Fungal diseases of tomatoes and their inhibition by local strains of microorganisms

Nigora Bekmukhamedova[1*], *Mukhiddin* Mamiyev[2], *Anvar* Tadjiyev[3], *Tulqin* Quziyev[2], *Shohrukh* Norimov[3] and *Dildora* Rustamova[3]

[1]Institute of Microbiology of the Academy of Sciences of the Republic of Uzbekistan, 100140 Tashkent, Uzbekistan.

[2]Tashkent State Agrarian University, 100140 Tashkent, Uzbekistan.

[3]Urganch State University, 220100 Urganch, Uzbekistan.

Abstract. Research has been carried out to determine the antagonistic properties of local strains of actinomycetes and microscopic fungi to the main pathogens of plant diseases - *Alternaria alternata, Verticillium dahliae.* It was shown that among the studied strains of microorganisms, the cultures of *Streptomyces sp.31* and *Stachybotrys sp.13* showed high antagonistic activity to the studied phytopathogens. At the same time, the zone of suppression of phytopathogens reached up to 36-40 mm in diameter.

1 Introduction

Tomato (*Solanum lycopersicum L. var. lycopersiciun*) is one of the most widespread vegetable crops in the world, comprising about 90 genera and at least 2,500 species. This is explained by its high yield, variety of uses, high biological value and high taste of the fruit [3,14].

Tomato is the most widespread vegetable crop not only in our country, but throughout the world. It is grown in all countries of the world, and in countries with warm climates it occupies a leading place among vegetable crops. World production of tomato fruits is 112-115 million tons, and the acreage occupied by them is 4.3-4.4 million hectares.

In Russia and Vietnam, tomatoes are grown both in open and protected ground. However, annual tomato yields are unstable due to damage caused by late blight, fusarium, verticillium, cladosporiosis, tobacco mosaic virus, as well as non-infectious blossom end rot. With the advent of resistant tomato varieties and hybrids, vegetable growers are less likely to encounter fungal diseases on tomatoes.

Fungal diseases of tomatoes:

Alternaria blight (dry leaf spot, macrosporiosis) and rot of tomato fruits. Pathogen: *Alternaria alternataf. sp. lycopersici*.

Dry spot is a common disease in the greenhouse and in the soil. Mass manifestations of the disease are observed at the beginning of fruit formation, reaching a peak towards the

* Corresponding author: mamiev65@mail.ru

end of the growing season. Fruit yield losses from leaf spot in some cases can reach 70%. Damaged fruits are unsuitable for sale. In addition to their spoiled appearance, they accumulate mycotoxins that are toxic to humans, which are preserved during processing and can end up in tomato juice and paste.

Alternaria causes the formation of round spots up to 1.5 cm in size on the leaves. The first signs appear on the leaves at the beginning of flowering. Later, spots appear on the petioles and stems. They have an elongated shape and at high humidity a black fungal coating is noticeable on them.

All types of Alternaria blight are diseases of tomato fruits that can cause rot during ripening and storage. Round depressed spots appear on the tomatoes, which are then covered with a black coating. Alternaria spots often develop in places of microcracks, tears, wounds, sunburn, frostbite, damage by insects, damage by other fungi and bacteria.

In open ground tomato crops, out of the entire diversity of the phytopathocomplex, the dominant harmful species has been identified - Alternaria, which annually damages the fruit harvest by 20-40%, which helps reduce its profitability [2, 12, 15].

The authors conducted research to study effective and environmentally friendly measures of protection against the most important tomato diseases in protected soil. Monitoring of the spread of root rot and accompanying micromycetes on tomato and cucumber plants was carried out in the conditions of production greenhouses of the Moskovsky agricultural complex, as well as an assessment of the influence of various antagonist bacteria on the intensity of development of root rot [8].

The most common fungal diseases on tomatoes in Uzbekistan are brown, white, dry leaf spot, black leg, fusarium and verticillium wilt, root rot, and gray rot. Late blight, powdery mildew, various rots, anthracnose, and stem cancer are less common. As a result of studies conducted on tomato plants and fruits in the Fergana Valley, the authors identified 20 species from 19 genera, 11 families, 10 orders and 4 divisions, and the main diseases are seedling lesions, Fusarium wilt, brown spot, Alternaria spot, fruit rot during storage. Late blight and powdery mildew were observed less frequently [7, 9, 13, 17].

