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STUDY OF GREEN MOLD INFECTION ON POMEGRANATE FRUITS IN THE WESTERN PART OF AZERBAIJAN

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Abstract

The article presents information on green mold (*Penicillium expansum* Lk. et Thom.) infection on pomegranate bushes in the western part of Azerbaijan. The activities which was carried out in the period 2021-2023 and were focused on the phytosanitary condition of pomegranate orchards with large industrial importance located in the Ganja-Kazakh geographical zone (western part of the country) showed that fungal diseases were widespread in the region, including green mold (*Penicillium expansum* Lk. et Thom.) infection. In some years, the green fruit mold (*Penicillium expansum* Lk. et Thom.) has reduced the productivity of pomegranate bushes by 50-100%. The local cultivars of pomegranate were the object of the study and the results showed that all varieties were highly susceptible to the pathogen. The article presents also results of laboratory experiments about effectiveness of some fungicides - 0.4% Selphate (37.5% copper oxychloride + 16% zineb), 0.05% Azoxifene (20% azoxystrobin + 12.5% difenoconazole), 0.05% Conazole (25% difenoconazole) and 0.3% P-oxide (50% copper oxychloride) against green mold infection. The assessment of biological effectiveness was based on reduction of growth of green mold (*Penicillium expansum* Lk. et Thom.) on the treated variants relative to the control. Among the tested fungicides, the product Selphate showed the highest biological effectiveness.

Keywords: pomegranate, pathogen, *Penicillium* Link., protection, fungicides

INTRODUCTION

Pomegranate (*Punica* L.) is a deciduous fruit shrub or tree, reaching a height of 5-6 m (Figure 1/a). The branches are thin, prickly, the leaves are glossy, the flowers are funnel-shaped orange-red with a diameter of 2.5 cm or more (Figure 1/b, c). Pomegranate bushes produce spherical fruits, which are, large berries with a leathery pericarp and a retained calyx. The color of the peel varied from orange-yellow to brown-red (Fig. 2, a, b) (Bullard & Mortensen, 1970, Akhmedov, 2010, Doroshenko et al., 2019, Huseynova, 2020, Namazov, 2021). One of the reasons for low yields of pomegranate bushes in the western part of Azerbaijan is loss of production from fungal diseases, the most harmful of which is the green mold of fruits (*Penicillium expansum* Lk. et Thom.). Under favorable conditions, the green mold (*Penicillium expansum* Lk. et Thom.) reduces the yield of pomegranate fruit by 50-100% (Huseynova, 2020, 2021, 2022). During 2021-2023, it was established that the disease affected pomegranate fruits during the growing season and storage. Often the fruits became infected while still on the bush in the garden. Collecting and storage of infected fruits, often results in the intensive mold development and ultimately rotting of all collected fruits.

When the disease occurs, pomegranate fruits rot like watery rot. Green mycelium, belonging to the phytopathogenic fungus *Penicillium expansum* Lk, forms inside the rotten fruits. Gradually the fruit softens and its internal content rot (Figure 3). Affected fruits

are completely unsuitable for consumption and processing. The observation of the current study implied that disease more often occurs in fruits that were damaged mechanically during fruit harvesting or by pests during the growing season.

Fig. 1. General view of a pomegranate bush (a), leaves (b), flowers (c)

Fig. 2. Pomegranate cultivars: Pink gyulosha (left), Krmyzy kabuh (right)

Fig. 3. Green mold on pomegranate fruits

Hyphomycetales are the most extensive morphologically and ecologically diverse order of imperfect fungi (*Anamorphic fungi*). *Hyphomycetes* are included in numerous ecological groups of fungi: soil, xylophilous (living on wood), plant parasites, carnivorous (catching microscopic animals and feeding on them), aquatic, mycophilic (living as parasites on other fungi), entomophilous (parasitic on insects), etc. (Gorlenko et al., 1980, Becker, 1988, Adamzhanova, 2009). One of the most common representatives of hyphomycetes is the genus *Penicillium* Link. Among hyphomycetes fungi of the genus *Penicillium* Link. rightfully occupy the first place in distribution. Their natural reservoir is soil, and they, being mostly cosmopolitan, are confined more to the soils of northern latitudes (Garibova & Lekomtseva, 2005). They are most often found in the form of mold deposits, consisting mainly of conidiophores with conidia, on a variety of substrates, mainly of plant origin (Popkova, 1989, Koval et al., 2016).

The main goal of the research work is to study the prevalence, intensity of development and harmfulness of the green mold on the pomegranate bushes (*Penicillium expansum* Lk. et Thom.) in the western part of Azerbaijan. To investigate the spread and development of the disease in the western part of Azerbaijan the current study focused on: 1) to identify the main environmental factors contributing to the spread of green pomegranate mold (*Penicillium expansum* Lk. et Thom.) in the Ganja-Kazakh geographical zone of Azerbaijan (western part of the country); 2) to clarify some biological features of the causative agent of green mold fungus - *Penicillium expansum* Lk. et Thom. trough field and laboratory experiments; 3) to develop measures to combat green mold of pomegranate fruits (*Penicillium expansum* Lk. et Thom.).

