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Pathogenicity Characteristics of *Alternaria Alternata* and Fusarium Solani which Cause Diseases in Pistachio

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Abstract: In the conditions of Uzbekistan, a total of 10 types of fungal diseases have been identified in pistachios, among which the dominant species are alternarium, fusarium and septorium. Due to the fact that alternarium and fusarium of pistachio are being studied for the first time, studies were conducted on their pathogenicity.

Keywords: pistachio, alternaria alternata, fusarium solani, pathogenicity, fungi.

The pathogenicity characteristics of *Fusarium solani* (Mart.), *Alternaria alternata* (Fr.) Keissler Sacc., which were identified during the study of diseases caused by fungi in pistachio, were studied in order to determine the disease-causing nature of the fungus. To do this, experiments were based on the Koch method, that is, isolating a pure culture of the pathogen from a diseased plant and artificially infecting pistachios with this fungus and isolating a pure culture of this fungus on a diseased plant.

Pathogenicity of *Alternaria alternata* to pistachio. To determine the pathogenicity of *A.alternata* to pistachio, its strains isolated from diseased samples were used. To conduct the experiments, seven strains of *A.alternata* (7, 9, 15, 20, 24, 27, 31) were grown in beer wort at a temperature of 24-26⁰ C for 7 days (pic.1).

According to literary sources, the titer of its spores in the culture fluid of $2 \cdot 10^3$ colony forming unit/ml is enough to damage plants with this fungus [1]. This titer of spores in the culture liquid was used in experiments to test the pathogenicity of *A. alternata*.

In order to artificially infect container-grown pistachio seedlings, their leaves were sprayed with a spore suspension of the above titer, and the seedlings were wrapped in cellophane to preserve moisture. They were placed in a greenhouse with light for 16 hours and a temperature of 24-26⁰ C. 10 seedlings were taken for each experimental option (pic.2).

After 3-4 days, it was observed that small dark-gray spots appeared on pistachio leaves. After 7-10 days, the formation of fungal conidia was noted on the surface of the spots. The development of the disease in these seedlings was monitored until the end of the growing season.





Pic.1. The strains of the A.Alternata

Pic.2. selected seedlings of pistachio for experiment

As a result of the experiments, strain 24 showed the strongest pathogenicity, in which 80% of the seedlings were infected, and it was found that the stem growth of the infected seedlings was behind by 32.5% compared to healthy seedlings (Table 1). The lowest rate was observed in the 7th strain, in which up to 20% of the seedlings were affected by the disease, and the stem growth was delayed by 8.1%. It was observed that 30-60% of the rest of the seedlings were affected by this disease, while the stem growth was 11.2-27.9% behind that of healthy seedlings. When leaf samples taken from diseased seedlings were planted in a humidity chamber in a Petri dish, and a pure culture of the pathogen was isolated from them, it was revealed that this fungus species was an *A.alternata*.

Table 1. Pathogenicity of *A. alternata* fungal strains to pistachio seedlings

Laboratory researches, 2019-2020 yy.

No	Fungal strains of	Number of artificially	Diseased seedlings		Seedlings stem	The backwardness of the plant
	A.alternata	infected seedlings, pcs	pcs	%	length, cm	compared to the healthy one, %
1.	7	10	2	20	18,1	8,1
2.	9	10	4	40	16,5	16,2
3.	15	10	6	60	14,2	27,9
4.	20	10	6	60	14,5	26,4
5.	24	10	8	80	13,3	32,5
6.	27	10	5	50	15,5	21,3
7.	31	10	3	30	17,5	11,2
8.	Control (undamaged)	10	-	-	19,7	-

 $EKF_{0.5}=1,3$

To study the effect of *A. alternata* fungal culture fluid titer on pistachio leaf disease, strain 24, which showed strong pathogenicity, was selected for further experiments. For this, the 24th strain of A. alternata was grown in beer wort for 7 days at a temperature of $24-26^{\circ}$ C. Pistachio seedlings were artificially infected with its spore suspension of several titers $(1\cdot10^{3}, 2\cdot10^{3}, 1\cdot10^{4}, 2\cdot10^{4}, 3\cdot10^{4})$. Conducting this experiment and taking into account the disease was carried out in the above-mentioned way.

