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RESEARCH ARTICLE

Antagonistic Features of Species Belonging to The Genus Trichoderma Spread in Azerbaijan

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ABSTRACT

In research were studyed antagonistic properties of strains of 10 species from the genus Trichoderma isolated from different(soil, plant and water) biotopes of Azerbaijan, and taken to the pure culture against to the phytopathogenic fungi. It became clear that in all the studied strains in relation to cultures such as Alternaria alternata MB-1056, Aspergillus flavus MB-1217, Bipolaris nodulosa MB -2011, Fusarium oxysporum MB-7012, F.solani MB-7034 and *Penicillium cyclopium* MB-11276 which used as a test shows such a feature. It manifests itself at the highest level in the strains T.asperellum G-3, as well as T.citrinoviride G-26 and T.koningi G-43. Although there is no correlation between the quantitative indicator of antagonistic activity of fungi from the genus Trichoderma and their cultural morphological type, the strains of T.asperellum G-3 and T.coningi G-43 synthesizes metabolites with strong fungicidal activity, which are considered promising for practical use.

KEYWORDS:
Trichoderm genus, phytapathogenic fungi, antagonism, antibiotic activities

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INTRODUCTION

As is known, fungi from the genus Trichoderma are in the antagonistic attitude with phytopathogens, and these features of theirs are used for a long time in in biological struggle[6, 16-18]. From the research conducted in this regard became clear that, the biological activity of fungi from the genus Trichoderma depends on the type of soil, hydrothermal regime, and the composition of mycobiota specific to that biotope, and use the strains isolated from that places in concrate condition are more effective[4, 9, 13-14]. Thus, the biotic and abiotic factors of each region are characterized by different quantitative and qualitative indicators and this is reflected in the fungi biota that inhabits in that region[15]. As a result, are formed strains characterized by specific physiological and biochemical properties peculiar for that region.

Although there is literature about of the distribution of species of fungi from the genus of Trichoderma in Azerbaijan[2], among those no researches on their use in biological control. Taking this into consideration, assessment of anatagonal characteristics of fungi species belonging to the genus Trichoderma was set as a goal in the presented work, and for this purpose was used from monospore strains of noted genus.

MATERIALS AND METHODS

The researches were carried out in the territory of the Greater Caucasus, Kur-Araz lowlands of the Republic of Azerbaijan in 2014-2020 and the samples for research were taken mainly from the soils rich with plant remains, as well as from the water of the Araz and Kura rivers.

Sampling, their preparation for laboratory analysis, taking

fungi to the pure culture were carried out in accordance by the methods and approaches accepted in mycology and currently widely used in similar work [8, 12].

Strains isolated from pure cultures were identification to the species based on determinants [1] compiled according to cultural-morphological and physiological characteristics.

During the study of antagonistic and antibiotic features [7, 13] were used from the phytopathogenic test cultures such of fungi as Alternaria alternata MB-1056, Aspergillus flavus MB-1217, Bipolaris nodulosa MB -2011, Fusarium oxysporum MB-7012, F.solani MB-7034 and Penicillium cyclopium MB-11276. These fungal strains were presented by the laboratory of mycological biotechnology of the Institute of Microbiology of ANAS.

To describe the interaction between Trichoderma and phytopathogenic fungi was used from the scale Canson and Carla modification by F.K Alimov [3] .

All experiments performed in the course of research were performed at least 4 repetitions, and the obtained results were statistically processed [10].

RESULTS AND THEIR DISCUSSION

As a result of the analysis of 250 soil, plant (feed, food and technical) and water samples taken from the Greater Caucasus and Kur-Araz lowlands of the Republic of Azerbaijan in 2014-2020, 1340 strains from the Mycota were taken to the pure culture. 270 of them belong to the genus Tricoderma. During determining their species composition, became clear that in general, the registered strains belong to 10 species and the distribution of the registered strains by species is shown in Table 1. As seen, most of the isolated strains, more precisely 67.1%, belongs to 4

Table 1: Distribution of fungi strains from the genus Trichoderma by individual species

N	Species	Number of strains belonging to a specific species (units)	Share (%) in total number
1	T.album	17	6.3
2	T.asperellum	35	13.0
3	T.atroviride	12	4.4
4	T.citrinoviride	21	7.8
5	T.harzianum	45	16.7
6	T.hamatum	48	17.8
7	T.koningi	19	7.0
8	T.longibrachiatum	12	4.4
9	T.oblongisporum	8	3.0
10	T.viride	53	19.6
Total		270	100

species(T.asperellum, T.harzianum, T.hamatum and T.viride). Inside the individual species T.viride are characterized by the highest, and T. oblongisporum with the lowest number of strains.

