

Effects of biostimulative fertilizers on the quality of *Rubus idaeus* L.

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Abstract

The aim of this paper is to analyse the influence of foliar application of a biostimulative fertilizer on some of the elements of raspberry fruit quality of the Polka variety. The research was conducted in 2015, according to the system of controls and treatment. Slavol VVL, a foliar fertilizer with biostimulating effects was applied for treatment. A total of 12 quantitative and qualitative properties were analyzed depending on the influencing factor, namely: total sugar content, reducing sugars, invert sugars, sucrose, water content, dry matter, total acidity, vitamin C, total phenols, total flavonoids and antioxidant capacity, and fruit weight. After the completed analyzes, it can be concluded that raspberry plants treated with Slavol VVL were characterized by the highest values of total acidity (2.07%), dry matter (14.86%), and vitamin C content (25.15 mg/100 g of fresh weight).

Key words: Polka, fertilizers, biostimulators, Slavol VVL, control

Introduction

Raspberries have long been known for their healing properties, which is confirmed by Ward (2011) in his work, who states that the ancient Greeks and Roman observed that *Rubus* plants and its fruits collected in nature were used for medicinal purposes in fresh or processed condition. In the territory of Bosnia and Herzegovina, tradition of intensive growing is not long, as it was only introduced

in 2006 (Hodžić, 2013). Owing to the quality fruits, high yield, and simple cultivation, Bosnian producers quickly accepted this fruit's horticulture named *Polka* (Hodžić, 2013).

Grajkowski and Ochmian (2007), on the other hand, researched and examined the influence of three types of biostimulative fertilizers (Atonik SL, Biochikol 020 PC, and Tytanit) on the yield and fruit quality of three Polish raspberry varieties: *Pokusa*, *Polka*, and *Poranna rosa* during 2006-2007.

The aim of this study is to verify the impact of a biostimulative fertilizer, as well as its effect on the quality elements of the *Polka* raspberry. The specific goal of this paper is to analyse the effect the biostimulative fertilizers have on the quality elements of raspberry fruit, which is genetically predisposed to mostly use potassium, nitrogen, and phosphorus. After a statistical analysis of the results, certain conclusions and recommendations were made pertaining to the increase of the effectiveness of fertilizers in order to increase yields, as well as the rationality and economic justification of its use.

Material and methods

The experiment was conducted on a private raspberry plantation of the *Polka* variety in 2015. The raspberry orchard was planted in the fall of 2011 on the plot "Ribića Otoka" which is located in the village Bakšaiš, near the center of Bihać, with a total area of approx. 9.5 ha. Of the total area of this plot, the *Polka* raspberry variety occupies an area of 1.5 ha. Dry matter content was determined by a refractometric method (AOAC, 1995), and the water content of the raspberry fruit was determined by the drying method at 105°C. The content of reducing and total sugars was determined by the Luff Schoorl volumetric method (Reff) (sucrose content is obtained by multiplying invert sugar by 0.95). The content of total acids is determined by the titration method with an indicator (AOAC, 2002). The titrimetric method with 2,6-dichlorophenol-indophenol (AOAC, 2002) was used to determine vitamin C (L-ascorbic acid). Determination of total phenols and flavonoids in raspberry fruits was performed by the UV/ VIS spectrophotometric method (Ough and Amerine, 1998). The FRAP (Ferric Reducing Antioxidant Power) method was used to determine the antioxidant activity of raspberry fruit (Reff).

Measurement of fruit weight, chemical analysis of fruit, and determination of vitamin C was performed from fresh matter. The content of total phenols, flavonoids, and antioxidant capacity of raspberry fruit was determined from dry matter. Fruit sampling was performed at full harvest, in late July and early August. Raspberry fruits were taken from all parts of the plant, in order to avoid possible mistakes that may occur due to various microecological factors. The harvested fruits were placed in previously prepared crates in such a way that each

crate separately represented an average sample of fruits of one tested variant and transported to the laboratory.

Raspberries are grown in a trellis system of free individuals with a type of backrest commonly called a T-system.

This method of cultivation means that raspberry shoots grow freely in a zone 60 to 80 cm wide (30-40 cm on both sides of the middle of the row) along the entire row, without a clearly defined number of increments per meter. Up to 20 shoots per meter were usually formed.

