Phytosanitary Monitoring of Vineyards of Uzbekistan

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ANNOTATION.
This article describes the results of a study of phytosanitary control of microorganisms in grape varieties in vineyards located in the Syrdarya region of the Republic of Uzbekistan. In this study, microorganisms of the genera *Fusarium, Aspergillus, Alternaria alternate, Trichoderma, Rhizoctonia, Botrytis cinerea,*

Streptomyces were isolated from the grape varieties “Toifi”, “Charos”, “Qorachilgi”, “Khusayni”, “Mersedes” and “Rizamat ota’ and the degree of contamination was tested in experiments. While *Fusarium* was noted as the dominant micromycetes, fungi belonging to other genera were noted in the following order: *Aspergillus, Alternaria alternate , Trichoderma, Rhizoctonia, Botrytis cinerea*. It was also noted that 2 isolates were bacterial isolates.

KEYWORDS: *Plasmopora viticola, Botrytis cinerea, Fusarium, Aspergillus, Alternaria alternate, Trichoderma, Rhizoctonia, Botrytis cinerea, Streptomyces*

INTRODUCTION.

Viticulture is the world's most profitable industry, therefore, it requires special agrotechnological measures. It is known from scientific sources that grape growing around the world is characterized by its high profitability and special agrotechnological measures, including irrigation, microbiological and pest control. In general, vine agrotechnology is also important because of its low demand for water compared to many agricultural plants.

According to scientific sources, the total area of grape cultivation in the world is 7.3 million hectares. Global grape production in 2022 will be 80.1 million tons, three percent less than in 2021, while the People's Republic of China is the main exporter with 15.6 million tons of grapes, followed by Italy (8.1 million tons), it is noted that the country of France occupies the third place (6.2 million tons).

Although the damage caused by harmful insects and microorganisms in the process of growing grapes on a global scale is small compared to other plants, the death of grapes in a large number of fields due to the influence of observed pests and microbiological organisms, this process is of significant importance impact on subsequent stages of grape storage and production of grape products; improvement of measures to combat diseases and pests.

Fig. 1. Annual world grape production
(*-The yield of 2022 is added to the expected yield results of 2023)
[https://www.statista.com/statistics/237600/world-grape-production-in-2007-by-region/]

Microorganisms that are common in the processes of microbiological damage to vines and the risk of their spread on the organs of the plant are shown in Fig.1 [Armijo G, 2016]. It can be seen from Figure 1 that, according to the generally accepted theory, the main microbiological damage of vines is caused by microorganisms such as *Plasmopora viticola*, *Botrytis cinerea*, *Xylella fastidiosa*, *Agrobactrium vitis*. So, in the microbiological damage of vine, a complex of microorganisms can be found, including bacteria, fungi, oomycetes and viruses.

We know that, like all plants, the vine plant is one of the plants rich in microbiota [Turabekova D.B., et al., 2023]. When grapevines are infected with fungi, pathogens disrupt the integrity of the vascular tissue in the grape stems, which disrupts the movement of water and nutrients throughout the plant. Scientists studied the impact of the disease on the yield and quality of wine products from infected vineyards. The Riesling Italico and Cabernet Sauvignon varieties were used as objects of the experiment. Using a spectrophotometer, the physicochemical parameters of wine products made from infected and uninfected grapes were analyzed. To check the statistical significance of the results, the Tukey test was used ($p \leq 0.05$). In this case, 51.29 g / l of Cabernet Sauvignon and 23.72 g / l of Italian Riesling were found in a sample taken from infected grapes. It was found that the degree of damage to plants has a significant impact on their quality [Janás, M. et all., 2025].

During the analysis of scientific sources, it was noted that the microbiota found in the vine plant is diverse. Including *Bacillus megaterium*, *Bacillus niacini*, *Bacillus cereus*, *Staphylococcus warneri*, *Staphylococcus epidermidis*, *Micrococcus luteusni*, *Sphingomonas sp.*, *Pseudomonas kilonensis*, *Microbacterium arborescens*, *Kloeckera apiculata*, *Lactobacillus*, *Acinetobacter*, *Arthrobacter*, *Pantoea dispersa*, *Pantoea aglomeranlar*, *Pseudomonas corrugata*, *Serratia rubidaea*, *Enterobacteriaceae*, *Enterobacter gergoviae*, *Klebsiella oxytoca* [Subden et al., 2003; Renouf et al., 2005; Nisiotou et al., 2011; Kántor et al., 2015, Wei R-t et all., 2022; Turabekova D.B., et al., 2022].

