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EFFECTS OF ALGAE PRODUCTS ON NUTRIENT UPTAKE AND FRUIT QUALITY OF APPLE

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Abstract

Nowadays the main task of scientists and farmers is to find natural ways to avoid negative effects of climatic anomalies and improve plant productivity lead to environmental friendly agriculture.

Biostimulants have a great potential to achieve these aims but unfortunately there is little information about its application in apple growing mostly in East Hungary.

For this reason, foliar nutrition experiment was made in the region of Nyírség (East Hungary) to investigate the effect of different biostimulants (algae products) on yield, leaf nutrient concentration and quality parameters of cv. 'Gala Must' apple (Malus domestica Borkh.) variety. The study was conducted in 2012 at Nyírbátor in East Hungary in a 14 years old apple plantation. Treatments (application time and doses) were adjusted to the phenological phases of apple and the control was used an untreated check. Effect of treatments was monitored by leaf diagnosis and apple quality measurements.

The results demonstrate that the treatments increased the external fruit parameters (diameter, weight, shape index) but not affected consequently the leaf macronutrient status compared to the control. We suppose that, stable treatment effect on leaf nutrient status can be observed in long-lasting experiment only. The applied products significantly increased the amount of flavonoid and phenolic compounds and water soluble antioxidant capacity value compared to the control. Our fruit analysis results supported that the applied biostimulators had no effect on fruit acid and ash content. Moreover, the applied products resulted higher sugar, vitamin C and dry matter content despite the unfavourable, very dry climatic conditions. In sum, results showed that foliar application of biostimulants had a positive effect on yield and resulted bigger and healthier therefore more marketable fruits.

Key words: algae based biostimulants, fruit nutrition, fruit quality, nutrient uptake

INTRODUCTION

Global demographic pressure and unexpected climatic events and their growing rate on agricultural production calls for novel and sustainable approaches toward satisfying the ever-growing demand for plant biomass destined for human food, animal feed, and energy production. Conventional agricultural practice has relied overwhelmingly on non-renewable inputs of fertilizers and pesticides (Calvo et al., 2014).

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Currently, legislation restricts the use of mineral fertilizers and pesticides and thus forces a new approach to reducing the use of chemical products through either parallel application or partial replacement with formulations capable of enhancing the efficiency of conventional treatment. Feeding a growing population requires yield increases and enhanced crop quality, both of which are fostered by biostimulants (European Biostimulant Industry Council (EBIC), 2012; Jardin, 2015; Chiaiese et al., 2018).

Plant biostimulants (PBs) attract interest in modern agriculture as a tool to enhance crop performance, resilience to environmental stress, and nutrient use efficiency (Bulgari et al., 2014).

According to recent EU Regulation, PBs are defined mainly through their claimed action, therefore PBs encompass diverse organic and inorganic substances (humic acids and protein hydrolysates) as well as prokaryotes (e.g., plant growth promoting bacteria) and eukaryotes such as mycorrhiza, N-fixing bacteria and macroalgae (seaweed) (European Commission, 2016; Yakhin et al., 2017; Chiaiese et al., 2018).

Among the natural materials of such capability are algae, which contain a variety of biologically active compounds verified to have a beneficial influence on plants (Balconi, 2012; Dmytryk, Chojnacka, 2018). Algae are increasing crops' performance, optimizing qualitative traits, reinforcing abiotic stress resistance and recovery, give greater profitability for the farmers. Biostimulants can enhance quality attributes of produce, including sugar content, colour, fruit seeding, etc. Enhanced quality can mean higher incomes for farmers, better storage and more nutritious food for consumers (Khan et al., 2009; Khan et al., 2012; Battacharyya et al., 2015).

Biostimulants foster plant growth and development throughout the crop life cycle from seed germination to plant maturity in a number of demonstrated ways, including but not limited to:

- Improving the efficiency of the plant's metabolism to induce yield increases and enhanced crop quality;
- Increasing plant tolerance to and recovery from abiotic stresses;
- Facilitating nutrient assimilation, translocation and use;
- Enhancing quality attributes of produce, including sugar content, colour, fruit seeding, etc.;
- Rendering water use more efficient;
- Enhancing soil fertility, particularly by fostering the development of complementary soil micro-organisms.

