**ANNALS OF THE
UNIVERSITY OF CRAIOVA**

Series: ✓ *Biology*
✓ *Horticulture*
✓ *Food products processing
technology*
✓ *Environmental engineering*

Vol. XXV (LXII) - 2021

**RESEARCH ON THE EFFECT OF A PLANT BIOSTIMULANT APPLIED
TO VINEYARD (*CHASSELAS DORE VARIETY*)**

Cioroianu Traian¹, Sîrbu Carmen^{1*}, Grigore Adriana¹, Marin Nicoleta¹

¹National Research and Development Institute for Soil Science, Agro-chemistry and Environment Protection-RISSA, Bucharest, 61 Mărăști Blvd., 011464, District 1, Bucharest, Romania

* Correspondence author. E-mail: carmene.sirbu@yahoo.ro

Keywords: *plant biostimulant, algae, protein hydrolysate, foliar application, vineyard*

ABSTRACT

*In recent years, plant biostimulants have been increasingly used in agriculture. The paper focuses on a new plant biostimulant with a complex matrix of organic substances based on protein hydrolysates (a mixture of peptides and free amino acids), algae extract (*Ascophyllum nodosum*) and secondary nutrients, used in viticulture. The experimental biostimulant was tested in the vineyard (*Chasselas dore*) by foliar application, in a concentration of 0.5%, three treatments in the vegetative phase. The trials were performed in comparison to a non-fertilized control. The use of foliar biostimulant led to yield increases of 29.1% compared to the control. The application of the three foliar treatments with the experimental plant biostimulant increased by 25 - 36% the process of photosynthetic assimilation to the vineyard, cultivated on the Horticultural Antrosols.*

INTRODUCTION

In Romania, certified areas in organic farming have increased since 2016. This leads to a higher requirement of farmers for inputs from the class of fertilizers and protection products. The increasing trend of organically cultivated agricultural areas is evident in the last 5 years in Romania. However, only 1,531,000 ha are fully converted to organic farming and 1,331,000 ha are under conversion to organic farming (https://ec.europa.eu/info/food-farming-fisheries/farming/organic-farming/organic-action-plan_en). In Romania, this area represents only 3% of the total agricultural land (<https://www.madr.ro/docs/agricultura/agricultura-ecologica/2021/Dinamica-operatorilor-si-a-suprafetelor-agri-eco-update-28.06.2021.pdf>.) while at the level of the European Union organic agriculture is 8.5%. At present, there is a target on the European Green Deal at EU level for organic farming to be practiced on 25% of the agricultural land by 2030 (Brussels, 19.4.2021 COM(2021) 141 final/2).

The need to formulate and test new products with a role in plant nutrition that can be used in organic agriculture is obvious. The permitted inputs used in the organic farming system must comply with specific legislation but must also be effective.

The application of biostimulants on plants leads to the accumulation of a higher content of nutrients in their tissue and to positive metabolic changes. For these reasons, the development of new biostimulants has become a point of scientific interest (Nardi et al. 2016).

There are many examples and information that the application of plant biostimulants can improve not only the yield but also the nutritional quality of fruits (Rodrigues et al. 2020). The Annex I to Regulation EC 889/2008 with fertilisers, soil conditioners and nutrients contains algae and products from algae and protein hydrolysates of plant origin.

Application by spraying to cultures with biostimulant plant containing *Ascomyces nodosum* extracts (Frioni et al. 2018) and protein hydrolysates (Boselli et al. 2019) can have positive effects on the quality of cultivated wine grapes.

In order to support the effects of plant biostimulants, there must be experimental data generated under controlled conditions (laboratory, greenhouse, growth chamber, phenotyping, etc.) and / or in the field (field studies) (Ricci et al. 2019).

To evaluate the effect of the Eco Plus biostimulant on vineyard, it was applied 3 times at a dose of 2.5 L / ha and yield, influence on photosynthesis and nutrition were evaluated.

