

Antifungal potential and biotechnological evaluation of strains of the genus *Trichoderma* as the basis for the biopreparation Ecostern *Trichoderma*

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ABSTRACT

A biotechnological evaluation was conducted on strains of the genus *Trichoderma* as key components of biodestructors and soil-improving preparations in the Ecostern® range. Their cultural and morphological characteristics, antagonistic activity against phytopathogenic microfungi, as well as the stability and antifungal efficacy of the Ecostern® *Trichoderma* biological preparation (gel form) during storage for 3 months and its effect on the structure of the soil microbiota were investigated. The ability of *Trichoderma* strains to effectively inhibit the growth of *Cladosporium herbarum*, *Botrytis cinerea*, *Alternaria alternata*, *Drechslera sorokiniana*, *Fusarium* spp., *Rhizoctonia solani* and *Sclerotinia sclerotiorum* was established. The highest antifungal activity was recorded on potato-dextrose agar and a medium containing barley malt extract, where the strains *T. harzianum* 11/16 TM demonstrated the maximum antagonism index (30–31 points), while *T. harzianum* 23/1 and *T. viride* 44/13 LM exhibited high levels of activity (27–28 and 28.5 points, respectively). On Chapek's medium, antifungal activity decreased by an average of 2.2-fold. The strain of BTU Biotechnology Company *T. harzianum* surpassed its foreign counterpart in biocontrol potential, demonstrating pronounced mycoparasitism and inhibition of sclerotium formation in *S. sclerotiorum*. After 3 months of storage, the biopreparation was characterized by a stable titre ($1.3\text{--}3.0 \times 10^7$ CFU/cm³) and high antifungal activity (79.9–100%) regardless of dilution. A positive effect on the soil mycobiota was also established, with a 5.3-fold reduction in pathogen numbers and an increase in the proportion of saprotrophic fungi to 97.9%. A comprehensive assessment of the antifungal activity and synergistic interaction of microbial strains of the genus *Trichoderma* enables the identification of patterns governing the development of their biological activity and viability. The results obtained provide a theoretical and practical basis for the development of optimally balanced biological products with high efficacy against plant pathogens.

Keywords: *Trichoderma*, antifungal activity, biodestructors, plant pathogens, titre stability.

INTRODUCTION

The global market for biostimulants, biofungicides, biofertilizers and other biological products is currently experiencing rapid growth. This trend is driven by the agricultural sector's accelerated transition towards sustainable farming practices and the growing demand for biological products that minimize human impact on agroecosystems (Bolokhovska et al., 2025; Cortez-Lázaro et al., 2025). The global market of bioproducts based on *Trichoderma* spp. biopesticides, estimated to be worth approximately \$1.5 billion in 2025, will grow at a compound annual growth rate (CAGR) of 8.0% from 2025 to 2033, reaching an estimated market value of \$2.8 billion by 2033 (Meher et al., 2020).

Species of the genus *Trichoderma* have a wide range of applications, including use as biofungicides for plant disease control, biofertilizers or plant growth promoters. The vast majority of studies on *Trichoderma* species have focused on investigating the efficacy and mechanisms of mycoparasitism (adhesion, penetration and infection of pathogen hyphae), the production of enzymes with various modes of action, antibiotic synthesis, competition for nutrients and ecological niches. Thus, certain strains of the genus *Trichoderma* are characterized by a high capacity for rapid colonization of plant seeds and the rhizosphere, which ensures the competitive displacement of phytopathogenic microorganisms when biopreparations are used to treat seeds or the root system of plants (Sharma et al., 2019; Mukhopadhyay et al., 2020). A bibliometric analysis of publications indexed in Scopus (2000–2025) revealed an exponential growth in research clusters of scientific output regarding the shift from applied research to studies focusing on the molecular-genetic and physiological mechanisms of ‘plant-microorganism’ interactions, the mediated activation of induced resistance systems in plants to biotic and abiotic stresses, the ability of *Trichoderma* spp. to colonize the root system as endophytes, and other issues (Mukherjee et al., 2022; Cortez-Lázaro et al., 2025). Significant attention is being paid to the study of the synthesis of various secondary metabolites, which include different classes of natural compounds, among which are both volatile compounds (alcohols, aldehydes, ketones, ethylene, hydrogen cyanide, monoterpenes) and non-volatile biologically active substances, in particular peptaibols, glyotoxin and

glyovirin of diketopiperazine-like nature (Vinale et al., 2020; Mukherjee et al., 2022).