MATERIALS AND METHODS

In 2021-2023, research work was carried out to study green mold on pomegranate fruits (*Penicillium expansum* Lk. et Thom.). Biological material was collected from industrial pomegranate gardens located in the western part of the republic (in the regions of Shamkir, Goranboy and Kazakh). Mycological and phytopathological studies were carried out at the Central Phytosanitary Laboratory of the Azerbaijan Institute of Food Safety. The harmfulness of green mold of pomegranate fruits (*Penicillium expansum* Lk. et Thom.) was studied on the susceptible cultivars "Krmyzy kabukh" and "Pink Gulosha".

The field experiments were consisted of five variants with three replications. Observations and notes about the prevalence and development of green mold of pomegranate fruits (*Penicillium expansum* Lk. et Thom.) were made systematically throughout the entire growing season following standard procedures in phytopathology (Chumakov et al., 1974, Khokhryakov, 1976, Dospehov, 1985, Khokhryakov et al., 2003). Pomegranate orchards were treated with fungicides during the growing season: at the first signs of the disease appearance, and subsequently at intervals of 7-10 days. Prevalence (P, %) was determined after counting diseased and healthy plants using the formula:

$$P=100n/N,$$

where n is the number of diseased plants in the sample; N is the total number of plants examined.

Disease development (R, %) was determined using the following formula:

$$R=(100\sum(ab))/Nk$$

where a is the number of diseased plants; b - corresponding score of their defeat, N is the total number of plants under observation (sick and healthy), k is the highest score on the accounting scale.

The dynamic of development was determined by the weather conditions of the growing season.

Biological efficiency (BE), expressed as a percentage, was calculated using the formula (Guliyev & Huseynova, 2022):

$$BE=(M_k-M_o)/M_kx100,$$

where M_k is the indicator of disease development in the control (protective measures were not carried out); M_o-indicator of disease development in the experiment (with protective measures).

RESULTS AND DISCUSSION

In the conditions of the western part of Azerbaijan, the green mold infection on pomegranate fruits (*Penicillium expansum* Lk. et Thom.) causes damage both in the field and in storage conditions. The distribution and intensity of development of green mold of pomegranate fruits (*Penicillium expansum* Lk. et Thom.) in the western part of Azerbaijan (2021-2023) were determined (Table 1).

As can be seen from the data that the green mold infection on pomegranate fruits (*Penicillium expansum* Lk. et Thom.) occured in all surveyed areas with significant severity. Thus, in 2021, the distribution of green mold (*Penicillium expansum* Lk. et Thom.) on the cultivar "Krmyzy Kabukh" by region ranged from 60.2 to 67.9%, in 2022 from 63.7 to 68.8%, in 2023 from 65.6 to 69.1%. Of all the pomegranate cultivars, "Krmyzy kabukh" and "Pink Gulosha" are especially susceptible to green mold, but as can be seen from the table, the Krmyzy kabukh cultivar was the most severely affected by this disease in all western regions where the research was carried out. The fruit rot begins with a small watery light brown spot, which, as it grows, becomes slightly depressed and becomes folded. When lightly pressed the skin bursts and droplets of moisture are released. The spot is covered first with white mycelium, and then with sporulation of the fungus in the form of a mass of greenish-bluish

or gray-green pads, and therefore the rot is often called bluish moldy. The rotten pulp has an unpleasant sour taste and musty smell, which is easily transferred to the healthy part of the fruit.

The occurrence of the disease is directly influenced by factors such as agrometeorological conditions, especially temperature. An analysis of the agrometeorological conditions of 2021-2023 showed that in these years' temperatures in the range of 12-35^{0}C were very favorable for the spread and development of the disease. Also during the years of research, frequent rainfall, cool and wet weather were among the factors stimulating the disease. Thus, an analysis of agrometeorological conditions shows that during 2021-2023, humidity was high and temperature was optimal. Agrometeorological conditions were favorable for the spread of the disease on a large scale. In the conditions of the western part of Azerbaijan, the disease causes damage both in the field and in storage conditions. The disease develops especially strongly when harvesting rules are violated. The affected areas of tissue soften, become watery and are easily squeezed.