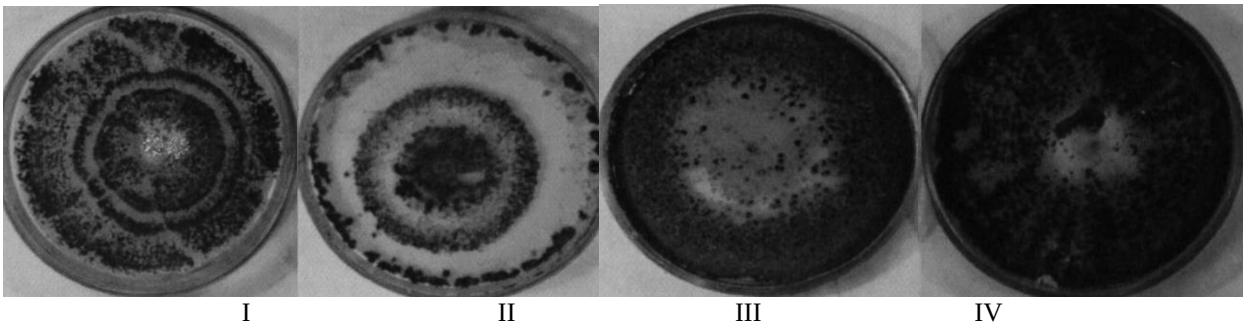
From the study of antagonistic properties of strains belongs to the recorded fungi become clear that in all fungi from the genus Trichoderma distributed in Azerbaijan were shown anatagonistic feature to the test cultures used. However, they differ from each other by the coefficient of antagonism, and in the formation of this difference are involved both the test cultures used and different species belonging to the genus Trichoderma(tab. 2). As seen, T. asperellum strains have the highest, and T. longibrachiatum strains the lowest indicator. The remaining strains takes middle position. In addition, individual strains differs by the quantitative indicator of the anatogenic coefficient, and sometimes this difference is more than twice. According to this indicator, the advantage belongs to strains of species such as T. asperellum and T. coningi.

Table 2: Characterization of antagonistic activity of fungi from the genus Trichoderma

		Number of strains used	Antagonistic coefficient			
			Alternaria alternata	Fuzarium oxysporym	F.solani	Bipolaris nodulosa
1	T.album	6	20-27	21-28	22-29	20-25
2	T.asperellum	8	20-52	27-57	30-59	31-53
3	T.atroviride	5	38-43	39-49	37-49	39-48
4	T.citrinoviride	5	20-40	24-50	27-51	27-44
5	T.harzianum	8	18-38	27-43	25-45	21-38
6	T.hamatum	7	26-30	25-29	25-28	27-29
7	T.koningi	6	32-50	39-54	35-51	32-51
8	T.longibrachiatum	5	18-24	23-30	24-29	19-27
9	T.oblongisporum	6	20-23	23-30	21-28	22-26
10	T.viride	7	20-31	24-32	27-35	23-34

Antagonistic coefficient of strains of species T.album. T.hamatum,T.longibrachiatum and T.oblongisporum are characterized by the lowest indicators. It should be noted that, the high antagonistic activity are considered strains those antagonist coefficient is 50 and higher[13]. From the obtained results became clear that among the studied strains, there are several that are prone to this feature and the most active of them are the strains such as T.asperellum G-3, as well as T.citrinoviride G-26, and T.koningi G-43. Interestingly, that all 3 of these strains were isolated from agrocenoses. Apparently, the high antagonistic activity of strains isolated from agrocenoses is due to the fact that phytopathogens are more common in such conditions, and the antagonistic potential of fungi against them gets into the active state. For strains isolated from water, this is not clearly observed, since fungi are migratory organisms for aquatic biotopes and the resistance of such ecosystems is relatively low.

According to some literature date, the antagonistic properties of one or another species of Trichoderma genus depend to some extent on their cultural morphological type (CMT), and in generally for fungi has been identified 4 CMT[13]. Each of these types also has its own characteristics, which are reflected in Figure 1.



I II III IV

To determine how accurate this, was determined the cultural morphological type of the fungi species belonging to the genus Trichoderma used in the study. It became clear that, some of these fungi have the characteristics of all groups of CMT, and some of them have the characteristics of several, but their relative quantities are different, and according to CMT, colonies characterizing the strains recorded in the studies mainly belong to group II (tab. 3).