Verticillium wilt. Pathogens: imperfect fungi: *Verticillium albo-atrum and Verticillium dahlia.*

Both species cause verticillium wilt of tomatoes - the same symptoms - one-sided yellowing starting from the leaves of the lower tier, curling of the leaf blades and their general wilting.

When infected with Verticillium wilt, chlorotic areas first appear on aging leaves. These signs are similar to a lack of nitrogen and magnesium in the diet. During the day the plant withers, but during the night it restores turgor and in the morning it seems quite healthy. Later, unilateral or general chlorosis on the leaves leads to their curling and drying out, which spreads from the lower leaves to the top. As a result, the plant dies completely.

If you make a cut in the stem, you can notice darkening of the blood vessels, and other infections and pests also settle on diseased bushes. When these pathogens enter the plant, they cause blockage of blood vessels and, as a result, wilting of the entire bush. These diseases can become chronic and cause a decrease in overall yield.

Tomato is the main crop of Uzbekistan. It is cultivated here on an area of more than 60 thousand hectares, and the gross harvest of its fruits is about 1.6 million tons. About 70% of the harvest is used for processing, 15-20% on the domestic market and 10-15% is exported to other countries. Fresh fruits and their processed products are in great demand on the foreign market, and therefore tomato production in the republic is constantly growing.

Tomatoes in Uzbekistan are grown using seedlings and without seedlings. The seedling method allows you to get the harvest earlier and extend the growing season of plants. Thanks to the seedling method, early tomato varieties in Uzbekistan can be grown as a repeat crop after harvesting grains and early vegetables [4, 23-34].

Chemical control of pathogens is labor-intensive, costs a lot of money, is not always effective enough, and in many cases is harmful to human health and the environment. Therefore, priority remains for the creation of highly productive tomato hybrids that are resistant to biotic and abiotic stresses [1, 10, 19, 21, 22].

To date, more than 8,000 antibiotics have been isolated from microorganisms, and about 4,000 have been obtained from other organisms: lichens, plants and animals. The most important and broad class of antibiotic producers are microorganisms [16, 20].

Antibiotics produced by microorganisms belong to different groups of compounds according to their chemical structure. The amount of antibiotics produced directly depends on the conditions for cultivating microorganisms, such as temperature, pH, medium composition, etc. Media for cultivating microbial cultures are selected individually for each producer. They must contain sources of carbon, nitrogen, various micro- and macroelements [11].

The Microbiology Laboratory of Shanxi Normal University conducted research to determine the antimicrobial activity of actinomycete isolates isolated on synthetic Gause agar against pathogenic strains of bacteria of the genera *Escherichia coli* E1, *Staphylococcus aureus* S4, *Serratia sp.* H1, *Dickeya duckantii sp*. and microscopic fungi of the genera *Penicillium sp.* P1, *Candida tropicalis* C1, *Verticillium dahlia* V2, *Fusarium oxysporum* FO1, *F.solani* FS3, *F.sulphureum* FS1, *F.vasinfectum* FOV1 and *Didymella bryoniae* DB1. It has been established that newly isolated strains of actinomycetes actively suppress the growth of the studied phytopathogens of bacteria and microscopic fungi [18].

Thus, the goal of our study was to isolate local strains of microorganisms from the tomato rhizosphere that have antagonistic activity against pathogens of fungal diseases of tomatoes.

2 Materials and Methods.

The object of the study are actinomycetes and microscopic fungi isolated from the rhizosphere of tomatoes in the Tashkent region, Kibray region.

To grow micromycetes, we used Czapek nutrient agar (g/l: sucrose - 30; NaNO3 - 2,0; KH2PO4 - 1,0; KCl - 0,5; MgSO4 - 0,5 FeSO4 - 0,01; tap water - 1. 0; pH = 6.5 agar-agar - 20.0). For actinomycetes, starch-ammonia agar (CAA, g/l) was used: NaNO3 - 1,0; MgSO4 - 1,0; K2HPO4 - 1,0; CaCO3 - 3,0, NaCl - 1,0, starch - 10.0; tap water - 1.0; pH = 6.8-7.0; agar-agar - 20.0) [5].