Moreover, biostimulants help protect and improve soil health by fostering the development of beneficial soil microorganisms. Healthier soil retains water more effectively and better resists erosion (Dudás et al., 2017).

To handle limiting factors and to insure high quality crops in the future lot of authors recommend the using of plant biostimulants to enhance nutrition efficiency, abiotic stress tolerance and/or crop quality traits, regardless of its nutrients content (Khan et al., 2009; Balconi, 2012; European Biostimulant Industry Council (EBIC), 2012; Battacharyya et al., 2015; Jardin, 2015). Their use provides an opportunity for growers to mitigate and correct the increasing effects of abiotic stress situations.

Furthermore, biostimulants contribute to socio-economic development. By making existing agricultural practices more efficient and improving post-harvest storage, biostimulants help reduce waste throughout the agri-food chain. Less waste means lower costs, which ultimately benefit the consumer who has access to high-quality, affordable food (European Biostimulant Industry Council (EBIC), 2012). The proper orchard management practises are the main key factors in the production of high and qualitative yields of fruits (Bramlage, 1993; Nagyné Demeter, 2010; Nagy et al., 2016).

In Hungary, in recent years, there has been a growing perception of the strengthening of the ecological approach in fruit nutrient management (Demeter, 2014), which requires the use of environmentally conscious cultivation technologies based on biostimulants.

The aim of this paper is to provide further data about biostimulants applying and their effects on yield and fruit quality. We wanted to study how effect the algae treatments on the mineral uptake of apple trees and the internal and external parameters of apples.

MATERIAL AND METHOD

The study was performed at the orchard of F.N. Fruit Ltd. at Nyírbátor in 2012. Comparison and evaluation of the effects of different biostimulants were performed in our study. The orchard was planted in 1998, grafted on M26 rootstock. Spacing between and within rows was 4.85 x 1.6 m. The apple cultivar was Gala Must. The orchard has drip irrigation system.

Applied treatments

In the experiment, beside the control, four algae biostimulants (Globalga, Goemar BM 86, Organic Green Gold (OGG) and Wuxal ascofol) were used to test their effects on fruit yield and quality.

Globalga is a reddish-brownish seaweed liquid, pH is 6.5 (in 10 % solution), contains: 7.0 % N and P_2O_5, 4.0 % K_2O, 6.0 % amino acids and EDTA as additives.

Goemar BM 86 is basically Ascophyllum nodosum, contains GA 142 algae cream, 1.67 % N, 9.6 % SO_3, 4.8 % MgO, 0.02 % Mo and 2.0 % B.

OGG is basically Chlorella vulgaris, green suspension, pH is 6.25 dry matter contain is 1 %, contains: 0.15 % N, 0.29 % P_2O_5, 0.25 % K_2O, 0.035 % Ca, 0.02 % Mg, 0.008 % B and 0.015 % Fe.

Wuxal ascofol is 50 % algae suspension, contains: 2.3 % N, 1.5 % K_2O, 0.195 % CaO, 0.033 % MgO, 3 % B, 0.005 % Fe, 0.5 % Zn, iodine, plant hormones.

The treatments were set up in three replications. Twenty trees per replication were treated. Algae products were sprayed on the foliage of the selected trees by a motorized knapsack sprayer. Applied dosages and the circumstances* of the application are showed in Table 1.

Table 1

Time and dosages of the applied treatments (l/acre)

Phenological stages	Application Time	Globalga	Goemar BM 86	Organic Green Gold	Wuxal ascofol
half blooming	26th April	2.0	3.0	3.0	-
Full blooming	30th April	2.0	3.0	3.0	10.0
Petal falling	4th May	2.0	3.0	3.0	-
2 week after full blooming	11th May	2.0	3.0	3.0	-
3 week after full blooming	18th May	-	-	-	10.0
4 week after full blooming	27th May	-	-	-	10.0

*- treatments were adjusted to the instructions of the manufacturers

Soil sampling and preparation and results

As the root system was most concentrated in the upper layer of the soil, soil samples were taken from 0-30 and 30-60 cm layers of the soil by using manual soil sampling equipment as described in Jackson (1958) using the Hungarian standard method MSZ-08 0202-77. Sampling was performed before the experiments were set up. The samples were dried, sieved, homogenized and stored in plastic boxes until the examination.