Table 3: Characteristics of fungi strains from genus Trichoderma, according CMT formed by colonies

Species of fungi	Number of tested strains	Characteristics of colonies formed by the tested strains by CMT			
		I	II	III	IV
T.album	11	9.1	54.5	18.2	18.2
T.asperellum	22	9.1	59.0	18.2	13.7
T.atroviride	7	-	57.1	-	42.9
T.citrinoviride	12	16.7	58.3	25.0	-
T.hamatum	26	11.5	57.7	15.4	15.4
T.harzianum	33	6.1	60.5	15.2	18.2
T.koningii	11	18.2	54.5	-	27.3
T.longibrachiatum	8	12.5	62.5	25.0	-
T.oblongisporum	5	-	60.0	40.0	-
T.viride	38	2.7	55.2	-	42.1
Total units(%)	163(100)	14(8.6)	112(68.7)	19(11.7)	18(11.0)

When the results(tab.1) were analyzed from this point of view, no signs of this addiction were found. Thus, as noted above, more than half of CMT's belonging to the all species are suitable the type II(tab 2), and the highest rate (62.5%) of the species in this group corresponds to T. longibrachiatum. In species belonging to group II, such as T.album, T.koningii the relative amount is characterized by the lowest (54.5%) indicators. During characterizing the antagonistic activity of the noted species, T. koningi is characterized by the highest, and the other two species are characterized by the lowest indicators. In the species T.atroviride, T.koningi and T.viride were not found type belonging to the group III but their antagonistic coefficient are also have different quantitative characterized. The same can be said about of indicators specific to other species. So that, CMT is not a characteristic by quantity as a stable indicator of the biological activity of fungi, primarily of fungi belonging to the genus Trichoderma.

It would be better to touch a point related to the antagonistic features of fungi from the genus Trichoderma. This is due to the the fact that whether is relationship between the area from which the pure culture is extracted, and their activity. All strains, used against all test cultures, and in some cases having high antagonistic properties, were separated from soils that generally considered relatively clean. Thus, technogenic influences are not only a factor limiting the spread of fungi belonging to the genus Trichoderma, at the same time, it can be assessed as a factor weakens their biological activity.

The above-mentioned 3 strains, with high antagonistic activity were selected as a favorable producer in biological control for further research. It should be noted that the activity of the use of these strains in biological control is not only due to their antagonistic activity, it may also be associated with the synthesis of compounds characterized as antibiotics, and this has been confirmed in some studies. The results of these studies are associated with that antagonistic properties of fungi belonging to the genus Trichoderma, are due to the synthesis substances with antibiotic effect [5, 11].

For this reason, in research was also considered expedient to clarify of biosynthesis of compounds with antibiotic effect of strains isolated as active producers.

For this purpose, the synthesis of antibiotic substances of these 3 strains was carried out in accordance with the known methods in the sample of test cultures. The selection of test cultures is based on the study of fungi from the genus Trichoderma for future to use in biological control in agriculture. Thus, all the fungi selected as test culture are currently one of the most dangerous pathogens of mycobiota in cultivated plants, especially grains, widely grown in the world. It should be noted that today, at least 10% of the world's products are lost due to fungal diseases, and it is also known that the main role in this loss is played fungi which selected as test cultures. From the result of the experiments carried for this regard became clear that all three strains tested have the ability to synthesize metabolites with fungicidal activity and in this case, the T. asperellum G-3 strain is characterized by a higher indicators (tab. 4). Thus, in all cases, the antibiotic (fungicide) activity of the materials obtained from it is characterized by figures with a strong effect.

Table 4: Antibiotic activity of some strains from the genus Trichoderma

	Strains	Diameter of the zone (mm) where was stopped the growth of test cultures					
		Alternaria alternata	Aspergillus flavus	Bipolaris nodulosa	Fuzarium oxysporym	Fuzarium solani	Penicillium cyclopium
1	T.asperellum G-3	31	29	35	32	33	30
2	T.citrinoviride G-26	24	27	26	26	28	29
3	T.koningi G-43	27	31	34	30	32	28

A similar situation is also observed in T.koningi G-43, and the only exception manifests itself in the relationship to A.alternata. T. citrinoviride G-26 in all cases is characterized by moderate antibiotic activity.

As known, the antimicrobial activity of this strain is considered strong when the quantitative indicator of antibiotic activity assigned according to this method is higher than 29[7]. In this regard, it should be noted that strains of T.asperellum G-3, T.koningi G-43 have the ability to synthesize antibiotic compounds with high antimicrobial activity. This allows to note its successfully use in biological struggle is promising.

Comparing obtained results with the literature[13], became clear that the antibiotic activity of strains such as T. asperellum G-3, as well as T. koningi G-43 does not lag behind known strains and is even higher in some cases. For example, diameter of the lysis zone for strains such as T.koningii - TSL-06; TSQ, T. asperellum – 31; 119 are varies between 17-45 mm.