The antagonistic properties of actinomycetes were studied using the agar block method [6]. The test cultures were collection strains of micromycetes - phytopathogens - pathogens of diseases in plants: *Alternaria alternata, Verticillium dahliae.*

3 Results and Discussion

40 isolates were isolated from the rhizosphere of tomatoes in the Tashkent region, Kibray region. Of these, 18 strains of actinomycetes and 15 strains of microscopic fungi were isolated into pure culture.

The results showed that out of 15 strains of microscopic fungi tested, 9 strains showed different inhibitory effects on pathogens of tomato diseases. Table 1 shows the zone of absence of growth of phytopathogens (D=mm) *Verticillium dahlia* and *Alternaria alternate* antagonists - micromycetes. Thus, all 9 studied strains, except *Acrimonium sp.*1, inhibited the growth of the pathogen *Alternaria alternata*, the causative agent of tomato Alternaria blight. At the same time, the zone of no growth of the pathogen reached from 14 to 30 mm

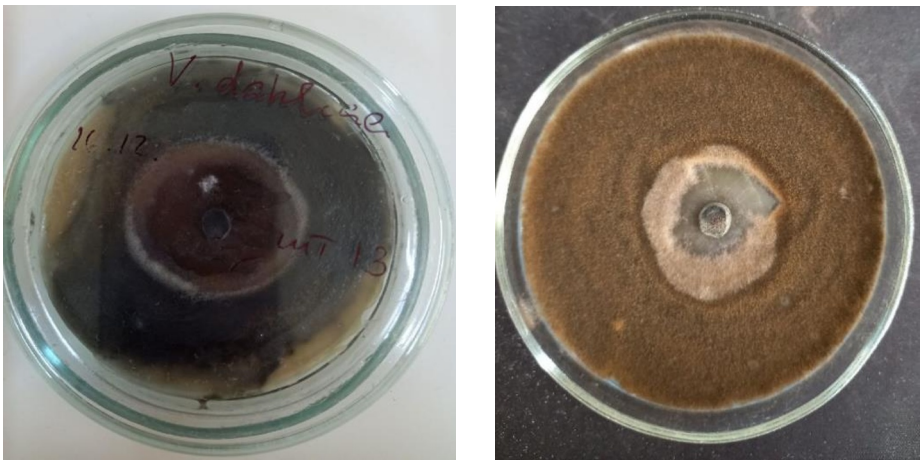
in diameter. The most active were strains of the genus *Aspergillus*, which inhibited pathogens up to 30 mm.

Table 1. Antagonistic properties of newly isolated strains of microscopic fungi to pathogens of tomato diseases

№	Micromycetes	Zone of no growth of phytopathogen, D=mm	
		Verticillium dahliae	*Alternaria alternata*
1.	*Acrimonium sp.1*	20	-
2.	*Penicillium sp.2*	24	20
3.	*Clodospora sp.3*	20	14
4.	*Aspergillus sp.5*	-	30
5.	*Penicillium sp.6*	-	14
6.	*Aspergillus sp.7*	-	27
7.	*Aspergillus sp.9*	-	16
8.	*Aspergillus sp.10*	-	22
9.	*Stachybotrys sp.13*	42	30

*Acrimonium sp.*1, *Penicillium sp*. 2 and *Clodospora sp*.3 inhibited the growth of *Verticillium dahliae*, the causative agent of verticillium wilt in tomatoes, while the pathogen-free zone reached up to 24 mm.

A high antagonistic activity of the micromycete *Stachybotrys sp.*13 was established, which inhibited the growth of pathogens from 30 to 42 mm in diameter (Figure 1).



1 2

Figure 1. Inhibition of phytopathogens by *Stachybotrys sp.*13
Verticillium dahliae (1), *Alternaria alternata* (2)

Studies were carried out to determine the antagonistic ability of 18 strains of actinomycetes newly isolated in pure culture. The results showed that out of 18 actinomycete strains tested, 10 strains suppressed the growth of *Verticillium dahliae* (Figure 2). In this case, the zone of no growth of the fungus ranged from 16 to 40 mm.

It has been established that the high antagonistic activity of *Streptomyces sp*. 31, reaching significant values, which inhibited the growth of the causative agent of Verticella wilt up to 40 mm.