MATERIAL AND METHODS

Eco Plus is a plant biostimulant with the following composition: organic nitrogen 3,3-3,5%, potassium 7.7-8,0%, organic phosphorus 0,2 – 0,3%, copper 0.05 – 0.06%, zinc 0.05-0.01%, iron 0.03-0.05%, manganese 0.01-0.02%, boron 0.01-0.02% and organic matter 45-48% from algae extract (*Ascomyces nodosum*) and protein hydrolysate. The raw materials used to obtain this product are allowed to be used in organic farming (Commission Regulation (EC) No 889/2008).

Due to the complex organic structure and trace elements, Eco Plus activates the biochemical processes in plant tissues and has a direct influence on the vital functions of cells and their activity.

The trials were performed by applying the Eco Plus (0.5% solution) to the vineyard farm. The experiment consisted of three foliar treatments performed with solution by fine atomization on the entire foliar surface, as follows: the first treatment - after the flowering phenophase; the second treatment – during the growing phase of the berries, the third after 10 days after the second.

The main physical, chemical and biological properties of Horti Antrosols were: a fine clay soil texture (39% and 45.5% clay); pH – weakly acidic (6.22 pH units) at the surface and neutral (6.82 - 7.11 pH units) below 60 cm depth; humus 3.12 - 3.22%; total nitrogen (0.150 - 0.135%); mobile phosphorus 42 - 35 ppm at the surface and 30 -25 ppm below 40 cm depth; mobile potassium 218 - 195 ppm at the surface and 140 -135 ppm below 40 cm depth, and base saturation of 85% at the surface and 90% below 40 cm depth.

The effects of the treatments were studied by analysis of variance (Fischer method) and Fisher's Least Significant Difference (LSD) test. All data are relative values as compared with the control (treated only with water) considered equal 100%.

RESULTS AND DISCUSSIONS

The plant biostimulant (Eco Plus) was characterized and tested in order to establish the agrochemical efficiency to the vineyard (*Chasselas dore*) in the intensive non-irrigated. The experimental fertilizer contained plant-derived protein hydrolysate, algae extract (*Ascomyces nodosum*) and secondary elements (Fe, Cu, Zn, Mn, B).

Based on the physico-chemical analyses, Eco Plus presented the following composition: organic nitrogen 3,3-3,5%, potassium 7.7-8,0%, fosfor organic 0,2 – 0,3%, 45-48% organic matter and 0.1 – 0.2% secondary elements (Fe, Cu, Zn, Mn, B).

The agrochemical trials assessed the evolution of production and the production yields, the process of photosynthetic assimilation, and the macronutrients content (N, P, K) present in vineyard leaves after fertilization.

The evolution of yields, photosynthesis activity and nutrient content in vineyard leaves, are presented in Tables 1, 2 and 3.

The productive efficiency resulted after the foliar application of Eco Plus to vineyard in intensive farm, on Hortic Antrosols is presented in Table 1.

Table 1

Productive efficiency (kg/ha) after foliar fertilization with Eco Plus (0.5% solution) on vineyard (*Chasselas dore*) in intensive farm, on Hortic Antrosols

Variant	Dose L/ha/ treatment	No. of treatments	Average prod. (kg/ha)	Yield	
				kg/ha	%
Control	-	-	9217	-	100
Eco Plus	2.5	3	11898	2681***	129.1

DL 5% - 934 kg/ha; DL 1% - 1357 kg/ha; DL 0.1% - 1881 kg/ha

It has been observed that organic components such as protein hydrolysates (Colla et al. 2017) and algae extracts (Ali et al. 2016) have demonstrated an important role of plant biostimulants by triggering physiological and molecular processes that stimulate growth and productivity (du Jardin 2015).

