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**STUDY OF THE COMPATIBILITY OF *TRICHODERMA* AND  
*BACILLUS* STRAINS FOR THE DEVELOPMENT OF A  
BIOTECHNOLOGICAL DRUG**

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**Abstract.** *A study of the antimicrobial activity of Trichoderma strains against Bacillus strains to determine the possibility of their compatibility in a biotechnological preparation was conducted. It was found that Trichoderma strains inhibited the growth of Bacillus strains with antimicrobial activity that were isolated from environmental objects, however, towards commercial strains (B. amyloliquefaciens QST 713 and B. subtilis M-22 VIZR), Trichoderma strains were neutral. Based on the obtained results, a combination of strains for the development of a new drug to protect plants from phytopathogenic fungi is proposed.*

**Keywords:** *antagonistic activity, strains compatibility, consortium, biotechnological preparation.*

**Introduction.** To create a modern biotechnological drug, a consortium is most often used. Consortium is a microbial association of two or more microorganisms, which can be archaea, fungi, bacteria, viruses or algae [7].

A combination of *Trichoderma spp.* and *Bacillus spp.* is one of the most commonly used for biological control of phytopathogens [2, 4, 5]. However, in this case, direct or indirect interactions between different biological agents may occur, which may lead to a negative or positive effect on the effectiveness of the biopreparation [6].

Therefore, we investigated the antagonistic activity of *Trichoderma* strains against different strains of the genus *Bacillus* obtained from different sources.

**Materials and methods.** To study the antagonistic activity of strains of *Trichoderma* against strains of microorganisms, some *Bacillus* strains obtained from different sources were used. *Bacillus megaterium* LBX.001, *Bacillus circulans* LBX-003, *Bacillus subtilis* LBX-288, *Trichoderma viride* LBX-174 (working name *Trichoderma viride* No. 8) and *Trichoderma harzianum* LBX-181 (working name *Trichoderma harzianum* No. 9) from the museum of enterprise «Scientific production association "Agrobioinovatika"»; *Bacillus*

*megaterium* MV B-7168 from the preparation "Biophosphorin"; *Bacillus amyloliquefaciens* QST 713 from the fungicide "Serenada ASO SC" ; *Bacillus subtilis* M-22 VIZR from the drug "Ghamair"; *Bacillus subtilis* ONU 559 from collection of the Department of Microbiology, Virology and Microbiology of Odesa I.I. Mechnikov National University.

*Bacillus* strains were cultivated on MPA medium. *Trichoderma* strains were cultivated on Sabouraud's and MPA media.

To evaluate the antagonistic activity of micromycetes, the method of mixed (counter) cultures was used [3]. Petri plates with Sabouro and MPA nutrient media were inoculated with microorganisms at the opposite poles: *T. viride* No. 8 and *T. harzianum* No. 9 against *Bacillus* strains.

The results were calculated on the 5th day of cultivation at a temperature of 28 °C.

**Results and discussion.** According to the results shown in Table 1, we can see that the tested *Trichoderma* strains showed high antagonistic activity against *B. megaterium* LBX.001 and *B. circulans* LBX-003 strains. It should be noted that when MPA medium was used, the level of antagonistic activity of *Trichoderma* strains was somewhat lower (Table 1).

Table 1

**Antagonistic activity of *T. viride* No. 8 and *T. harzianum* No. 9 strains against *Bacillus* strains**

Test strains	Medium MPA		Medium Sabouro	
	<i>T. viride</i> № 8	<i>T. harzianum</i> № 9	<i>T. viride</i> № 8	<i>T. harzianum</i> № 9
<i>B. megaterium</i>	++	++	+++	+++
<i>B. subtilis</i> M-22 ВИЗР	n	n	n	n
<i>B. subtilis</i> ONU 559	++	++	+++	+++
<i>B. amyloliquefaciens</i> QST 713	n	n	n	n
<i>B. megaterium</i> MB B-7168	n	++	+++	+++
<i>B. subtilis</i> LBX-288	n	++	+++	+++
<i>B. circulans</i> LBX-003	++	++	++	++

Note: n – neutralism; ++ weak antagonism; +++ strong antagonism

A similar pattern was observed when studying the effect of *Trichoderma* strains on *B. subtilis* LBX-288 (Table 1).

