DOI: https://doi.org/10.53040/cga12.69 UDC: 634.8:632.4:632.95:631.95

TRENDS IN COMBATING VINE PHYTOPATHOGENS WITH THE APPLICATION OF EPIPHYTIC MICROORGANISMS

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Abstract. *The management of phytopathogens plays a decisive role in the sustainable development of agriculture, especially in viticulture. A particularly serious impact is recorded in the development of pathogens of the vine, especially of powdery mildew (Uncinula necator (Schwein) Burrill). In addition to measures promoted by modern agricultural biotechnologies the particular interest are the epiphytic microorganisms, which populate the leaf surface of plants, constituting complex heterogeneous habitats from various extended communities and numerous transients from various systematic entities. Through effective management and control of plant diseases, crop losses can be reduced and the quality and quantity of agricultural products can be improved. The article includes the analysis of the role of different biotic and abiotic factors, which change the relationships within these communities, as well as the adaptation processes of some entities of epiphytic and phytopathogenic microorganisms on the leaf surface of the vine.*

Keywords**: **biocontrol, ecological agriculture, epiphyte, phytopathogen agent

INTRODUCTION

The worsening of phytosanitary problems against the background of climate change lies in the modification of the epidemiological particularities of phytopathogenic agents and the increase in the prevalence of pests by manifesting various difficulties in carrying out technological processes for the production of agricultural crops [1]. The main problem of contemporary viticulture lies in increasing the receptivity of existing varieties to the attack of phytopathogens and specific harmful arthropods, which reduce grape harvests by 30-35% or cause total compromise of wine production.

From the great diversity of pathogens, a special role is played by diseases of cryptogamic etiology, and during the last decades there has been an increase in the pathogenicity of grapevine powdery mildew (*Uncinula necator (*Schwein*)* Burrill). Being widespread everywhere, powdery mildew in the conditions of climate change and the predominance of dry summers, and the pathogen attacks all the vegetative and generative organs of the plant. The manifestation of the disease is recorded by browning and falling of young bunches and abortion of flowers, as well as necrotization of berries at different stages of development, which requires the development and application of protection systems [2].

Ensuring the biological efficacy of the measures to reduce the impact of the pathogen requires increasing the efficiency of the protection systems oriented to the application of natural mechanisms by using the principles and techniques in the research of the efficiency of the action of the biological preparations proposed for the elaboration of the concept of synergism aimed at increasing the effectiveness of the elements proposed for conventional and organic farming systems [3].

Measures to reduce the impact of pathogens on agricultural crops are carried out by investigating the relationships between agricultural plants and microorganisms, which populate their leaf area, which largely determine the health status of agricultural crops. This is explained by the ubiquity of microscopic entities in natural environments (soil biota, aquatic environment and bioaerosols). Unlike the relationships of agricultural plants with pathogens in soil, water and air, epiphytic communities are not mechanisms for triggering pathogenic processes, and some interactions between the plant and epiphytic microorganisms are symbiotic, in which various synergistic mechanisms are manifested [4]. These include closely co-evolved partnerships between plants and mycorrhizal fungi or nitrogen-fixing bacteria. Most plant species are populated by numerous microorganisms, gathered within different microbiomes, consisting of microscopic entities with a lower degree of association and are influenced by the organisms that inhabit that part of the soil, and whose development is determined by the functionality of the plant rhizosphere. The manifestation of these microorganisms is recorded by the activity of rhizobacteria, which promote the growth of endophytic plants and fungi, which

participate in increasing resistance to pathogens and stressful conditions.

Plants have various microbial niches in the form of rhizosphere and phyllosphere, the representatives of which host representatives of various mutual, commensal and pathogenic microorganisms. They influence microbial communities in a species- and genotype-dependent manner, through secondary metabolites, exudates, and immune responses, all of which vary depending on developmental stage, plant compartment, and biotic interactions. Numerous species of bacteria, microscopic fungi, algae, viruses and protozoa have been isolated and identified on the surface of the photosphere [5], which represent a perfect model for analyzing the evolution of microflora and genetic exchange, as well as being a significant source of microorganisms, which diffuse to other spheres.