Secondary metabolites of *Trichoderma* spp. exhibit significant antimicrobial activity and also have a positive effect on plant metabolism, enhancing disease resistance by activating the plants' defense system and promoting their growth (Mukhopadhyay et al., 2020; Vinale et al., 2022).

The growth of the market for biological products based on *Trichoderma* spp. is driven by a range of interrelated factors, among which a key role is played by increased awareness among agricultural manufacturers of the benefits of biodestructors, biostimulants, biofertilizers and biocontrol agents as environmentally safe alternatives to chemical products. Additionally, the increasing prevalence and virulence of soil-borne plant pathogens, as well as tighter government regulation aimed at implementing the principles of sustainable agriculture, contribute to this growth.

Around 60% of the fungal-based biofungicides available on the market are products in various formulations based on species of the genus *Trichoderma*. Their effectiveness and demand are due to the presence of a complex of complementary biocontrol mechanisms, including antibiosis, rapid colonization, competition for nutrients and ecological niches, as well as direct mycoparasitism. As a result, fungi of the genus *Trichoderma* demonstrate high efficacy against a broad spectrum of field phytopathogens, including those of the genera *Sclerotinia*, *Rhizoctonia*, *Fusarium*, *Macrophomina*, *Phytophthora*, *Phyllactinia*, *Colletotrichum*, *Cladosporium* and others, as well as fungi that infect produce during storage, such as species from the genera *Penicillium*, *Aspergillus*, *Rhizopus*, *Botrytis* and others (Meher et al., 2020).

A significant contribution to the effectiveness of biological control agents is made by manufacturers of biological products through research into and the identification of highly effective strains of *Trichoderma* spp., as well as improvements in cultivation methods and formulations. At the same time, there are a number of limiting factors, including the instability of the quality of biological products during storage, a reduction in the biological activity of strains, the difficulty of scaling up production for widespread commercial distribution, and the lack of unified and standardized application technologies, which hinders the widespread adoption of biological control agents in production and requires further

scientific and technological solutions (Pedrero-Méndez et al., 2021; García-Latorre et al., 2025).

The development of cost-effective biological products, adapted to specific soil and climatic conditions and the needs of individual crops, plays a decisive role in the development of this market segment, including in Ukraine. Of particular importance are innovations in formulations aimed at enhancing the antifungal activity and stability of products, extending shelf life, optimizing application methods and ensuring compatibility with other chemical and biological agents (Bolkhovskiy et al., 2025). A particularly promising strategy is to combine several isolates of the genus *Trichoderma* with different modes of action, thereby enhancing the efficacy and stability of biocontrol.

The effectiveness of biological products based on fungi of the genus *Trichoderma* is attributed to the biological activity and viability of strains, which is a relevant issue for research. An important and insufficiently researched aspect is the comprehensive study of antifungal activity and the synergistic interaction of different strains of microorganisms, leading to the development of optimally balanced microbial preparations.

The aim of the research is to investigate the antifungal potential and biotechnological evaluation of *Trichoderma* spp. strains as the basis for soil-improving and soil-restoring products in the Ecostern® range manufactured by the Ukrainian biotechnology company BTU.

MATERIAL AND METHODS

The subjects of the study

The study was conducted at the mycology laboratory of the Institute of Applied Biotechnology Ltd (Kyiv, Ukraine). The subjects of the study were strains of the genus *Trichoderma* (*T. viride* 44/13 LM, *T. koningii* 1220-KL, *T. reesei* 320-TN/L, *T. harzianum* 21, 23/1, 11/16 TM), test cultures of phytopathogenic micromycetes from the Collection of Microscopic Fungal Cultures of the biotechnology company BTU (Ukraine), the Ecostern® *Trichoderma* biodestructor, and the soil mycobiota during rapeseed cultivation under the influence of Ecostern® *Trichoderma* in the Western Forest-Steppe region of Ukraine.

