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EFFECT OF BACTERIAL FERTILIZER ON GROWTH, YIELD AND GRAPE QUALITY OF POTTED CABERNET SAUVIGNON (VITIS VINIFERA L.)

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ABSTRACT

In these studies, one commercial bacterial fertilizer - BactoFil B10 was used during the first and second growing seasons of the potted vines. The influence of the fertilization was investigated on the vine growth – total leaf area, shoot diameter, dormant pruning weight and fresh root weight. The most expressed effect of Bactofil application was in the first vegetation with increases of the total leaf area of 9%, shoot length of 6% and shoot pruning weight of 14.6%, in comparison to the control. At the end of the second vegetation, plants were removed from the pots and the fresh root weight was measured. It was found that applied treatment was not influenced on the variations of the average root weight. The first grapes were obtained in the second vegetation, and treatment with BactoFil was not influenced on the differences in the yield, grape and berry weight. Also, treatment was not influenced on the must quality which was expressed over the dry matter content and total acid content.

INTRODUCTION

The established practice in conventional viticulture production in Serbia involves the use of mineral fertilizers whose long-term utilization, together with the use of pesticides and the impropriate mechanization, leads to disturbance of soil properties, the accumulation of toxic compounds in soil and plant tissues. By introducing the practice of bio fertilization, especially with the use of bacteria, the biological properties of the soil are improved by significantly increasing the types and number of useful rhizosphere bacteria that positively affect the structure and fertility of the soil, the accessibility of nutrients, especially N, P and K, and the synthesis of other stimulating compounds. The direct influence of rhizospheric bacteria on the vine is reflected as the increase in the content of readily available macro and micro nutrients in the rhizosphere, the synthesis of hormones and other stimulating compounds. The increased availability of accessible N in the rhizosphere is due to the activity of symbiotic and non-symbiotic azotofixators (*Rhizobium*, *Azospirillum*, *Azotobacter*, etc.). The translation of bound and insoluble P and K into forms directly accessible to the plant takes place thanks to the bacteria of the species *Bacilus*, *Pseudomonas*, *Aspergilus* and others. Further, the synthesis of siderophores, molecules containing easily accessible Fe, is significant in preventing of the chlorosis on carbonate soils. In translating the insoluble Fe^{+3} into easily accessible molecules - siderophores, the bacteria of genera *Aeromonas*, *Azotobacter*, *Bacillus*, *Pseudomonas*, *Rhizobium* and others play a major role. The special significance of rhizospheric bacteria is in the ability to synthesize phytohormones such as auxins, cytokinins, giberellins and ethylenes that have a positive effect on the growth of the organs, branching and extending of the root, shoots, and growth of berries. The ability of the phytotohormone synthesis is distinguished by 80% of the rhizosphere bacteria (Vessey, 2003). In addition to direct, no less significance has the indirect effect of rhizosphere bacteria on the vine. This influence is based on the synthesis of various compounds that promote the resistance of plants to unfavorable

environmental factors and pathogenic microorganisms (Barka et al., 2006; Solomon et al., 2014). The most important are both the synthesis of antibiotics by the genus *Bacillus* and *Streptomyces*, *Stenotrophomonas* sp., *Pseudomonas* sp., and the synthesis of various enzymes by some types of rhizobacteria (*Pseudomons* sp. and *Trichoderma* sp.) An important element in the prevention of pathogen colonization on the root is the synthesis of a wide spectrum of polysaccharides, which forming protective biofilm on the root surface (Gupta et al., 2015).

Numerous studies have confirmed the positive effect of the interaction of rhizospheric bacteria and roots on the different properties of the vine. Positive influence on the growth of vegetative organs, extinction and branching of the root, growth of the shoot and leaves has been stated by Barka et al., 2000; Compant et al., 2005; Rolli et al., 2012; Shaheen et al., 2013. The most important effect of biofertilization has been shown as the quality of grape cuttings and rootstocks observed over the callus, root and shoot development (Sabir, 2013). The positive effect of the application of various biological fertilizers on bud fertility and grape yields is stated by Hazarika and Ansari (2007). The direct influence of fertilizer application on rizobacteria on the grape structure is given by El-Sabagh et al. 2011.

This study was carried out to evaluate the effect of bacterial fertilizer on vegetative growth, yield and fruit quality on young potted grapevine.

MATERIALS AND METHOD

The study was carried out during 2015 and 2016 growing seasons on the potted grapevine grafts of cv. Cabernet Sauvignon on Kober 5BB rootstock. A field study was conducted at the experimental garden of the Faculty of Agriculture, University of Belgrade, Serbia (44^{o}50' N, 20^{o}24' E). Experimental grapevines grafts were planted in 20 plastic pots (20 l) containing of the pre-prepared substrate Terracult TC8, with added perlite (10% of total vol.) and mineral fertilizer Osmocote (30 g per pot). A treatment with bacterial fertilizer was prepared by adding of the BactoFil B10 (AgroBio, Hu.) solution (25 ml / 0.5 l H_20) and adding this solution two more times during the growing season at the intervals of 30 days. BactoFil B10 is a commercial microbiological fertilizer composed of nitrogen-fixing and phosphorus mineralizing bacteria (*Azotobacter vinelandii, Azospirillum lipoferum, Azospirillum brasilence, Bacillus megeterium, Bacillus subtilis, Bacillus cirkulans, Bacillus polymixia, Micrococcus roseus and Pseudomonas fluorescens*). According producers specification, the bacterial titer in the preparation is 5.2×10^{9} mL^{-1}. The control variant was prepared with planted grafts in a same substrate without adding a bacterial fertilizer. The pots with grafts were randomly divided into two rows with 2.5 $\times$ 1 m pot spacing, and North-South orientation of the rows. After that, trellis from poles and wires was placed over the pots and vines were spur-pruned on the two buds. During the growing season, the main shoots were tied to wires in an upright position, allowing the undisturbed and uniform development of the main and lateral shoots. During the summer, the vines were kept under net mesh having 70% of the sun transmission and drip irrigation was optionally included according the moisture sensor readings. Optimum protection against pest and diseases was applied.

