

REVIEW



on dissertation for obtaining the educational and scientific degree "Doctor" in: field of higher education 6.0 Agricultural Sciences and Veterinary Medicine, professional field 6.1 Crop science, scientific specialty Viticulture

Author of dissertation: Hristo Shterev Kozarov, full-time doctoral student at the Department of Viticulture and Fruit Growing at the Agricultural University, Plovdiv.

Dissertation Topic: Application of organic products and their impact on the growth and reproductive performance of Merlot variety grown under non-irrigated conditions.

Reviewer: Prof. Dsc. Slavcho Georgiev Pandeliev, Academician of the Bulgarian Academy of Sciences and Arts-Sofia, field of higher education 6.0 Agricultural Sciences and Veterinary Medicine, professional field 6.1 Crop science, scientific specialty Viticulture, appointed as a member of the Scientific Jury by Order RD 16-775/23.06.2026 of the Rector of the Agricultural university – Plovdiv.

1. Brief introduction of the candidate.

Master Hristo Shterev Kozarov was born on 29.03.1998 in the city of Sliven. In 2017, he graduated from secondary education and in 2021, he graduated from the Agricultural University in the city of Plovdiv. During this period, he also acquired the qualification of Master of Engineering in Technology of Wine and High Alcoholic Beverages at the University of Technology - Plovdiv. He is the winner of the Rector's Award of the AU - Plovdiv for the best student of the class, 2021. He has participated in the work of many university projects and events of student organizations in Bulgaria and abroad.

By Order of the Rector of the AU - Plovdiv RD - 26- 03/09.01.2023, he was enrolled as a full-time doctoral student in the Department of Viticulture and Fruit Growing. He has collected 194 credits from participation in educational, research and teaching activities. Of course, he received a maximum of 130 credits from research activities.

On 29.01.2026, a positive opinion of the department for discharge with the right to defense was adopted. He passed the exams in English, statistical data processing, teaching methodologies, scientific ethics and work with information sources with an excellent score. He has published 2 scientific articles. He has conducted 49.4 hours of exercises with full-time students of the Plant Protection and Agricultural Tourism specialty. He is fluent in English and Greek.

2. Relevance of the problem.

The problem with organic soil improvers is not new. Since ancient times, winegrowers have used various materials to improve the mechanical and chemical composition of the soil. Very nutrient-poor soils have been transformed into a rich nutrient medium for grapevine plants. However these processes have not been carried out systematically. In many cases, due to

ignorance and not fully understanding the nature of the various substances, negative results have been obtained. However this work deals with the use of substances with a known nature and chemical composition. One of these organic products is "Humat Rost". The doctoral student is correctly directed to use this substance as a means of enriching the soil with basic nutrients, improving its structure, mechanical and chemical composition, as well as preserving and utilizing soil moisture. I find the task thus set to be correctly formulated and relevant for modern viticulture developed under non-irrigated conditions. When developing the scheme of the experimental work, the scientific supervisors have also taken into account the specific climatic conditions of the northern slopes of the Rhodope Mountains.

3. Degree of knowledge of the state of the problem and creative interpretation of the literature review.

The doctoral student has reviewed 275 literary sources, including 19 in Cyrillic and 256 in Latin. They have been examined in a structured and critical manner in the following sequence: Climate - a natural feature that directly affects the grape formation and wine derivatives, soil fertility - a complex that determines the yield, quality of grapes and wine; Possibilities for maintaining a high level of soil biological activity in vineyard plantations; Phenology - external changes in plant organisms during the vegetation and dormancy periods; Physiology - factors determining the course of processes in grape cultivation; Root and foliar application of organic improvers; Characteristics of wine - changes in the physico-chemical and tasting potential. With this literature, the doctoral student has accumulated a lot of knowledge that has helped him distinguish the known from the unknown and correctly formulate the goals and objectives of the research.

4. Aim, objectives and research methods.

The purpose of the study is to establish the capabilities of the organic product "Humat Rost" and its impact on the growth and reproductive manifestations of the Merlot variety, grown under non-irrigated conditions. The use of this product to overcome stress conditions during the dormant and vegetative periods.

To achieve the goals, the following tasks have been set:

- Research on the climatic and soil conditions in the land of the town of Kuklen and their impact on the quantity and quality of grapes.
- Assessment of the metabolic activity of bacterial communities in the soil.
- Establishing differences in phenology, in the indicators characterizing the potential, actual fertility of the vines and the photosynthetic activity of the leaf mass.
- Determining the dynamics of ripening, yield and quality of grapes.
- Features of grape vinification and organoleptic evaluation of experimental wines.