The following plant pathogens were used as test cultures: *Alternaria alternata* (Fr.) Keissl. (the causative agent of *Alternaria* leaf spot,

ear rot and black seedling blight), *Drechslera sorokiniana* (Sacc.) Shoemaker (the pathogen responsible for root rot, leaf spot in cereal crops and black seedling blight), *Fusarium sporotrichioides* Sherb., *F. solani* (Mart.) Sacc., *F. verticillioides* (Sacc.) Nirenberg, *F. culmorum* (Wm.G.Sm.) Sacc. (pathogens causing fusarium root rot, fusarium head blight and fusarium wilt), *Rhizoctonia solani* J.G. Kühn (the causative agent of rhizoctonia or black scab), *Acrostalagmus luteo-albus* (Link) Zare, Gams & Schroers (syn. *Verticillium lateritium* Berk.) (the causative agent of dry rot and verticillium wilt), *Sclerotinia sclerotiorum* (Lib.) de Bary (the causative agent of white rot), *Botrytis cinerea* Persoon (the causative agent of grey rot), *Cladosporium herbarum* (Pers.) Link (the causative agent of olive mould and seed rot).

Study of morphological characteristics and antifungal activity of *Trichoderma* spp. strains

To study the morphological characteristics of strains of the genus *Trichoderma*, they were inoculated onto potato dextrose agar (PDA) medium (Borkar, 2017). The plates were incubated at a temperature of 25 °C. On the 4th and 14th days, the morphological structures were examined using light microscopy. The antifungal activity of *Trichoderma* spp. strains against plant pathogens was investigated using the double culture method (agar block method) on PDA medium (Badalyan et al., 2002, 2004). Agar blocks with a diameter of 5 mm were obtained from seven-day-old colonies of *Trichoderma* spp. and phytopathogens grown on PDA at a temperature of $+25\text{ °C} \pm 2\text{ °C}$.

Agar blocks containing *Trichoderma* spp. and the phytopathogen were placed opposite each other, 1 cm from the edge of the Petri dish. In the control, blocks of the corresponding phytopathogen test cultures were cultured separately, also placed 1 cm from the edge on one side of the dish. The antagonism index of *Trichoderma* strains was assessed using a rating scale based on 5 main reaction types (A, B, C, D, E) and 4 subtypes (C_{A1} , C_{B1} , C_{A2} and C_{B2}) (Atamanchuk et al., 2024) and calculated using the Equation 1:

$$AI = A(n \times 1) + B(n \times 2) + C(n \times 3) + C_{A1}(n \times 3.5) + C_{B1}(n \times 4) + C_{A2}(n \times 4.5) + C_{B2}(n \times 5) + D(n \times 0) + E(n \times 0) \quad (1)$$

where: n – the frequency of each type or subtype of reaction associated with a given category or categories ($A, B...D$), resulting in a corresponding score; greater weight was given to categories representing inhibitory reactions.

The biotechnological evaluation of the strains involved studying the antifungal activity of *Trichoderma* spp. strains under the influence of culture medium components. The initial cultures were grown on PDA and a medium containing barley malt extract (BME), which were subsequently transferred to PDA, Čapek's medium and a medium containing BME for comparison of antagonistic potential. The plates were incubated at +25 °C. On the 4th and 14th days, the following parameters were examined using light microscopy: colony color, medium pigmentation, production of a specific odor; shape and size of fungal structures, structure of conidiophores, and growth rate.

The assessment of the antagonistic potential of domestic (*Trichoderma harzianum* (BTU, Ukraine)) and foreign (*T. harzianum*, China) microbial preparations against plant pathogens was carried out in the microbiological laboratory of Agricultural LLC “Druzhba-Nova”, using the method described by Kurchenko et al., 2021). The test cultures used were *Fusarium boothii* (O'Donnell, T. Aoki, Kistler & Geiser) (the causative agent of fusarium head blight in cereal crops and stalk rot in maize), *Macrophomina phaseolina* (Tassi) Goid (the causative agent of charcoal rot in soybeans, sunflowers and maize), *V. lateritium* Berk., *S. sclerotiorum* (Lib.) de Bary.