Strains 22 and 68 do not inhibit the pathogenic micromycete *Alternaria alternata*, but other tested streptomycetes inhibited the growth of the causative agent of *Alternaria* from 16 to 24 mm.

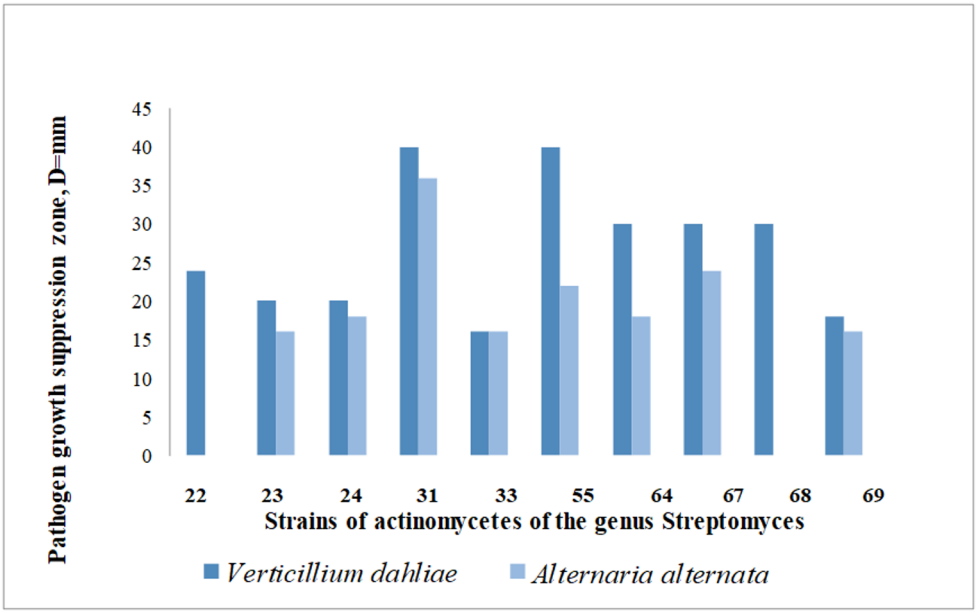
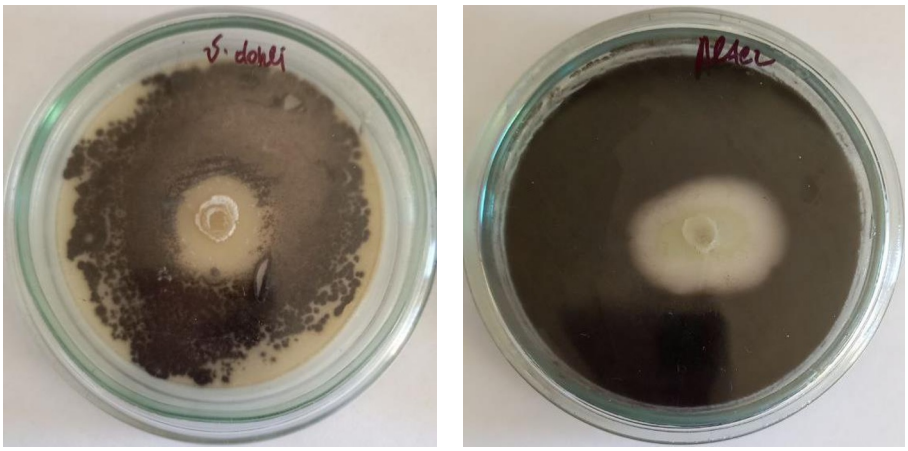


Figure 2. Inhibition of phytopathogens by active antagonists - actinomycetes



1 2

Figure 3. Inhibition of the growth of tomato pathogens *Verticillium dahliae* (1), *Alternaria alternata* (2)

4 Conclusion

Thus, it has been established that proven active strains of microorganisms *Stachybotrys sp*.13 and *Streptomyces sp*.31 with high antagonistic ability are practically important for recommending their theoretical and practical use in agriculture and the development of new biological products for the destruction of fungal diseases of tomatoes, such as - *Verticillium wilt* and *Alternaria blight*.