According to the Basic Pedological Map, this area was mapped as an alluvial-carbonate sandy soil on sands. It is located in the lower parts of the valley along the Una River and often alternates alluvially with alluvial, carbonate, loamy, and clayey poorly gley soils. The characteristic of the land in this locality is high natural drainage, the water is lost very quickly so the soil is very drained and dry.



Fig. 1. Experimental orchard on the plot "Ribička Otoka" (Source: <http://katastar.ba/>, 2019).

Raspberries were planted in specially separated fields for control or treatment, for easier fertilization control and monitoring of plant growth stages.

Fruit sampling was performed at full harvest, in late July and early August. The samples were taken two times and the paper shows the average values of the obtained results. Harvesting was performed on each marked field and the harvested samples have been treated with the Slavol VVL fertilizer, and then combined into one average sample, as well as for the control field. Raspberry fruits were harvested from all parts of the plant, in order to avoid possible mistakes which may occur due to various microecological factors.



Fig. 2. Fruits harvested from the field treated with the Slavol VVL fertilizer (Photo: Handanović, 2015).

The obtained results of average values of the examined chemical properties of the Polka raspberry fruit are presented with standard deviation and results of the statistical data processing by using the One-Way ANOVA and Tukey's test; primarily due to the volume of results and a large number of individual tables.

The research was set up and statistically evaluated according to the split plot scheme, where the main one is the fertilization factor, and subfactor variant of biostimulator application, with three repetitions. All determined results were analyzed by usual methods of statistical data processing - analysis of variance (ANOVA), statistical significance test impact of applied treatments - F test and Tukey's test. In addition to the analysis of variance, the analysis of principal components (PCA - Principal Component Analysis) Hotelling was performed, based on the correlation matrix in the PAST computer program. The principal components analysis (PCA) was performed using mean values of quantitative traits.

Research results and discussion

Chemical properties of the Polka raspberry fruit

The results of all 12 mentioned parameters are presented in three tables (chemical, antioxidant properties, and fruit weight), according to the statistical processing data in line with the treatment with the Slavol VVI biostimulative fertilizer and the control treatment.

As Table 1. shows, a statistically significant difference was found in all observed chemical properties of the *Polka* raspberry fruit (total sugars, reducing sugars, inverted sugars, water content, dry matter content, and total acidity).

Tab. 1. Average values of chemical properties of the raspberry fruit

Treatment	Total sugars	Reducing sugars	Invert sugars	Sucrose	Water	Dry matter	Total acidity
Slavol VVL	4.61 ± 0.23 ^a	4.14 ± 0.14 ^b	0.52 ± 0.30 ^a	0.44 ± 0.23 ^a	85.14 ± 0.27 ^a	14.86 ± 0.27 ^a	2.07 ± 0.16 ^a
Control	5.46 ± 0.28 ^b	4.04 ± 0.24 ^b	1.42 ± 0.21 ^b	1.35 ± 0.20 ^b	86.20 ± 0.27 ^b	13.80 ± 0.27 ^b	1.60 ± 0.09 ^b
ANOVA	p ≤ 0.05	p ≤ 0.05	p ≤ 0.05				

Lab. a, b - values marked with the same letter do not differ statistically.

Based on the data from Table 1, a statistically significant difference was found in the observed chemical properties of the Polka fruits depending on the treatment. It was shown that the factor of the foliar fertilizer with biostimulative effects impacts some observed chemical properties of the Polka fruits ($p \leq 0.05$). The content of total sugars in the fruits of the Polka raspberry, depending on the foliar treatment of two types of biostimulant fertilizers and the control, ranged from 4.61% (Slavol VVL) to 5.46% (control and humus). Reducing sugars in raspberry fruit ranged from 4.04% (control) to 4.14% (Slavol). Higher content of invert sugars and sucrose was obtained in fruits from the control (1.42% and 1.35%), and lower from the field treated with Slavol VVL (0.52% and 0.44%).