Table 1. Distribution and intensity of development of green mold of pomegranate fruits (*Penicillium expansum* Lk. et Thom.) in the western part of Azerbaijan (2021-2023)

Western regions of Azerbaijan	Pomegranate cultivars	Year					
		2021		2022		2023	
		P, %	R, %	P, %	R, %	P, %	R, %
Shamkir	*Krmyzy Kabukh*	67.9	33.3	68.8	33.7	69.1	33.5
	Pink Gyulosha	44.1	21.0	45.0	21.7	45.6	21.9
Goranboy	*Krmyzy Kabukh*	65.5	31.9	66.6	31.2	66.9	31.8
	Pink Gyulosha	33.9	18.8	34.9	19.0	36.1	19.5
Kazakh	*Krmyzy Kabukh*	60.2	29.8	63.7	30.0	65.6	30.4
	Pink Gyulosha	30.9	17.8	31.7	17.7	31.9	18.1

Legend: P - prevalence, %; R - intensity of development, %

The details of the structure of conidiophores (smooth or spiny, colorless or colored), the sizes of their parts can be different in different species, as well as the shape, structure of the shell and the size of mature conidia. The results of a laboratory study of the fungus *Penicillium expansum* Lk et Thom are presented. Analysis of laboratory studies indicated that *Penicillium expansum* Lk. et Thom. colonies on Czapek agar were fast-growing, velvety or flaky, less often tufted, with white mycelium. Sporulation was moderately abundant, with blue-green to yellow-green color. Exudate was usually absent, or less often present, colorless or brownish in color. The water-soluble pigment released into the medium and the reverse of the colonies were cream, orange-brown to dark brown in color, or there was no water-soluble pigment, the reverse was whitish. There colonies have strong smell, somewhat fruity. Colonies on agar with malt extract were flat, velvety to the point of forming cores, often without aerial mycelium, often with a more pronounced grayish tint of sporulation than on Czapek's medium. Conidiophores were three-tiered, sometimes 2-4 layered, smooth-walled, sometimes rough in the lower part, 200-500 μm long and 3-4 μm thick. Conidia were elliptical to almost spherical, smooth-walled, 3.0-3.5x2.5-3.0 μm, in long irregular chains.

The results from the current study showed that the chemical measures were successful in the fight against the green mold infection on pomegranate fruits (*Penicillium expansum* Lk. et Thom.). The fungicides which were tested and their effectiveness are presented (Table 2).

Table 2. Effect of fungicides on the spread and development of green mold of pomegranate fruits (*Penicillium expansum* Lk. et Thom.) in the western part of Azerbaijan (2021-2023)

Fungicides/concentration/active ingredients	Biological or technical efficiency, %								
	Year								
	2021			2022			2023		
	P	R	BE	P	R	BE	P	R	BE
Azoxifene 0.05% (20% azoxystrobin + 12.5% difenoconazole)	21.0	13.7	66.0	21.1	13.6	66.1	19.0	13.0	68.0
0.05% Conazole (25% difenoconazole)	18.0	11.1	72.2	17.7	11.0	73.0	17.1	10.5	74.1
0.4% Sulfate (37.5% copper oxychloride + 16% zineb)	16.8	10.0	75.0	16.0	9.5	76.3	15.1	8.0	80.3
0.3% P-oxyride (50% copper oxychloride)	19.1	12.7	68.2	19.5	12.2	70.0	18.8	11.9	71.0
Control (without chemical treatment)	78.7	39.9	0	79.0	40.1	0	79.9	40.5	0

Legend: P - distribution, %; R- development, %; BE - biological effectiveness, %

As can be seen from the table, among the fungicides used against penicillium of pomegranate bushes (*Penicillium expansum* Lk. et Thom.) the product Sulfate showed the highest result for all three years. Among other fungicides, Conazol also showed good results. The preparations Azoxifene and P-oxiride showed were slightly less effective and were unable completely to protect the fruit crop from the destructive effects of mold (*Penicillium expansum* Lk. et Thom.).

CONCLUSION

Thus, green fruit mold (*Penicillium expansum* Lk. et Thom.) is a very harmful disease of pomegranate bushes in the western part of Azerbaijan. It is found in almost all western regions (Goranboy, Shamkir, Kazakh) of crop cultivation, affecting ripening and ripe pomegranate fruits. The disease often begins with softening and the formation of green mold, the rotting process spreads to the remaining parts of the fruit. On the rotten tissue of the pomegranate fruit, abundant green mycelium, hyphae, conidia, and conidiophores of the parasitic fungus *Penicillium expansum* Lk. et Thom. are formed. The pomegranate fruits affected by green mold (*Penicillium expansum* Lk. et Thom.) fall off.

An effective means of combating green mold of pomegranate fruits (*Penicillium expansum* Lk. et Thom.) was spraying with fungicides - 0.4% Sulfate, 0.3% Azohifen, 0.05% Conazol and 0.05% P -oxyride Among the tested fungicides of pomegranate fruits (*Penicillium expansum* Lk. et Thom.) in the 2021-2023 research years, the product Sulfate showed the greatest biological effectiveness.

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