adaptation of the spot of the noble variety. Similarly, in the study upon common motherwort, Kiełtyka-Dadasiewicz and Król [2012] showed that plants treated with Bio-algeen S90 significantly increased the thickness of shoots during the growing season. According to own research, the three-fold application of Bio-algeen S90, regardless of concentration, caused an increase in the diameter of the root crown. Similarly, Hanzal et al. [2015] and Lorenc et al. [2016] showed significant increase in the diameter of root crown and other parts of *Picea abies* after the use of Bio-algeen S90. The action of Bio-algeen and other bio-alginates leads to a significant improvement in the quality of seedlings. Depending on the tree species, environmental conditions, application techniques and interaction with other factors, the effect of application towards seedlings may vary. However, in most cases, these products affect the growth and development of seedlings positively, therefore they can be used as a means to strengthen trees in a nursery [Lorenc et al. 2016].

Measurement of chlorophyll fluorescence is a non-invasive method for determining the physiological status of plants [Roháček and Barták 1999, Zlatev and Yordanov 2004, Efeoğlu et al. 2009, Kałużewicz et al. 2018, Rios et al. 2018]. The present study conducted on young *R. multiflora* plants was aimed at determining the effect of Bio-algeen S90 on the following parameters of chlorophyll fluorescence:  $F_0$ ,  $F_m$ ,  $F_v/F_m$ . The research has showed that the number of Bio-algeen applications had significant impact on average  $F_0$  values. The highest value of this indicator was recorded with the double use of the bio-preparation at the concentration of 0.1 mg·dm<sup>-3</sup> (Tab. 4). Similarly, the highest mean  $F_m$  values were recorded with bio-preparation at the concentration of

0.2 mg·dm<sup>-3</sup> with two applications. With the help of the  $F_v/F_m$  index, the maximum efficiency of the PS II photosystem is determined, the measurement of which allows determining the activity of the photosynthetic apparatus [Dąbrowska et al. 2011]. It is assumed that its optimal level should be close to 0.83, then it indicates the maximum efficiency of photosynthesis and is treated as a reference point for various deviations resulting from stress acting on the plant [Johnson et al. 1993, Maxwell and Jonson 2000]. The obtained results indicate that the chlorophyll fluorescence index of *R. multiflora* on average had the highest value when plants were treated with the bio-preparation at concentration of 0.4 mg·dm<sup>-3</sup>, although the values did not differ statistically. There was no effect of the bio-preparation application frequency on this indicator. The obtained results indicate that in all treatments, the photosynthetic efficiency index was slightly lower than the optimal one and it was within the limits of 0.75–0.66 (Tab. 4). It can be assumed that under the influence of environmental factors, the PSII efficiency decreased, which resulted in slightly lower  $F_v/F_m$  values. This is confirmed by the research of other authors that the impact of biotic and abiotic factors causes a decrease in the performance of the PSII photosystem [Cetner et al. 2016].

## CONCLUSIONS

Bio-algeen S90 positively influenced the tested parameters of *R. multiflora*. All concentrations of the bio-preparation and frequency of application stimulated the length and number of shoots and the diameter of the root crown. However, the most beneficial was the two-fold bio-preparation application at the concentration of 0.4 mg·dm<sup>-3</sup>. Bio-algeen also positively influenced the chlorophyll fluorescence parameters. The highest mean values of  $F_0$  and  $F_m$  were recorded with double application of the preparation. There was no significant effect of the bio-preparation on the  $F_v/F_m$  index, which was within the limits of 0.75–0.66.

## ACKNOWLEDGMENTS

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