It was also found that the tissues of the vine are dominated by bacteria belonging to the genera *Pseudomonas*, *Sphingomonas*, *Frigoribacterium*, *Curtobacterium*, *Bacillus*, *Enterobacter*, *Acinetobacter*, Erwinia, *Citrobacter*, *Pantoea*, and *Methylobacterium* [Bokulich et al., 2014; Perazzolli et al., 2014].

As a result of the analysis of the mycological composition of vines worldwide, it was found that fungi belonging to the families of *Ascomycetes* and *Basidiomycetes* are dominant, while fungi belonging to the families of *Zygomycota* and *Chytridiomycota* are found in very small quantities [Pinto et al., 2014; Gao et al., 2019, Liu et al., 2021; Wei et al., 2022].

In addition, mycelial fungi belonging to the genera *Aspergillus*, *Alternaria*, *Penicillium*, *Cladosporium*, *Lewia*, *Davidiella*, *Erysiphe* and *Botrytis* are common, and from yeast fungi Aureobasidium pullulans and *Aureobasidium pullulans* ҳамда *Hanseniaspora*, *Issatchenkia*, *Pichia*, *Candida*, *Rhodotorula*, *Lachancea*, *Metschnikowia*, *Cryptococcus*, *Filobasidiella*, *Sporobolomyces* and yeasts belonging to the genus *Torulaspora* have been found to occur depending on the samples taken from the plant (soil, stem, leaf, flowers, branches, leaves, fruit, fruit products, etc.) [Bokulich et al., 2014; Pinto et al., 2014; Wang et al., 2015; Filippis et al., 2017; Liu et al., 2021].

In neighboring countries, including Afghanistan, in 2017, in Nari and Kunar regions, it was reported that the fungus Fusarium equiseti is causing great damage to vineyards, and for the first time, signs of brown rot were detected in the leaves and roots of grapes grown in Afghanistan [Nasir Ahmed Rajput et al., 2020].

MATERIALS AND METHODS.

Plant sources used. Grape varieties "Toifi", "Charos", "Korachilgi", "Husaini", "Mercedes" and "Rizamat", vineyards infected with *Fusarium fungus*. Experimental work was carried out on a 6.0-hectare plot of the “Alijon Kuvonchbek Bogi” farm, which specializes in horticulture and viticulture, and on a 10-hectare plot of “Erkhulota orzusi” LLC.

Used microbiological sources. Microorganisms of the *Fusarium, Aspergillus, Alternaria alternate, Trichoderma, Rhizoctonia, Botrytis cinerea, Streptomyces* series were isolated and tested in experiments as the main objects of the research.

Nutrient media used in the cultivation of mycological objects.

Czapek-Dox medium (g/l): distilled water – 1.0; sucrose - 30.0; $NaNO_3$ – 2.0; K_2HPO_4 – 1,0; $MgSO_4$ – 0,5; KCl - 0,5; $FeSO_4$ – 0,01; nutrient agar – 15.0; pH 7.3±0.2. Sterilization is carried out for 20 minutes at 121 ºC under a pressure of 1 atmosphere.

Sabouraud agar (g/l): distilled water – 1.0; glucose - 40.0; peptone - 10.0; nutrient agar - 18.0; pH 6.5±1. Sterilization is carried out for 20 minutes at 121 ºC under a pressure of 0.5 atmospheres.

Mendels nutrient medium (g/l): KH_2PO_4 - 2.0; $(NH_4)_2SO_4$ - 1.4; $FeSO_4 *7H_2O$ - 0.0027; $MnSO_4 *7H_2O$ - 0.0016; $ZnSO_4 *7H_2O$ - 0.0014; $CoCl_2 *6H_2O$ - 0.0037; $MgSO_4*7H_2O$ - 0.6; $CaCl_2 *2H_2O$ - 0.4; peptone - 0.75; urea 0.3 and glucose 30. pH - 4.8. Sterilization is carried out at 121 ºC for 20 minutes.