CONCLUSION

From the carried out of research became clear that in the territory of Azerbaijan spread 10 species of fungi belonging to the genus of Trichoderma, and among them are strains that have high antagonism. T. asperellum G-3, and T. koningi G-43 strains that are characterized by the highest rate of antagonism, at the same time have the ability to synthesize substances with antibiotic activity, and these indicator does not lag behind of those currently use in this area.

REFERENCE

1. Aleksandrova, A.V., Velikanov, L.L., Sidorova, I.I. 2006. A key for identifying the species from the genus Trichoderma. Mycology and phytopathology, 40,6: 457–468.
2. Alieva, G.R., Yusifova, A.A., Safaralieva, E.M., Agaeva, T.S., Ismailzade, N.N. 2020. Species composition of fungi of the genus Trichoderma Pers., spread in different cenoses in Azerbaijan. Journal "Modern Science: Actual Problems of Theory and Practice", Series: Natural and Technical Sciences, 2:6-9.
3. Alimova, F.K. 2006. Biological diversity of species of the genus Trichoderma (Fungi, Ascomycetes, Hipocreales) and their role in the functioning of microbiota and their role in the functioning of microbiota and plant protection in agrocenoses of various soil and climatic zones on the territory of the Republic of Tatarstan. Diss .... doct. biol. sciences. Kazan, , p 657 .
4. Abdel-Lateif, Kh.S. Trichoderma as biological control weapon against soil borne plant pathogens. African Journal of Biotechnology, 2017, vol. 16(50), p.2299-2306
5. Al-Mekhlafi, N.A., Abdullah, Q.Y., Al-Helali, M.F., Alghalibi, S.M. 2019. Antagonistic Potential of Native Trichoderma species against Tomato Fungal Pathogens in Yemen. Int. J. Mol. Microbiol., 2(1):1-10.
6. Anees, M., Tronsmo, A., Edel-Hermann, V., Hjeljord, L.G., Heraud, C., and Steinberg, C. 2010. Characterization of field isolates of Trichoderma antagonistic against *Rhizoctonia solani*. Fungal Biology 114:691-701
7. Egorova, N.S. 1995. A guide to practical exercises in microbiology. Textbook. Benefit. 3rd ed., Rev. and additional M .: Publishing house of MSU., 224:
8. Handbook of Mycological Methods, 2006. Retrieved from: http://www.fao.org/fileadmin/user_upload/agns/pdf/coffee/Annex-F.2.pdf
9. Hassan, M.M., Gaber, A., El-Hallous, E.I. 2014. Molecular and morphological characterization of Trichoderma harzianum from different Egyptian soils. Wulfenia J., 21:80–96
10. Kobzar, A.İ. 2006. Applied Mathematical Statistics. M .: FIZMATLIT, 816.
11. Kushwaha, M., Verma, A.K. 2014. Antagonistic activity of Trichoderma spp. (a bio-control agent) against isolated and identified plant pathogens. IJCBS, 1:1–6
12. Netrusov, A.İ., Egorova, M.A., Zakharchuk, L.M. and oth. 2005. Practical workon Microbiology. M .: Publishing Center "Academy", 608.
13. Sadikova, V.S. 2012. Ecology of fungi of the genus Trichoderma (Pers: Fr.) of the Yenisei River basin, their biological properties and practical use. Diss... doct. biol. sciences. Moscow, 417.
14. Sadikova, V.S., Kurakov, A.V., Kuvarina, A.E., Rogojin, E..A. 2015. Antimicrobial activity of strains of fungi from the genus Trichoderma from Central Siberia. Applied Biochemistry and Microbiology, 51,3:340-347
15. Ranelli, L.B., Hendricks, W.Q., Lynn, J.S., Kivlin, S.N. and Rudgers, J.A. 2015. Biotic and abiotic predictors of fungal colonization in grasses of the Colorado Rockies. Diversity and Distributions, 21:962–976
16. Singh, A., Shukla, N., Kabadwal, B., Tewari, A., Kumar, J. 2018. Review on plant-Trichoderma-pathogen interaction. Int. J. Curr. Microbiol. Appl. Sci. 7: 2382–2397.
17. Waghunde, R.R., Shelake, R.M. and Sabalpara A.N. 2016. Trichoderma: A significant fungus for agriculture and environment. African Journal of Agricultural Research, 11(22):1952-1965.
18. Yashenko, E.E., Grodnitskaya, N.D. 2000. Influence of Trichoderma fungi to soil micromycetes causing infectious lodging of coniferous seedlings in forest nurseries of Siberia. Microbiology, 69,6:850-854.