As a result of the experiment, it became known that as the spore titer in the culture liquid of A.alternata fungus increases, the incidence of pistachio leaves also increases. When the titer of the spore suspension was $1 \cdot 10^3$ colony forming unit/ml, the damage of seedling leaves was 6.3-28.5%, when the titer of the spore suspension was $3 \cdot 10^4$ colony forming unit/ml, this indicator was 38 was 7-80.2% (Table 2). When pistachio leaves were artificially infected with the A.alternata fungus, it was noted that the young leaves on the tips of the leaves on the lower side of the stem were relatively more affected by the disease. Damage on the 2nd leaves at the top of seedlings was 6.3-38.7%, on the 3rd leaves it was 10.9-42.1%, and on the 4th leaves it was equal to 28.5-80.2%.

Table 2. Effects of *A. alternata* **strain 24 spore suspension on pistachio seedlings** Laboratory researches, 2019-2020 yy.

	A.alternata spore titer (cfu/ml)	Number of artificially	Damage to seedling leaves, %			
№		Number of artificially infected seedlings, pcs	2 leaves	3 leaves	4 leaves	
		infected seedings, pcs	from the top	from the top	from the top	
1.	1.10^{3}	10	6,3	10,9	28,5	
2.	2.10^{3}	10	11,4	22,3	37,0	
3.	1.10^{4}	10	25,8	34,5	58,1	
4.	2.104	10	34,2	40,6	75,7	
5.	3.10^{4}	10	38,7	42,1	80,2	

 $EKF_{0.5} = 1,4$

Based on the data obtained as a result of the experiment, it can be concluded that the disease of alternariosis in pistachios is caused by the fungus *A.alternata*. The higher the number of spores of this fungus in the environment, the stronger the damage to the leaves by the pathogen. Older pistachio leaves are more severely damaged by this fungus.

Fusarium solani ning xandon pistaga nisbatan patogenligi. To determine the pathogenicity, 10 seedlings were selected for each option for artificial infection with strains of the F.solani fungus. To determine the pathogenicity of the strains, the stem of the selected seedlings was cut in the shape of a "T" using a sterilized scalpel [2]. The skin of the seedlings in the cut area was slightly bent and artificially infected by using a syringe from culture fluids grown in beer wort with F. solani strains for 7 days (pic.3).

The infected part of the seedlings was tightly wrapped with scotch tape. From the seventh day, the progress of the disease in infected seedlings was monitored. Symptoms characteristic of Fusarium disease were observed on the leaves, branches and stems of seedlings. Phenological control of artificially infected seedlings showed that the disease does not show any external symptoms in the initial period.





Pic.3. a) F. solani fungal strain and the culture fluids grown in beer wort; b) appearance of the fusarium after artificially infecting

Internal symptoms of the disease: the mycelium of the fungus, moving through the conductive tubes in the wood tissues, destroyed the tissue cells in the places where they entered. As a result, it was noted that the dead cells turned brown under the influence of poisonous substances, that is, toxins of the fungus.

The internal symptoms of the disease in the seedlings appeared 20 days after the experiment. Symptoms of the disease are clearly manifested in these seedlings, and when their stem is cut crosswise, it was found that the primary and secondary wood cambium tissues darken even to the core.

Among the strains obtained for the experiment, strain 16 showed the highest pathogenicity, and it was found that 70% of seedlings were infected in this variant (Table 3).

Table 3. Pathogenicity of *F. solani* fungal strains to pistachio seedlings

Laboratory researches, 2019-2020 yy.

No	Fungal strains of	Number of	Diseased		Seedlings	The
	F.solani	artificially	seedlings		stem length,	backwardness of
		infected seedlings,	dona	%	cm	the plant
		pcs				compared to the
						healthy one, %
1.	3	10	3	30	37,8	18,4
2.	5	10	5	50	33,2	28,3
3.	8	10	2	20	39,8	14,0
4.	13	10	1	10	42,5	8,2
5.	16	10	7	70	29,8	35,6
6.	19	10	6	60	31,8	31,3
7.	Control	10	-	-	46,3	-
	(undamaged)					

 $EKF_{0.5} = 1,4$

The least manifestation of the disease was recorded in 13 strains, and in this variant, up to 10% of seedlings were infected with the disease. Damage of seedlings up to 20-60% was observed in the remaining strains. Due to the disease, it was found that the development of the stem of the seedlings lags behind by 8.2-35.6% compared to the control.

Phenological observations made on artificially infected seedlings showed that these seedlings are weak compared to the control and lag behind in growth and development.

Based on the results of the experiment on the pathogenicity of *F. solani* fungus, it was concluded that it is this type of fungus that causes fusariosis in pistachio sprouts and seedlings.

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