Potato glucose agar nutrient medium (g/l): distilled water - 1.0; starch or sucrose - 10.0-30.0; $NaNO_3$ – 9,2; KCl - 0,5; $MgSO_4 *7H_2O$ - 0.5; KH_2PO_4 – 1,0; $FeSO_4$ – 0,01; nutrient agar 20.0-30.0; pH 4.9-5.2. Sterilization is carried out for 20 minutes at 121 ºC under a pressure of 1 atmosphere.

RESULTS AND DISCUSSION.

Phytosanitary control of the vineyards of Syrdarya region and scientific research conducted in 2020-2023 for the purpose of studying endophyte and rhizosphere microorganisms related to grape varieties. In particular, when the vineyards of 13 farms specializing in horticulture and viticulture in the Syrdarya region were subjected to phytosanitary control as of June 2020, the areas infected with signs of fungal diseases amounted to 22.33% (Table 1). In the results of phytosanitary analyzes and phenological observation, it was found that there were no signs of fungal disease in 31 seven-year-old and 21 fifteen-year-old Muscat vines. Charos, Red Husaini, Chilgi, Black chillaki varieties were also noted as the least affected grape varieties. As the vine varieties with the most frequent signs of fungal disease, Husaini (5-year-46.43%, seven-year-36.36), Toifi (two-year-37.50%, four-year-26.47%, five-year-41 .94%, fifteen-year-32.41%) varieties, Rizamat (two-year-32.14%, five-year-42.11%, seven-year-32.20%) varieties were noted.

TABLE 1

The dynamics of occurrence of signs of fungal diseases in the cross section of vines

№	The variety of vines studied	Vine age, year	The number of	The part of the vine where the fungal disease was observed	Number of infected vines	The number of diseased vines compared to the

			studied vines, pcs	stem	leaf		total number of vines, %
1	Charos	10	38	+	-	2	5,26
2	Muscat	7	31	-	-	-	0,00
3	Muscat	15	27	-	-	-	0,00
4	Husaini	5	28	++	+++	13	46,43
5	Husaini	7	22	+	++	8	36,36
6	Red Husaini	7	74	-	++	5	6,76
7	Red Husaini	12	43	-	+	3	6,98
8	Toifi	2	72	++	+++	27	37,50
9	Toifi	4	68	++	+++	18	26,47
10	Toifi	5	62	++	+++	26	41,94
11	Toifi	15	28	-	+	9	32,14
12	Rizamat	2	62	++	+++	37	59,68
13	Rizamat	5	57	+	+++	24	42,11
14	Rizamat	7	59	-	++	19	32,20
15	Rizamat	10	47	-	+	4	8,51
16	Kelin barmok	10	78	-	+	7	8,97
17	Chilgi	6	42	-	+	3	7,14
18	Chilgi	15	33	-	+	2	6,06
	Total:		927			207	22,33

In the course of observations, during the study of 47 vines of the 10-year-old Rizamat variety, only 4 vines, 8.51%, were found to have signs of fungal disease. It was concluded that fungal diseases have started to develop in this vineyard.

The next phase of the research was continued on the varieties of vines with the most signs of fungal disease. On the basis of mycological analysis, mycological samples were taken and analyzed from the stem and leaf of the vine as one object and from the soil of the growing area of the vine. The obtained results are shown in Table 2. During the mycological analysis, it was noted that the number of isolates isolated from the leaves and stems of the vine was 22.04% less than the number of isolates found in the soil. During the research, it was observed that 28.26% of isolates were found in the leaves and stems of Husaini vine, and 71.74% in the soil.

TABLE 2

Proportion of mycological isolates isolated from vine leaf, stem and soil

Grape varieties	Number of isolates isolated from the plant	Number of isolates isolated from vineyard soil	The occurrence of isolates in the plant, %	The occurrence of isolates in the soil, %
Husaini	13	33	28,26	71,74
Toifi	18	13	58,07	41,93
Rizamat	15	13	53,58	46,42

Total:	46	59	43,81	56,19

The analysis showed that in the vine variety Toifi, 16.14% more was found in the plant leaves and stems than in the soil. The same situation was observed in the Rizamat variety of the vine, and it was found that the isolates were 7.16% more frequent in the leaves and stems than in the soil. Research was conducted on 46 isolates isolated from plant organs from a total of 105 isolates isolated during the research. Fungal isolates isolated from diseased plant organs were cultured on different nutrient media and classified according to their growth colony morphology, mycelium, macro- and microconidia under the microscope. Based on mycological analysis, it was noted that 46 isolates belonged to 7 categories (Table 3).