Reference

1. Avdeev A.Yu. Selection and testing of tomato varieties for individual and collective farms in the Volga region. *Abstract for the candidate of agricultural sciences* (2006).
2. Vinogradova O.V., Surkova T.A., Zayats V.G. The influence of antagonist bacteria on the development of symptoms of major tomato diseases in protected soil. *Bulletin of RUDN University, series agronomy and livestock breeding*. **3**, 30-35 (2008).
3. Gavrish S.F. Tomatoes. *Veche*, 160 (2005).
4. Demidov E.S. Methods of selection and seed production of vegetable crops for the creation of environmentally friendly technologies in the southern regions of the CIS countries. *Abstract for the candidate for Doctor of Agricultural Sciences* (2000).
5. Egorov N.S. Guide to practical classes in microbiology. *Publishing house. Moscow State University*. 205 (1995).
6. Egorov N.S. Fundamentals of the doctrine of antibiotics. *Publishing house. Moscow State University.*104 (2003).
7. Madikhanova Sh.N., Mirzaitova M.K., Dekhkonova D.K. Features of tomato, *"Science and Education" Scientific Journal,* **2**, 138-142 (2021).
8. Malashkina E.A., Kotova V.V., Pakina E.N., Surkova T.A., Slobodyanyuk K.V. The influence of bacteria and micromycetes - antagonists on the development of root rot in tomato and cucumber plants in production greenhouses of the Moskovsky agricultural complex. *Bulletin of RUDN University. series Agronomy and animal husbandry*. **1.** 56-64 (2009).
9. Omonova N.M. The influence of different air temperatures on the growth of pathogenic fungi in tomatoes. *Life Sciences and Agriculture*. **2-3.** 94-97 (2020)
10. Palastrova O.A. The effectiveness of fungicides in the fight against tomato diseases. *Bulletin of the Kurgan State Agricultural Academy*, **2.** 41-43 (2018).
11. Popova E.D. Selection and optimization of nutrient media for the cultivation of actinomycetes - antibiotic producers. *International scientific research journal.* **3** (34). (2015).
12. Problems of scientific support for vegetable growing in the South of Russia. *Collection of scientific papers*. 135-139 (2009).
13. Rasulov U.Sh., Uktamzhonova Kh.A., Mamurova Sh.N. Fungal diseases of tomatoes. *Science and Education" Scientific Journal.* **2**, 41-46 (2021).
14. Siddikova N.K., Mirzaitova M.K., Abdullaeva G.D. Fungal diseases of conifers. *Bulletin of Science*, **1(12)**, 257-259 (2019).
15. Khromova L.M., Sarbasheva A.I., Shidova L.H., Khromova D.A. Alternaria blight is a dangerous disease of tomatoes. *Bulletin of the Altai State Agrarian University*. **12 (158)**, 26-31 (2017)
16. Schmid R. Visual biotechnology and genetic engineering. *Binomial. Knowledge Laboratory*. 40 (2014).
17. Yunusov Z.R., Soloviev A.A., Mikhailenko S.N., Komakhin R.A., Zhuchenko A.A. The influence of transgenes on meiotic recombination in higher eukaryotes, using the example of tomato plants. *Agricultural biology*. **3**, 52-59 (2009).
18. Dong-sheng Wang, Hai-ke Ren, Erihemu, Zhi-yi Zheng. Isolation, identification and antagonistic activity evaluation of actinomycetes in barks of nine trees, *Arch Biol Sci*. **69 (2)**, 345-351 (2017). https://doi.org/10.2298/ABS160429109W