Soil pH was determined from a soil solution of 0.01 M $CaCl_2$. Plasticity index (K_A) and humus content were measured according to Hungarian guideline (MSZ 20135:1999). Nitrogen forms of each soil sample were quantified according to Houba et al. (1986). For extracting the available P and K content of soils, ammonium-lactate solution (so called AL extractant) was used, then the amount of phosphorus was quantified colorimetrically with the phospomolybdovanadate method (Hungarian standard MSZ 20135:1999). Potassium content was quantified by flame atom emission spectrophotometry (Hungarian standard MSZ 20135: 1999). For determining Ca, Mg, Mn, Cu and Zn contents of the soil Lakanen-Erviö solution (LE) was used (Lakanen, Erviö, 1971). Soil Ca, Mg, Mn, Cu and

Zn contents were quantified using flame atomic absorption spectrophotometry (Hungarian standard MSZ 20135: 1999).

The results of soil analysis are showed in Table 2. Orchard soil type was slightly acidic, non-calcareous sandy soil with very low humus content. The pH of soil was near neutral and slightly decreased by the depth. Water capacity of soil was low according to the soil type. The texture grade of soil was sandy according to the soil plasticity index (K_A) (Table 2).

Table 2

Results of soil analysis (Nyírbátor, 2012)

Parameters	Depth (cm)	
	0-30	30-60
pH (KCl)	7.47	6.38
Water soluble salts (%)	< 0.02	< 0.02
Plasticity index (K_A)	29	27
Humus content (%)	1.001	0.557
(NO_3+NO_2)-N (mg/kg)	9.12	4.61
P_2O_5 (mg/kg) (AL)	746	216
K_2O (mg/kg) (AL)	164	89.4
Mg (mg/kg)	80.5	66.2
Mn (mg/kg)	67.8	98.5
Cu (mg/kg)	4.586	1.594
Zn (mg/kg)	8.93	1.898

The soil organic matter content was low and decreased by the depth. The N-supply of the soil was medium, which was godd correlation with the measured mineral N-forms. The mineral N fraction of the soil was dramatically decreased by the depth (Table 2).

Carbonate content of soil was not detectable. Available soil P (AL soluble) was high mostly in the upper layer of the soil. Available soil K content (AL soluble) was low and decreased by the depth. These results pointed out that the macronutrients were concentrated in the upper layer of the soil. Soil Mg and micronutrient contents were suitable for fruit growing (Table 2). The data on micronutrient contents correspond to the values characteristic to sandy soil with low humus content and pH value.

Leaf sampling and preparation

The leaves of the selected cultivar were used for plant sampling. Leaves were taken from twenty trees from the treatment plots at the standard sampling time (at the beginning of August). For sampling, healthy, well-developed, mature leaves (twenty leaves per replication) were taken from the mid-third portion of extension shoots of the current year as described in the international and Hungarian plant sampling guidelines for fruit orchards (Stiles, Reid, 1966; Hungarian standard MI-08 0468-81).

Fruit analysis

The concentration of flavonoids was measured by Kim et al. (2003), the total phenolic compounds were determined by Singleton and Rossi (1965) while the total water soluble antioxidant capacity (FRAP value) was evaluated according to Benzie and Strain (1996).

The soluble solid content of fruits (SSC) was measured by handle refractometer Brix (MT-032ATC, detection limit: ±0.20%) (MSZ EN 12143:1998), the titratable acid content was measured by potentiometric titration according to Hungarian standard (MSZ ISO 750:2001). The dry matter content of fruits was determined by loss-ignition method. The ash content of fruits was determined according to Hungarian standard MSZ ISO 5520:1994. The vitamin C content was measured by iodine titration.

Statistical analysis

All the obtained data were tabulated and statistically analyzed according to Svab (1981) using the L.S.D. test at 5% level to recognize the significance of the differences between various treatment methods. The effects of the different treatments were assessed within ANOVA and Fisher's least significant differences were calculated following a significant ($P \leq 0.05$) F test.