Table 2

Influence of foliar fertilization with Eco Plus biostimulant on photosynthesis in vineyard leaves (*Chasselas dore*) in intensive non-irrigated farm

Indicators / Experimental variant	Control	Eco Plus
Chlorophyll a (mg/g fresh substance)	1.388	1.889
Difference compared to the control (%)	100	136.10***
Chlorophyll b (mg/g fresh substance)	0.973	1.218
Difference compared to the control (%)	100	125.18**
Carotene (mg/g fresh substance)	0.589	0.724
Difference compared to the control (%)	100	122.92**
Total pigments (mg/g fresh substance)	2.950	3.831
Difference compared to the control (%)	100	129.86***

Chlorophyll a (mg/g)	Chlorophyll b (mg/g)	Carotene (mg/g)	Total pigments (mg/g)
DL5%: 0.242	DL5%: 0.136	DL5%: 0.087	DL5%: 0.431
DL1%: 0.371	DL1%: 0.205	DL1%: 0.124	DL1%: 0.581
DL0.1%: 0.484	DL0.1%: 0.272	DL0.1%: 0.198	DL0.1%: 0.874

Table 3.

Influence of foliar fertilization with Eco Plus biostimulant on mineral nutrition in vineyard leaves (*Chasselas dore*) in intensive non-irrigated farm (dry matter)

Indicators / Experimental variant	Control	Eco Plus
Total nitrogen (Nt %)	1.863	2.427
Difference compared to the control (%)	100	130.27**
Phosphorus (P2O5 %)	0.633	0.729
Difference compared to the control, (%)	100	115.17**
Potassium (K2O %)	0.851	1.142
Difference compared to the control, (%)	100	134.20***

Nt (%)	P2O5 (%)	K2O (%)
DL5% - 0.312%	DL5% - 0.073%	DL5% - 0.162%
DL1% - 0.488%	DL1% - 0.132%	DL1% - 0.203%
DL0.1% - 0.623%	DL0.1% - 0.196%	DL0.1% - 0.284%

The obtained results (Table 2) show a statistically distinctly significant and very significant yield compared to the control, for each assimilatory pigment and for the total content of assimilatory pigments after the three foliar treatments (0.5% solution). The results also showed that the increase of the photosynthesis was supported by the production yields. The photosynthetic efficiency promoted the increase of the production and the absorption of substances by the plant cell.

It can be noticed (Table 3), that the application of three foliar treatments with Eco Plus in the critical periods and of maximum necessity for plant nutrition at vineyard, has a positive influence on the macronutrients content (N, P, K) present in the foliar metabolism, respectively 15% for phosphorus, 30% nitrogen and 34% potassium. The experimental results showed statistically assured yields, distinctly significant compared to the control for nitrogen and phosphorus content and very significant for potassium contents (Table 3).

According to the results, the use of Eco Plus led to the increase of: mineral assimilation, chlorophyll content and cellular multiplication. Consequently, this led to: strong development of the foliar surface; better regulation of fruit differentiation; improved quality and quantity of production; increased resistance to stress and temperature variations; faster absorption and translocation in plant; increased resistance to diseases and pests.

The increases obtained by applying Eco Plus biostimulant compared to the control can be explained by the fact that the soil was well supplied and the biostimulating plant determined the plant to assimilate the necessary nutrients. (Rouphael et al. 2018, Baltazar et al. 2021).

It has been observed that the use of plant biostimulants does not have a negative impact on crops and the environment and can be used with confidence in agriculture. (Frioni et al. 2018; Taskos et al. 2019)

CONCLUSIONS

The use of the foliar Eco Plus biostimulant led to yield increases of 29.1% compared to the control. The three foliar treatments determined statistically significant yields compared to the control for the carotene content in leaves and very significant for the content of chlorophyll pigments, as well as for the total content of

assimilatory pigments, in the range of 25,2% and 36,1%. The foliar fertilization had a positive influence on the productivity and quality indicators, as well as on the photosynthesis activity. The foliar fertilization also stimulated the biosynthesis of assimilatory pigments, by decreasing the extent of the organogenesis stages

ACKNOWLEDGMENT

This research was conducted under the ADER Program, Contract ADER 144 /2019, Project "Identification, evaluation, testing, development and validation of methods of analysis for nutrients and contaminants from inputs used in organic farming".