TABLE 3

Composition of microbiota depending on the varieties of the vine in the case of Syrdarya region

№	Series of isolates	Meeting in vine varieties			Total number of isolates	Relative to the total number of isolates, %
		Husaini	Toifi	Rizamat		
1	*Fusarium*	4	8	5	17	36,96
2	*Aspergillus*	2	3	2	7	15,22
3	*Alternaria*	3	-	2	5	10,87
4	*Trichoderma*	2	2	3	7	15,22
5	*Rhizoctonia*	2	1	3	6	13,04
6	*Botrytis cinerea*	-	2	-	2	4,35
7	*Streptomyces*	-	2	-	2	4,35
Жами:		13	18	15	46	100,00

Fusarium (37%) was recorded as the dominant micromycete, while fungi belonging to other genera were found in the following order: *Aspergillus* (15.2%), *Alternaria alternate* (10.9%), *Trichoderma* (15.2%), *Rhizoctonia* (13 .0%), *Botrytis cinerea* (4.3%). It was also noted that 2 isolates were bacterial isolates (4.3%). Bacterial isolates were isolated from the genus Toyfi and were found to belong to the genus *Streptomyces* (№. 7).

Also, the proportion of isolates belonging to the *Fusarium* genus was determined in the section of species: although it was determined that they belonged to the species *F.oxysporium, F.solani, F.culmorum, Fusarium poae,* it was not possible to determine the type of 11 isolates based on morphocultural and some biochemical tests (Table 4).

TABLE 4

Mycological composition of the vine depending on the varieties in the case of the Syrdarya region

№	Types of fungi	Meeting in vine varieties			Total	Relative to common fungi, %
		Husaini	Toifi	Rizamat		
1	*F.oxysporium*	1	1	–	2	11,76

2	*F.solani*	–	1	1	2	11,76
3	*F.culmorum*	–	1	–	1	5,88
4	*Fusarium poae*	–	–	1	1	5,88
5	*Fusarium spp.*	2	6	3	11	64,71
Жами:		3	9	5	17	100,00

Based on the conducted research, it was noted that in the vineyards of Syrdarya region, species belonging to the genus *Fusarium* were more common than other micromycetes. As a continuation of further research, the goal was to develop biological control tools against these mycological objects.

CONCLUSION.

As a result of the conducted research, the following conclusions were presented:

1. In the results of phytosanitary analyzes and phenological observation, it was found that 31 seven-year-old and 21 fifteen-year-old Muscat vines did not show signs of fungal disease. Charos, Red Husaini, Chilgi, Vinniy, Black chillaki varieties were also noted as the least affected grape varieties. As the vine varieties with the most frequent signs of fungal disease, Husaini (5-year-46.43%, seven-year-36.36), Toifi (two-year-37.50%, four-year-26.47%, five-year-41.94%, fifteen-year-32.41%) varieties, Rizamat (two-year-32.14%, five-year-42.11%, seven-year-32.20%) varieties were noted.

2. During mycological analysis of the vine varieties with the most fungal disease symptoms, it was noted that the number of isolates isolated from the leaves and stems of the vine was 22.04% less than the number of isolates found in the soil. During the research, it was observed that 28.26% of isolates were found in the leaves and stems of Husaini vine, and 71.74% in the soil.

3. On the basis of mycological analysis, it was noted that 46 isolates belonged to 7 categories. *Fusarium* (37%) was recorded as the dominant micromycete, while fungi belonging to other genera were found in the following order: *Aspergillus* (15.2%), *Alternaria alternate* (10.9%), *Trichoderma* (15.2%), *Rhizoctonia* (13 .0%), *Botrytis cinerea* (4.3%). It was also noted that 2 isolates were bacterial isolates (4.3%).

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