The subject of the study was the Merlot variety, grafted on the Berlandieri x Riparia SO4 rootstock. The vines were planted in 2011 at the Experimental and Implementation Base of the Department of Viticulture and Fruit Growing at the University of Plovdiv, located near the village of Brestnik, Rhodope Municipality and the land of the Kuklen town. The vines were planted at distances of 3.0 m between rows and 1 m between vines in the row. The training

system is a double cordon, medium-stemmed and the bud loading of the vines was carried out by pruning 6 spurs with 2 buds per vine /12 buds/. The organic agent "Humat Rost" is a liquid concentrate with a rich dark brown color. The soil derivative has a pH of 8.23, organic substances are 80.66%. It is rich in phosphorus, potassium, nitrogen, magnesium, calcium, zinc, boron, humic acids. The composition of the leaf extract is presented, which contains nitrogen, phosphorus, potassium, magnesium, calcium with a pH of 7.5 - 9.5.

Seven variants are provided with foliar treatment in the phenophases - appearance of the first leaf, before flowering, after mass flowering, berry growth and veraison with the following doses:

V0 - control variant /without treatment/;

V1 - treatment with a dose of 200 ml/da;

V2 - with a dose of 300 ml/da;

V3 - with a dose of 400 ml/da;

V4 - treatment with a dose of 500 ml/da;

V5 - root treatment at the beginning of the vegetation with a dose of 15kg/da;

V6 - root treatment at the beginning of the vegetation with a dose of 25kg/da.

-Climatic characteristics of the area.

-Characteristics of the soil cover and chemical composition of the soil.

-Determination of the metabolic activity of microbial communities using the Biolog system /Eco Platetm/, which includes Amines - 2 pcs., Amino acids - 6, Carbohydrates - 10, Carboxylic acids - 7, Polymers - 4 pcs. and Phenolic compounds - 2 pcs.

-Phenological observations during the vegetation of the vines.

-Indicators characterizing the potential and actual fertility of the vines.

-Studies on the photosynthetic activity of the vines.

-Dynamics in the growth and maturation of shoots.

-Study on the yield and quality of grapes.

-17 physicochemical indicators were studied to evaluate the produced experimental wines.

-Quantitative determination of total protein substances.

-Isoelectric separation and determination of protein fractions by electrophoresis.

-Storage of annual canes with nitrogen, phosphorus, potassium, calcium, magnesium, starch and reducing sugars.

-Mathematical data processing and graph preparation were performed using the Excel program.

To identify statistically significant differences, analysis of variance with LSD, a multiple comparison test at a significance level of $P < 0.05$, was applied.

5. Visualization and presentation of the results.

The results of the research are presented in 29 tables and 54 figures. The large volume of experimental material is processed precisely and presented in a comprehensible manner in a

different, but accessible form for viewing. In all figures, the differences between the variants are clearly shown, which makes it easier for the reader.

6. Discussion of results and literature used.

During the three years study, the climatic conditions were not uniform, which made it possible to reveal the most important properties of the applied fertilizer. The humus reserves in the surface soil layer are high. The content of active Ca is very high, of nitrogen relatively low, of easily digestible phosphorus is low to medium, of potassium is very high. The soil reaction is slightly alkaline. Under such conditions, "Humat Rost" should appear. It has been established that under similar conditions of soil treatment and the absence of inhibiting factors, the development of soil microorganisms proceeds according to a model typical of ecoplates. The absorption of the groups of substrates is not uniform, therefore in individual repetitions of the ecoplates, high values of standard deviation and standard error are obtained. No proven statistical differences have been established in the duration of the phenophases, depending on the concentrations of the organic fertilizer used. It has been established that the growth vigor, the percentage of fruiting shoots and the size of the inflorescences are positively affected by organic fertilization. The results obtained for grape yield show that it is strongly affected by water deficit and very weakly influenced by fertilization with different doses from 200 ml to 500 ml of "Humat Rost". The content of assimilable nitrogen in grapes is significantly influenced by the specific conditions in individual years. For example, grapes in 2024 contained about twice as much assimilable nitrogen as compared to 2023 and 2025. It is imperative to clarify the reasons for this phenomenon, because the content of assimilable nitrogen in grapes has important technological significance. It has been established that at a content lower than 150 mg/dm³ of must, alcoholic fermentation slows down and the process may stop before the sugars in the grape juice are fully fermented. The data also show that grapes from vines treated with "Humat Rost" /V4, V5 and V6/ accumulate more anthocyanins, which also determines more intense, saturated and bright red color of the wines. The content of total and sugarfree extract is within normal limits from 25 to 30 g/dm³. No direct relationship was established between the concentration with which the vines were treated and the extract content of the wines produced. The content of total phenols was within normal limits between 2600 and 3000 mg/dm³. The wines produced were highly alcoholic, especially in the 2024 and 2025 harvests. The differences between the treated vines and the control variant were insignificant. The shade of color and the percentage of yellow-brown, red and blue-violet colors were within normal limits for young red wines. The wines from the grapes treated with 500ml in all three years of the study have the lowest shade of color. The lower shade is an indexation for a higher content of coloring matter and is a prerequisite for a more intense, more saturated and brighter red color of the wine.