RESULTS AND DISCUSSION

Results of leaf analysis

Results of leaf analysis were shown in the Table 3.

Foliar application of Wuxal and OGG significantly influenced the N content of apple leaves. The other treatments not affected significantly leaf N content (Table 3).

Table 3

Results of leaf analysis (Nyírbátor, 2012)

Treatments	N (%)	P (%)	K (%)	Ca (%)	Mg (%)
Control	2.24 a	0.177 b	1.34 b	1.84 a	0.377 a
Globalga	2.15 a	0.186 b	1.62 bc	1.96 b	0.356 a
Goemar BM 86	2.18 a	0.146 a	1.08 a	1.72 a	0.377 a
OGG	2.44 b	0.159 a	1.02 a	1.39 a	0.400 b
Wuxal Ascofol	2.39 b	0.163 a	1.00 a	1.50 a	0.391 a

In each column, means followed by the same letter are not significantly different ($P < 0.05$).

Leaf P, K and Ca were increased by the Globalga treatment only. Leaf P, K, and Ca content were lower when applying other treatments compared to the control (Table 3). Leaf Mg was significantly affected and increased by the OGG treatment only.

Independently of the treatments the leaf macronutrient status was optimal for all examined nutrients. It was found that the leaf macronutrient status was not affected by the treatments consequently. This result is highly similar to the findings of Khan et al. (2012) who reported that foliar treated grapevines (mixture of amino acids and seaweed extract) showed no significant change in the leaf mineral contents.

Results of fruit analysis

Fruit samples were taken at the time of full ripening (31th August). The results of fruit analysis (external parameters) are shown in Table 4. Fruit size (diameter), mean weight and shape index were measured as external fruit parameters. Fruit diameters were measured at two time: at the middle of June and at the end of August (picking time).

Table 4

Results of fruit analysis (external parameters) (Nyírbátor, 2012)

Treatments	Fruit diameter (mm) (15.06.)	Fruit diameter (mm) (31.08.)	Mean weight* (g)	Shape index**
Control	32.86 a	60.98 a	101.75 a	0.86 a
Globalga	36.31 c	65.93 b	126.00 c	0.88 b
Wuxal Ascofol	35.73b c	66.99 b	128.75 c	0.85 a
Goemar BM 86	36.38 c	67.82 b	132.75 c	0.84 a
OGG	34.51 b	62.54 a	112.50 b	0.86 a

In each column, means followed by the same letter are not significantly different ($P < 0.05$).
*- mean weight of 100 fruits
**- ratio of height and width

All applied treatments significantly affected the fruit diameter, except OGG at the end of August. Goemar BM 86 resulted the highest increment in fruit diameter (11.2%). All treatments had significant positive effect on the weight of fruits. The Goemar BM 86 treatment resulted the highest mean weight. The increment was 30.5% compared to the control. These results are similar to those obtained from pear by Colavita et al., 2011.

Goemar BM 86 tends to have positive influence in acceleration of ripening and increase of fruit size. These results are in harmony with those obtained by Krok and Wieniarska 2008, who use of any biostimulator in primocane raspberry growing under conditions of Poland. that respect, its effects are notconsistent, however, varying depending on cultivar and or season.

The results of fruit analysis (internal parameters) are shown in Table 5, 6 and 7. Dry matter, ash and ascorbic acid content of fruits are showed in Table 5.

All applied biostimulator increased the fruit dry matter but only the Wuxal treatment resulted significant effect on it. Similar results were obtained regarding to the vitamin C content of fruits. Treatments had not significant effect on the ash content of fruits (Table 5).

Total sugar and acid content of fruits are shown in Table 6. All treatments increased the total sugar content of fruits, except OGG. But only the Goemar BM 86 treatment increased significantly the total sugar content of apples. However, the treatments had not significant effect on total acid content of apples. Total acid content of apples was varied between 1.1 and 1.4 g/l.