19. Jones J.B., Jones J.P., Stall R.E. and Zitter T.A. Compendium of Tomato Diseases. *APS Press*. 73 (1991).
20. Khidirov S.Yu., Gulmurodov R., Mamiev M.S., Yuldosheva D. Propagation of fungi in the soils: A case study of scientific experimental fields of forestry of Tashkent region. *IOP Conference Series: Earth and Environmental Science,* (2020).
21. Safiuddin, Sheila Shahab, Mohd. Mazid, Dania Ahmed. Comparative study of Fusarium oxysporum f sp. lycopersici and Meloidogyne incognita race-2 on plant growth parameters of tomato. *Agricultural Sciences,* **3**, (2012).
22. Tavga Sulaiman Rashid, Kamaruzaman Sijam, Hayman Kakakhan Awla, Halimi Mohd Saud, Jugah Kadir. Pathogenicity Assay and Molecular Identification of Fungi and Bacteria Associated with Diseases of Tomato in Malaysia. *American Journal of Plant Sciences*. **7**, 949-957 (2016).
23. Abdiev, Z., Khurramov, U., Karimov, J., Zokirov, K., Omonov, M., Jumanov, L., & Ruzmetov, G. The influence of nutrition and various materials on tomato cultivation in hydroponics. *E3S Web of Conferences*, Vol. **452**, 01012 (2023). https://doi.org/10.1051/e3sconf/202345201012
24. Abduvasikov, A., Khurramova, M., Akmal, H., Nodira, P., Kenjabaev, J., Anarkulov, D., & Kurbonalijon, Z. (2024). The concept of production resources in agricultural sector and their classification in the case of Uzbekistan. *Caspian Journal of Environmental Sciences*, ***22*(2)**, 477-488.
25. Atkhamova, S., Eshmatov, F., Fayzullaeva, N., Shomaksudova, K., Uzakova, S., & Zokirov, K. Determination of bioactive compounds of pomegranate peel to incorporate as a functional food ingredient in food production. *BIO Web of Conferences*, Vol. **95**, 01038 (2024). https://doi.org/10.1051/bioconf/20249501038
26. Buriev, K., & Zokirov, K. Seasonal and daily blooming rhythm of cucumber (Cucumis sativus) belonging to Cucurbitaceae family. *E3S Web of Conferences,* Vol. **452**, 01015 (2023). https://doi.org/10.1051/e3sconf/202345201015
27. Buriev, K., & Zokirov, K. Seasonal and daily blooming rhythm of watermelon (Citrullus lanatus) belonging to Cucurbitaceae family. *E3S Web of Conferences,* Vol. **452**, 01014 (2023). https://doi.org/10.1051/e3sconf/202345201014
28. Dissanayake, K., Rifky, M., Farmonov, J., Boynazarova, Y., Meyliyev, A., Zokirov, K., & Samadiy, M. Impact of incorporating inorganic additives in processed meat production. *E3S Web of Conferences*, Vol. **510**, 01037 (2024). https://doi.org/10.1051/e3sconf/202451001037
29. Dissanayake, K., Rifky, M., Hunupolagama, D., Harris, J. M., Zokirov, K., Azizova, K., & Samadiy, M. Inorganic additives in meat production and processing. *E3S Web of Conferences*, Vol. **510**, 01028 (2024). https://doi.org/10.1051/e3sconf/202451001028
30. Mirsaidov, M., Nimchik, A., Khodjiyev, O., Jesfar, M., Zokirov, K., Shamatov, S., & Kambarov, A. Analysing the chemical standards of the Fergana Mekhmash wastewater treatment plant and environmental processing. *E3S Web of Conferences*, Vol. **498**, 02016 (2024). https://doi.org/10.1051/e3sconf/202449802016
31. Nendissa, D. R., Alimgozhaevich, I. K., Sapaev, I. B., Karimbaevna, T. M., Bakhtiyarovna, S. Z., Abdullah, D., & Sharifovna, A. G. (2023). Sustainable livestock grazing in Kazakhstan practices, challenges, and environmental considerations. *Caspian Journal of Environmental Sciences*, ***21*(4),** 977-988.
32. Rifky, M., Abdusalomova, D., Dissanayake, K., Zokirov, K., Harris, J. M., Jesfar, M., & Samadiy, M. Seaweed as a functional ingredient and emulsifier in dairy processing.

BIO Web of Conferences, Vol. **108**, 18001 (2024). https://doi.org/10.1051/bioconf/202410818001

33. Zokirov, K. Effect of drip irrigation regimes and mineral fertilizer rates on watermelon yield in mulched fields. *E3S Web of Conferences*, Vol. **389**, 03067 (2023). https://doi.org/10.1051/e3sconf/202338903067
34. Zokirov, K. Irrigation regimes for drip irrigation of watermelon in mulched and non-mulched fields. *E3S Web of Conferences*, Vol. **389**, 03068 (2023). https://doi.org/10.1051/e3sconf/202338903068