REFERENCES

Ali, N., Farrell, A., Ramsubhag, A. and Jayaraman, J., 2016. The effect of *Ascophyllum nodosum* extract on the growth, yield and fruit quality of tomato grown under tropical conditions. *Journal of applied phycology*, 28(2), pp. 1353-1362.

Baltazar, M., Correia, S., Guinan, K.J., Sujeeth, N., Bragança, R. and Gonçalves, B., 2021. Recent Advances in the Molecular Effects of Biostimulants in Plants: An Overview. *Biomolecules*, 11(8), p. 1096.

Boselli, M.; Bahouaoui, M.A.; Lachhab, N.; Sanzani, S.M.; Ferrara, G.; Ippolito, A. 2019. Protein hydrolysates effects on grapevine (*Vitis vinifera* L., cv. Corvina) performance and water stress tolerance. *Sci. Hortic.*, 258, 108784.

Colla, G., Hoagland, L., Ruzzi, M., Cardarelli, M., Bonini, P., Canaguier, R. and Rouphael, Y., 2017. Biostimulant action of protein hydrolysates: unraveling their effects on plant physiology and microbiome. *Frontiers in plant science*, 8, p. 2202.

Du Jardin, P., 2015. Plant biostimulants: definition, concept, main categories and regulation. *Scientia Horticulturae*, 196, pp. 3-14.

Frioni, T., Sabbatini, P., Tombesi, S., Norrie, J., Poni, S., Gatti, M. and Palliotti, A. 2018. Effects of a biostimulant derived from the brown seaweed *Ascophyllum nodosum* on ripening dynamics and fruit quality of grapevines. *Scientia Horticulturae*, 232, pp. 97-106.

Nardi, S., Pizzeghello, D., Schiavon, M., & Ertani, A. 2016. Plant biostimulants: physiological responses induced by protein hydrolyzed-based products and humic substances in plant metabolism. *Scientia Agricola*, 73(1), 18-23.

Ricci, M., Tilbury, L., Daridon, B. and Sukalac, K., 2019. General principles to justify plant biostimulant claims. *Frontiers in plant science*, 10, p.494.

Rodrigues, M., Baptistella, J.L.C., Horz, D.C., Bortolato, L.M. and Mazzafera, P., 2020. Organic plant biostimulants and fruit quality—A review. *Agronomy*, 10(7), p. 988.

Rouphael Y., Colla G. 2018. Synergistic biostimulatory action: Designing the next generation of plant biostimulants for sustainable agriculture. *Frontiers in Plant Science*, 9, 1 – 17.

Taskos, D., Stamatiadis, S., Yvin, J.C. and Jamois, F., 2019. Effects of an *Ascophyllum nodosum* (L.) Le Jol. extract on grapevine yield and berry composition of a Merlot vineyard. *Scientia Horticulturae*, 250, pp. 27-32.

***Regulation (EU) 848/2018 of the European Parliament and of the Council of 30 May 2018 on organic production and labelling of organic products and repealing Council Regulation (EC) No 834/2007.

***https://ec.europa.eu/info/food-farming-fisheries/farming/organic-farming/organic-action-plan_en (accessed 6th September 2021).

***<https://www.madr.ro/docs/agricultura/agricultura-ecologica/2021/Dinamica-operatorilor-si-a-suprafetelor-agri-eco-update-28.06.2021.pdf> (accessed 6th September 2021).

***Council Regulation (EC) No 834/2007 of 28 June 2007 on organic production and labelling of organic products and repealing Regulation (EEC) No 2092/91.

***Commission Regulation (EC) No 889/2008 of 5 September 2008 laying down detailed rules for the implementation of Council Regulation (EC) No 834/2007 on organic production and labelling of organic products with regard to organic production, labelling and control.

***Brussels, 19.4.2021 COM(2021) 141 final/2: Communication from the Commission to the European Parliament, the Council, the European economic and social committee and the committee of the regions on an action plan for the development of organic production.