Table 5

Results of fruit analysis (internal parameters – I.) (Nyírbátor, 2012)

Treatments	Dry matter (%)	Ash (%)	Vitamin C (%)
Control	13.18 a	0.30 a	1.47 a
Globalga	13.43 a	0.29 a	1.47 a
Wuxal Ascofol	15.81 b	0.37 a	3.38 b
Goemar BM 86	14.53 a	0.33 a	2.35 a
Green Gold	14.18 a	0.35 a	2.05 a

In each column, means followed by the same letter are not significantly different ($P < 0.05$).

Table 6

Results of fruit analysis (internal parameters – II.) (Nyírbátor, 2012)

Treatments	Total sugar content (g/l)	Total acid content (g/l)
Control	108.30 a	1.20 a
Globalga	108.59 a	1.10 a
Wuxal Ascofol	109.59 a	1.40 a
Goemar BM 86	112.46 b	1.10 a
OGG	106.87 a	1.30 a

In each column, means followed by the same letter are not significantly different ($P < 0.05$).

Flavonoids, phenolic compounds and FRAP values of fruits are shown in Table 7. Flavonoid concentration in fruits varied between 0.187 and 0.346. The highest value was observed at the Wuxal treatment. Similarly to Wuxal, OGG had a strong, significant effect on the amount of flavonoids.

Table 7

Results of fruit analysis (internal parameters – III.) (Nyírbátor, 2012)

Treatments	Flavonoids (mg katechine ekv./100g fresh weight)	Phenolic compounds (mg gallic acid ekv./100g fresh weight)	FRAP (mg ascorbic acid ekv./100g fresh weight)
Control	0.196 a	47.593 a	34.604 a
Globalga	0.187 a	50.311 b	34.287 a
Wuxal Ascofol	0.346 b	69.234 c	55.250 c
Goemar BM 86	0.202 a	59.563 c	35.215 a
OGG	0.250 b	51.483 b	40.088 b

In each column, means followed by the same letter are not significantly different (P<0.05).

Total phenolic content in fresh fruit samples was significant affected by all applied treatments. It means, that the biostimulators increased the phenolic concentration in the apple samples. The Wuxal treatment was the most effective. The Wuxal treatment resulted the highest FRAP value similarly to those founded at flavonoids and phenolic compounds. It seems that among the treatments the Wuxal treatment had the strongest effect on measured so called "healthy protective" compounds. These results are in harmony with those obtained by Karim and Rahim, 2008 and Abd El-Motty et al., 2010 at Mango trees.

CONCLUSIONS

Our investigation was set up in a 14 years old apple orchard, planted on an acidic sandy soil, among unfavourable soil and climatic conditions. Four algae suspension as biostimulants were used in this comparing study.

Similarly, to the findings of Khan et al. (2012), it was found that the leaf macronutrient status was not affected by the treatments consequently and significantly. Furthermore, longer experiment is needed to study the effect of these products on leaf nutrient status.

Applied algae products and their doses significantly affected the fruit diameter and weight. It confirms the earlier findings that the PBs are useful to improve yield (Bulgari et al., 2014; Calvo et al., 2014; Yakhin et al., 2017).

Significant fruit diameter increment was observed in the early phenological stage, near after the foliar application.

All treatments increased the dry matter and the vitamin C content of apples compared to the control but significant effect was measured by using Wuxal Ascofol. Moreover, applied biostimulants had no significant effect on fruit ash and acid content. These results confirmed the earlier findings (Vernieri et al., 2005) that the efficacy of the biostimulants depends from the timing of application. Since biostimulants activate specific biochemical mechanisms, it is important to identify the best application time. The optimal dose is also very important because within a certain range the crop can positively respond to biostimulants application. Therefore, it is important to define for each biostimulant the optimal application range, too high or low concentrations can nullify the biostimulant effect (Toscano et al., 2018; Vernieri et al., 2005).

Moreover, from these results it would be foolhardy to state that applying biostimulants in fruit growing provides greater health benefits than those produced without them, but we suggest that these comparison studies should be expanded. The real benefit of these studies is that they can identify and establish the production input weaknesses and strengths that affect nutrition, so that changes can be made to improve both organic and integrated fruit production technologies.

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