The treatment with "Humat Rost" has a favorable effect on the organoleptic characteristics of the produced wines and their overall tasting assessment. Obviously, when using lower doses /200-300 ml/ there is no categorical impact on the organoleptic qualities of the wines.

The chlorophyll content in the area of the 10th leaf changes dynamically depending on the period of the study, the age of the leaf, the availability of nutrients and water. The main physiological processes of transpiration and photosynthesis of the leaves also occur in this direction. The linear growth of the shoots is not strongly affected by the applied fertilizer, but is directly dependent on the increased water deficit and high air temperatures. This is the main reason why the maturation of the shoots takes place with different durations from 85 days in 2023 to 35 days in 2024 and 34 days in 2025. No significant differences were found in the biochemical composition of the rods depending on the organic fertilizer "Humat Rost" used. The applied nutritional regimes within the three experimental years show a change in the amount of mature growth formed. The differences between the individual variants reach 1,780 kg.

7. Contributions of the thesis.

I accept the report on the contributions made with the dissertation work.

Scientific contributions

-For the first time, a scientific study is being conducted on the influence of the organic improver "Humat Rost" on the intensity of the physiological processes of transpiration and photosynthesis of leaves, the buds fertility, the yield and quality of grapes. The establishment of these complex dependencies represents new, up-to-date information in the world viticulture literature.

-The results obtained from the research are original, showing the relationship between the absorption of amino acids by microbial communities and the higher metabolic activity of vines treated with 15 kg/da "Humat Rost".

Scientific and applied contributions

-When treating the vines at the root with 15kg/da and 25kg/da "Humat Rost" an increase is achieved, but the amount of assimilable nitrogen, which favors the reproduction of yeast and the accumulation of microflora, leads to a smooth alcoholic fermentation of the grape juice.

-It has been established that the organoleptic characteristics of the resulting wines vary significantly depending on both the year of the harvest and the higher doses of treatment with "Humat Rost" used.

Applied contributions

- The applied regression analysis establishes a significant relationship between the amount of fertilizer, the period of feeding and the formation of the vegetative and reproductive organs of the vines.

-Treatment of the vines with "Humat Rost" stimulates the synthesis of coloring matter /anthocyanins/ in grapes and wine, as well as the intensity of the color and its brightness.

8. Critical comments and questions.

I do not make critical notes because I made them during the first review of the dissertation and they were taken into account in the final edit.

9. Published articles and citations.

Козаров Н. /2025/. Disruptions caused by abiotic factors affecting inflorescence development during the short Annual growth cycle of the grapevine. Journal of Mountain Agriculture on the Balkans /vol. 28, issue 5, pp. 576-601/. The article has been peer-reviewed and published in a reputable journal.

The presented abstract objectively reflects the structure and content of the dissertation.

CONCLUSION:

On the basis of the different research methods learned and applied by the PhD student, the correctly derived experiments, the made generalizations and conclusions, I consider that the presented dissertation meets the requirements of the FRAADB and the Regulations of the Agricultural university – Plovdiv for its application, which gives me the reason to evaluate it as **POSITIVE**.

I take the liberty of suggesting that the Honourable Scientific Jury also vote in the affirmative and award Hristo Shterev Kozarov the educational and scientific degree "Doctor" in the scientific specialty Viticulture.

20.07.2026 г.

Plovdiv

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