A single leaf area, the main shoot leaf area and the lateral shoot leaf area were determined by using the statistical model as suggested by Lopes and Pinto (2000) and modified by Beslic et al. (2010). Estimating the area of an individual leaf was based on a formula obtained by regression analysis that uses the sum of the length of two inferior leaf veins as an independent variable. To estimate the leaf area per main shoot and per lateral shoot we used the model derived from multiple regression analysis that has the following independent variables: the number of leaves, surface area of the largest and of the smallest leaf on the shoot.

The vine vegetative growth expressed as dormant pruning weight of mature shoots was determined by measuring the weight of all shoots per vine using a hand-held digital scale.

In the second growing season, at the time of full ripeness of grape, all bunches were counted from each vine and weighed to determine yield and average bunch weight. Berries were then removed, counted and berry weight per bunch was calculated.

Must quality was determined on base concentration of the total soluble solids content (SSC) and titratable acidity (TA). A digital refractometer (RS-500 Atago, Japan) was used to determine SSC, and TA was measured by titration with 0.1N NaOH to a pH 8.2 and point.

All data were analysed using the analysis of variance (ANOVA). Treatment effects were compared using mean separation by LSD and polynomial contrasts. Regression analysis was conducted to determine the relationship between different factors. All analyses were performed using the Statgraphics Plus 5.1 (Statistical Graphics Corp. 2001.). All reported correlation coefficients were significant at the p=0.05 level.

RESULTS AND DISCUSSIONS

The vegetative growth was expressed as leaf area per vine, shoot dimensions, dormant shoot pruning weight and root weight, and some of these parameters were affected by biofertilization, (Table 1). The total leaf area per vine was significantly higher for 9% and 10,6% in treatment with biofertilizer by comparing to control while differences in the single leaf area were not recorded. The average shoot length was increased by a maximum 6% (2015), while no effect on shoot diameter in both seasons. The dormant shoot pruning weights were 17,2 and 21,6% lower in the control treatment by the end of 2015 and 2016, respectively. Differences in fresh root weight were not recorded. By utilization of bacterial fertilization, the biological properties of the pot's substrate were improved, which had a positive effect on the accessibility of nutrients and the synthesis of other stimulating compounds (Khalil 2012, Shaheen et al., 2013). The activity of the symbiotic and non-sybiotic azotofixators from BactoFil (*Rhizobium*, *Azospirillum*, *Azotobacter* etc.) affected the increase in the availability of growth promoting accessible N in the rhizosphere. Consequently, due to the direct and indirect effects of the rhizospheric bacteria, there was a positive effect on the growth of vegetative organs, the extinction and branching of the root, the growth of shoots and leaves.

Table 1. The elements of vegetative growth of the potted grapevine Cabernet Sauvignon

	Single leaf area (cm^2)	Total leaf area (m^2 per vine)	Shoot length (cm)	Shoot diameter (mm^2)	Pruning weight (g per vine)	Fresh root weight (g per vine)
2015						
Bactofil	66.45	0.678^a	147.5^a	7.4	123.8^a	-
Control	65.28	0.617^b	138.6^b	8.4	105.7^b	-
Lsd (0.05)	0.01158	0.04941	0.08135	0.1252	0.03230	
2016						
Bactofil	65.82	0.695^a	145.2^a	7.3	123.7^a	98.8
Control	65.20	0.621^b	139.2^b	7.4	101.7^b	107.0
Lsd (0.05)	0.01224	0.02555	0.07448	0.1883	0.09846	8.6934

Means separated by LSD multiple range tests ($p \leq 0.05$). Data followed by different letter in each column are significantly different.

The second vegetation was first yielding year. The results of the Bactofil effect on yield, grape and must quality are shown in Table 2. Treatment with BactoFil was not influenced on the differences in the yield, grape and berry weight. Also, treatment was not influenced on the must quality which is expressed over the dry matter content and total acid content.

Table 2. Effect of bacterial fertilization on yield, grape and berry quality of potted grapevine on cv. Cabernet Sauvignon

	Berry weight (g)	Bunch weight (g)	Yield (g)	Dry matter (%)	Total soluble solids (g per vine)
Bactofil	1,34	32,3	170	21,4	7,2
Control	1,26	32,5	171	21,8	7,3
Lsd (0.05)	0,91115	0,88625	1,21845	0,65542	0,44422

The lack of significant influence of BactoFil on yield elements and grape quality is in contrast to the results of numerous studies in which the positive effect of the application of various bacterial fertilizers was confirmed (Hazarika and Ansari, 2007; El-Sabagh et al., 2011). Nagy and Pinter (2015) obtained the significant increase of the bunch and berry weight, but no effect on grape quality. In most studies, the fertilizers were used on the oldest grape vines that are already in the phase of regular yield. In our case, the two-year potted vines were examined, whereby the increased contents of K, P and main microelements in the leaves were not recorded in relation to the control vines (results not presented). It is well known that these elements influence on the bud fertility and grape quality.

CONCLUSIONS

The application of bacterial fertilizer - BactoFil on young potted vine showed significant effect only on some elements of the vegetative growth: average shoot length, dormant shoot pruning weights, and total leaf area per vine. The activity of the symbiotic and non-symbiotic azotofixators from BactoFil affected the increase in the availability of growth promoting accessible N in the rhizosphere. Treatment with BactoFil was not influenced on the differences in the yield, grape and berry weight and must quality.

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