In the studies conducted by Skupien et al. (2011) on the *Polka* fruits, grown organically and conventionally in different locations of western Pomerania (Poland), the content of total sugars was 5.82% in the organic sample. As opposed to the research conducted and published by Nikolić et al. (2008) during the 2006-2007 period on the *Polka* raspberries fruits, grown by conventional agricultural methods, which registered significantly higher values of the total sugar content in the fruit (7.10% and 8.42%). An average higher content of inverted sugars (6.82%) was thus observed and published by Nikolić et al. (2008) in their research study of the *Polka* raspberry fruit grown by using conventional horticultural methods over the experimental period of two years.

Based on the obtained research results, the highest dry matter content was recorded in raspberry fruits from the fields that were treated with the Slavol VVL foliar biostimulative fertilizer (14.86%), and the dry matter value in raspberry fruits from the control was 13.80%. The previous studies conducted by Nikolić et al. (2008); Lewandowski et al. (2015); Milivojević et al. (2011); Musić (2011); and Skupien et al. (2011) showed that the dry matter content in the raspberries fruit, grown using conventional methods, ranged from 10.04% to 14.49%.

Considering the average content of total acids in the fruits of the Polka raspberry cultivar, the highest was detected in fruits from fields treated with Slavol VVL (2.07%). Control fruits contained lower values of total acid content (1.60%).

The content of total acids, depending on the level of the applied biostimulative fertilizers, ranged from 1.68% (Biochikol 020), over 1.70% and 1.79% (control and Tytanit) to 1.82% (Astonik SL). These values are in the range of the obtained values of this study, with a significantly higher content of total acids recorded in this study in the fruits analyzed from the fields treated with Slavol VVL (2.07%). In a large number of studies, the content of total acids in raspberry fruit grown by using conventional horticultural methods, ranged from 0.8% to 1.87% (2011), as shown in the studies conducted by Nikolić et al. (2008); Skupien et al. (2011), which is significantly less than the value of total acid content in raspberry fruits harvested from the fields treated with Slavol VVL, and consequently analyzed during this study.

Antioxidant properties of the *Polka* raspberry fruit

Tab. 2. Average values of antioxidant effects of the raspberries fruit

Treatment	Vitamin C (mg/100 g FW)	Total phenols (mg / g DW, expressed as gallic acid)	Total flavonoids (mg/g DW)	Antioxidant capacity (μ mol Fe ²⁺ /g DW)
Slavol VVL	25.15 \pm 0.19a	10.14 \pm 0.35a	3.33 \pm 0.10a	12.13 \pm 0.40a
Control	22.18 \pm 0.10b	12.25 \pm 0.18ab	3.87 \pm 0.07b	14.18 \pm 0.28b
ANOVA	p \leq 0.05	p \leq 0.05	p \leq 0.05	p \leq 0.05

Lab. a, b - values marked with the same letter do not differ statistically.

Based on the results of the one-way analysis of variance (One-way ANOVA), it can be concluded that the treatment with a foliar biostimulation fertilizer affected the content of vitamin C.

The application of the Slavol VVL biostimulative fertilizer had a positive effect on the content of vitamin C, while for the other examined parameters higher values were in the control field.

When it came to the vitamin C levels, Skupien et al. (2011) obtained similar results. When analyzing the *Polka* raspberry fruit, grown by organic methods, their study showed vitamin C levels at 21.9 mg/100 g FW, whereas the *Polka* raspberry fruit, grown by conventional methods, showed vitamin C levels at 25.3 FW. The highest average content of vitamin C in the *Polka* raspberry fruit was registered in the fruits harvested from the fields treated with Slavol VVL (25.15 mg/100 g FW), whereas lower values of vitamin C were found in the fruits harvested from control fields (22.18 mg/100 g FW). The influence of three types of biostimulative fertilizers (Atonik SL, Biochikol 020 PC, and Tytanit) on the yield and fruit quality of three polish raspberry varieties: *Pokusa*, *Polka*, and *Poranna rosa* was conducted during the 2006-2007 period. In that paper,

Grajkowski and Ochmian (2007) discovered significantly higher content of vitamin C levels, which ranged from 43 mg/100 g FW (Tytanit), over 47.00 and 48.00 mg/100 g FW (Astonik SL and Biochikol 020) to 59.00 mg/100 g FW in the fruits harvested from the control field.