The contact of microorganisms with the surface of the leaves occurs with the help of raindrops and through the movement of air, as there are large fluxes of microorganisms in the atmosphere and at nearby surfaces. Landing on the leaf, microscopic organisms form aggregates of different sizes, separate individuals, as well as microcolonies. The peculiarities of the leaf surfaces lead to the formation of heterogeneous microhabitats covered by cuticle consisting of crystalline wax structures located on the protective tissues with a network of grooves, as well as the guard cell complexes around the stomata, which determine the permeability of the leaf apparatus. Penetrating into the essence of the mechanisms of the functioning of epiphytic communities and the relationships between the ecestora components and the phytopathogenic agents of plants represents a prospective way in reducing the impact of harmful organisms on agricultural crops.

RESEARCH MATERIALS AND METHODS

Various strains of bacteria, fungi and other pathogens of vine diseases were used for research, their control was carried out by testing the means of combating 4 randomized repetitions, in accordance with the general requirements of experiments of this kind [1, 6]. The protection programs against pathogenic organisms were developed based on the massive information recorded within the Laboratory of Phytopathology and Biotechnology.

The identification of biological products to combat biological agents will allow the improvement of integrated protection programs in conventional and organic farming systems. The situation in the field of plant protection and the environment was analyzed by applying the methodology of systemic approaches, as a tool of complexity management and as one of the essential paradigms of the future.

Laboratory and field experiment testing was carried out in 4 randomized trials, respectively, in accordance with the general requirements of experiments of this kind [7].

RESULTS AND DISCUSSIONS

Peculiarities of the pathogen of vine powdery mildew

Powdery mildew (*Uncinula necator)* is one of the most contagious diseases, affecting all vegetative and generative aerial organs and causing serious technological, economic and ecological problems. From the moment of initiation of the pathogenesis process, significant damage is recorded, which determines the need to apply effective means of prevention and control.

On the leaves there is a fine, spider's web-like mycelium, with a dusty appearance, which spreads forming whitish spots on both surfaces of the blade. Young shoots covered with whitish, and then brown, spots are stopped from growing. The bunches are browned and then dried. The berries attacked by mycelium crack, providing an optimal environment for the development of other fungal pathogens, and in rainy autumns they are covered with gray rot (*Botrytis cinerea*), which causes the quality of the grapes to depreciate.

The primary infection occurs through attacked shoots and cleistothecia (fig.1). The conidiospores, being spread by the wind, land on the foliar apparatus where they germinate and infect the vine by penetrating the haustors into the epidermal cells. The germination, infection and development of ascospores and conidiospores take 5-12 days, depending on the temperature, developing several cycles of infection throughout the growing season. It is worth mentioning that the development of the pathogen is favored by mild cloudy weather, moderate degree of lighting, optimal temperatures of 22-28°C with a range of 6-33°C, humid conditions necessary for sporulation [8].



Figure 1. Manifestation of *U.necator* symptoms on vine leaves and shoots

The phenomenon of climate change contributed to the early development of the pathogen. Weather conditions at the beginning of the season will affect management decisions to prevent outbreaks that occur later in the season (fig. 2). Maintaining wine plantations requires good air circulation, spray penetration, and exposure to sunlight.



Figure 2. Manifestation of *U. necator* symptoms on the generative organs of the vine

In order to achieve better spray coverage and prevent further development of the disease, the following practices are recommended: carrying out the first treatment at the stage of 2-3 true leaves; cutting shoots for aerating bushes; lifting threads to expose the bunches; removal of 2-3 leaves immediately above and below the bunches; adjustment of spray nozzles and direct airflow of spray machines; the use of large spray volumes and the highest rate of wetting agent; applying 2-3 sprays every seven to ten days to ensure maximum coverage of leaves and bunches [9].

Among the prevention measures, it is necessary to comply with the regulations for the correct application of maintenance works (cutting, tying, fertilizing, pinching, weed control), as well as a complex of prospective measures to reduce the impact of harmful organisms [1, 6].