The strain *B. subtilis* ONU 559, isolated from the bottom sediments of the Black Sea, showed high antimicrobial activity against conditionally pathogenic microorganisms [1]. However, in co-cultivation with strains of *T. viride* No. 8 and *T. harzianum* No. 9, the antagonistic effect of fungi against *B. subtilis* ONU 559 was recorded (Table 1).

Towards the *B. circulans* LBX-003 strain, the level of antagonistic activity of strains *T. viride* No. 8 and *T. harzianum* No. 9 was defined as "weak antagonistic activity" (Table 1).

Towards *B. amyloliquefaciens* QST 713 and *B. subtilis* M-22 VIZR strains, both *Trichoderma* strains demonstrated neutrality (Table 1).

The *B. megaterium* MV B-7168 strain is sensitive to the antagonistic activity of *T. viride* No. 8 and *T. harzianum* No. 9 strains under the conditions of their co-cultivation on Sabouraud's medium (Table 1).

When conducting this series of experiments, we also investigated the presence of mutual antagonistic activity of *Bacillus* strains. The obtained results are shown in the table 2.

Table 2

### Mutual antagonistic activity of *Bacillus* strains

Strains	Strains						Marking: n - neutralism + - antagonism ++ - antagonism with admiration // - bilateral antagonism
	1	2	3	4	5	6	
1		++	H	++	H	H	<b>Strains</b> 1. <i>B. megaterium</i> LBX.001 2. <i>B. subtilis</i> M-22 ВИЗР 3. <i>B. subtilis</i> ONU 559 4. <i>B. amyloliquefaciens</i> QST 713 5. <i>B. megaterium</i> MB B-7168 6. <i>B. subtilis</i> LBX-288
2	++		H	++	H	H	
3	H	H		H	H	H	
4	++	+	H		H	//	
5	H	H	H	H		H	
6	H	H	H	//	H		

As we can see, according to the above results, the strain *B. subtilis* ONU 559 was neutral to most strains, as were the strains *B. megaterium* MV B-7168 and *B. subtilis* LBX-288 (Table 2). The strain *B. amyloliquefaciens* QST 713 showed bilateral antagonism when it was co-cultivated with the strain *B. subtilis* LBX-288. The strain *B. subtilis* M-22 VIZR showed antagonistic activity against the strain *B. subtilis* ONU 559 and antagonism with capture against the strain *B. megaterium* LBX.001 (Table 2).

## Conclusions

1. *T. viride* No. 8 and *T. harzianum* No. 9 strains showed strong antagonistic activity against microorganisms of the genus *Bacillus*. Strains isolated from natural sources (*B. subtilis* ONU 559, *B. megaterium* LBX.001, *B. subtilis* LBX-288 and *B. circulans* LBX-003) were not resistant to the antimicrobial activity of *Trichoderma* strains, unlike commercial strains (*B. amyloliquefaciens* QST 713 and *B. subtilis* M-22 VIZR).

2. The proposed consortium for the creation of a new biotechnological drug is *T. viride* No. 8 or *T. harzianum* No. 9 + *B. amyloliquefaciens* QST 713.

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## ДОСЛІДЖЕННЯ СУМІСНОСТІ ШТАМІВ *TRICHODERMA* ТА *BACILLUS* ДЛЯ РОЗРОБКИ БІОТЕХНОЛОГІЧНОГО ПРЕПАРАТУ

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**Анотація.** Проведено дослідження антимікробної активності штамів *Trichoderma* по відношенню до штамів *Bacillus* з метою визначення можливості їх сумісності в біотехнологічному препараті. Було встановлено, що штами триходерми пригнічували ріст штамів *Bacillus* з антимікробною активністю, які були ізольовані з об'єктів навколишнього середовища, однак по відношенню до комерційних штамів (*B. amyloliquefaciens* QST 713 та *B. subtilis* M-22 ВІЗР) штами триходерми були нейтральними. На основі отриманих результатів запропоновано комбінацію штамів для розробки нового препарату для захисту рослин від фітопатогенних грибів.

**Ключові слова:** антагоністична активність, сумісність штамів, консорціум, біотехнологічний препарат.

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