The content of total phenols in the Polka raspberry cultivar, depending on the influential treatment, ranged from 10.14 mgGAE/Gdw for the fields treated with Slavol VVL to 12.25 mgGAE/gDW for the fruits from control fields. Higher values of total phenol content compared to these studies were published by Kazimierczak et al. (2015). The content of total phenols in this study for freeze-dried raspberry cultivar in organic cultivation was 18.64 mgGAE/g DW, and in conventional cultivation the content of total phenols was 15.17 mgGAE/g DW. Observing the content of total flavonoids in the dry matter, a higher content was registered in the control field (3.87 mg/g DW), and lower values in the fields treated with Slavol VVL (3.33 mg/g DW). Assessing the vegetative growth and fruit chemistry of some raspberry and blackberry varieties in southern Poland, Orzeł et al. (2016) in their research reported that the content of total flavonoids in three experimental years in fresh matter of the Polka fruit ranged from 19.90 mg/100g FW for 2011, 27.77 mg/100g FW for 2012, and 22.71 mg/100g FW for 2013.

The obtained values of antioxidant capacity indicate better reduction ability measured in fruits from the control field (14.18 $\mu\text{mol Fe}^{2+}/\text{g DW}$), and a lower registered value of total flavonoids was recorded in the fruits from the fields treated with Slavol VVL (12.13 $\mu\text{mol Fe}^{2+}/\text{g DW}$).

The weight of the *Polka* raspberry fruit

Tab. 3. Average values of the weight of the raspberry fruit

Treatment	Weight of the fruit (g)
Slavol VVL	3.35 ± 0.42 ^{ab}
Control	3.11 ± 0.45 ^b
Anova	$p \leq 0.05$

Lab. a, b - values marked with the same letter do not differ statistically.

Based on the data from Table 3, a statistically significant difference was found in the average fruit weight depending on the treatment. It has been shown that a factor of different types of foliar biostimulating fertilizers affects the fruit weight of the *Polka* raspberry fruit ($p \leq 0.05$). Higher fruit weight was found in the experimental fields treated with Slavol VVL (3.35 g), while in the controls it was 3.11 g. Significantly higher values of fruit weight expressed by the weight of 100 fruits were published by Grajkowski and Ochmian (2007) when

examining the influence of three types of biostimulative fertilizers (Atonik SL, Biochikol 020 PC, and Tytanit). The obtained values for the weight of 100 representative fruits of the *Polka* raspberry fruit were published in a study conducted by Krok and Wieniark (2008), who investigated the effect of the biostimulative fertilizer Goemar BM 86 (developed from an algae extract) on the development and fruit quality of dicotyledonous raspberry varieties, and are very much in accordance with the weight values obtained in this study.

Conclusion

Based on the results of one-year research, it can be concluded that the application of the Slavol VVL biostimulative fertilizer had positive effects in relation to the control of certain examined parameters. This is reflected in the higher values of fruit weight, reduced sugar content, dry matter, and total acidity, which is evident in the research results. The other examined parameters (total sugars, invert sugars, sucrose, water content, total phenols, total flavonoids, and antioxidant capacity) have higher values in the control field compared to the field treated with the Slavol VVL biostimulative fertilizer.

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Утицај биостимулативних гнојива на квалитет малине (*Rubus idaeus* L.)

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Сажетак

У овом раду анализиран је утицај фолијарне примјене биостимулативног гнојива на неке од елемената квалитет плода малине сорте Полка, на подручју града Бихаћа. Истраживање је проведено у 2015. години, по систему контрола и третман. За третман је примјењено фолијарно гнојиво са биостимулативним дјством Славол VVL. Укупно је анализирано 12 квантитативних и квалитативних особина у зависности од утјецајног фактора, а то су: садржај укупних шећера, редукујућих шећера, инвертних шећера, сахарозе, садржај воде, сухе твари, укупне киселости, витамина Ц, укупних феноли, укупних флавоноида и антиоксидативни капацитет и маса плода. Након завршених анализа може се закључити да су се биљке малине третиране Славолом VVL одликовале највишим вриједностима укупне киселости (2,07%), сухе твари (14,86%) и садржаја витамина Ц (25,15 mg/100 g свјеже масе).

Кључне ријечи: Полка, гнојива, биостимулатори, Славол VVL, контрола

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