Activities to expand the implementation of regenerative agriculture are based on the application of the agroecological principles of sustainable and resilient soil management, which are analogous to those of natural ecosystems and include: the permanent presence of living roots in the soil, permanent soil cover with plant debris, the great diversity of main and successive crops in rotation at the level of each individual lot and landshaft.

Synergistic phenomena – direction of increasing the effectiveness of the means of plant protection as natural sources of intensification of actions aimed at obtaining the integral effect of the joint application of several factors, which is greater than the simple sum of its parts and signifies "working together" and defines the increased effect that can be obtained by the simultaneous action of several physical elements, biological, economic, social.

The application of prophylactic measures remains as effective levers to reduce the impact of pathogens on the vine (selection of immune, resistant and tolerant varieties, choice of rootstock and stump shape, fertilizers, application of technological operations in green.

Development and application of technological programs for integrated protection of vines with the use of specific powdery mildew protection products, as well as the implementation of ecological agriculture.

Relationships between pathogens and epiphytic community elements on leaf surface

Starting from the needs of reducing biotic and abiotic stresses associated with leaf surface, epiphytic microorganisms are oriented towards the establishment of microhabitats capable of protecting host plants. The ability to navigate and stay in those niches in the leaf surface that are the richest in nutrients and water available is key to the epiphytic survival of both bacteria and fungi. To survive abiotic stress on the leaf surface, many microbes form biofilms composed of extracellular polysaccharides for cell aggregation. Biofilms are resistant to drying, antimicrobials and reactive oxygen species, which are important for plant protection. Biofilms can also be formed from yeasts and filamentous fungi, including a number of plant pathogens. The physical proximity of different microbial species in a stressful environment supports a high rate of horizontal gene transfer, especially for the exchange of plasmids and genes. The in-depth analysis of the relationships within epiphytic communities with the participation of bacteria is presented by [10], and filamentous microorganisms by [11].

Particularly important is the secretion of enzymes such as chitinases, esterases and lipases, which can release nutrients from the cuticle and, by degrading the cuticle components, increase its permeability to nutrients and the susceptibility of plants to infection. Epiphytic microorganisms produce siderophores containing iron chelates to increase iron absorption. The production of siderophores is also recorded in epiphytic yeasts, which have capacities to combat fungal pathogens.

In order to apply the capacities of epiphytic communities, it becomes necessary to highlight the similarities between epiphytic and phytopathogenic adaptations manifested in the production of plant hormones, the production of surfactants and the suppression of defenses, which cause the environment around pathogens to change, reducing penetration capacities and behaving as epiphytes during early leaf colonization [4]. Most leaf surface microorganisms occur in the same suitable micro habitats and can collaborate closely in biofilm formation with several species [12]. The interactions between them are not always oriented towards collaboration, becoming competitive in the conditions of limited nutritional resources. These interactions and their potential exploitation for the biological control of phytopathogens represent perspectives in order to ensure the high degree of plant health.

CONCLUSIONS

The extension of the phenomenon of climate change causes a series of changes in the phytosanitary status of agricultural crops, considerably increasing the impact of phytopathogens. There has been an increase in the virulence and aggressiveness of the pathogen *Uncinula necator* in the vineyard, which currently has a high degree of pathogenicity, and in hybrid varieties, which previously showed immunity to this pathogen.

The leaf surface represents an important habitat consisting of microorganisms with considerable potential for control of phytopathogens. Epiphytic communities are constituted by the host plant and under the influence of neighboring plants, soil, water, and air, which cause the formation of adaptations that manipulate the host plant and improve access to nutrients.

The application of the capacities of epiphytic communities requires the development and investigation of research models of interactions between microorganisms and the host plant, as well as the opening of potential pathways for the biological control of phytopathogens for the extrapolation of results aimed at preserving plant health.

Reducing the impact of the vine requires the application of the integral control system with the application of effective prophylactic and therapeutic means based on the achievements of modern agricultural biotechnologies.

Acknowledgements: The research was carried out within Subprogram 011103 "Development of environmentally harmless means to reduce the impact of pests of agricultural crops against the background of climate change", funded by the Ministry of Education and Research.

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