Study of the antifungal activity of the biological product Ecostern® *Trichoderma*

The antifungal activity of the biological product Ecostern® *Trichoderma* (BTU Biotechnology

Company, Ukraine), which contains spores and mycelium of antagonistic fungi of the genus *Trichoderma*, including *Trichoderma harzianum* 11/16 TM, *T. viride* 44/13 LM, *T. koningii* 1220-KL, *T. reesei* 320-TN/L, as well as their metabolic products and other biologically active substances, was assessed after storage for 3 months, using the method described by Kurchenko et al. (2021). Samples of the biological preparation in dilutions of 1:10, 1:100 and 1:500 were added in quantities of 1 ml to the PDA growth medium. After the medium had solidified, an agar block (5 mm in diameter) of a 5-day-old test culture of the phytopathogenic fungus was placed in the center of the surface. Plates containing sterile water (1 ml) served as controls. The cultures were incubated at +25 °C for 5–7 days. The diameter of the test culture colonies was measured on the 12th day of growth. The results were interpreted according to the activity scale (Table 1).

Analysis of the pathogenic and saprotrophic soil mycobiota

An analysis of the pathogenic and saprotrophic soil mycobiota under rapeseed crops following treatment with the product Ecostern® *Trichoderma* was conducted in Western Ukraine in accordance with DSTU 7847:2015 “Soil quality. Determination of the number of microorganisms in soil by the method of inoculation onto a solid (agarized) culture medium” and methods generally accepted in microbiology (Borkar, 2017).

Data analysis

All treatments were replicated three times. Statistical analysis was performed using Statistica 12 (StatSoft Inc., USA) and Microsoft Excel 2010. To evaluate the effects of strains and bio-preparations, a one-way analysis of variance (ANOVA) was conducted, followed by Tukey's HSD test. Differences were considered statistically significant at a p -value < 0.05.

RESULTS

The cultural and morphological characteristics of the *Trichoderma* strains

The biotechnological evaluation of *Trichoderma* strains involves systematically monitoring

Table 1. Assessment of the biological product's activity against test cultures of phytopathogenic fungi using the agar block method

Activity level	Activity of bioagents relative to the test culture, %
High	100–75
Medium	74–50
Low	<50

their antifungal activity, analyzing the stability of their viability and titre during storage, and experimentally validating their effectiveness under production conditions.

The first stage of the research (Figure 1) involved studying the cultural and morphological characteristics of the strains.

The strain *T. viride* 44/13 LM formed colonies with white, cotton-like mycelium; from the third day conidial sporulation was observed in the form of small green cushions; as the mycelium aged, it became prostrate and low. The underside of the colony showed no pigmentation. Conidiophores were tree-like; the branches mainly diverged oppositely at right angles, increasing in length from the apex to the base, thus forming a pyramidal shape. Conidia were spherical, broadly ellipsoidal.

Strain *T. koningii* 1220-KL formed colonies with a sparse, powdery mycelium in the center and a sparse, cobweb-like mycelium at the edges; from the third day, weak sporulation was observed, which over time took the form of large white and light grey-green pads arranged in concentric circles. The reverse side was pigmented, with yellow pigment being secreted into the medium. Conidia ranged from broadly ellipsoidal to ellipsoidal, and conidiophores were branched, predominantly with paired compact branches. On the 7th day of aerial mycelium growth of strain *T. harzianum* 21, conidial sporulation was observed in the form of light yellow-white cushions, which took on a greenish hue as the culture aged and formed a colony with a cottony mycelium. The reverse side of the colony was unpigmented. Microscopic examination revealed conidiophores with a narrow main axis, narrow-flask-shaped phialides that were not overfilled, and spherical, ovoid conidia. The strains *T. viride* 44/13 LM, *T. koningii* 1220-KL and *T. harzianum* 21, exhibited varying growth rates and characteristics of colony

development on PDA medium. The *T. koningii* 1220-KL strain exhibited the slowest growth rate.

The growth, morphogenesis and antagonistic activity of *Trichoderma* spp. strains depend significantly on the composition of the culture medium. The use of different culture media allows for a comprehensive assessment of the biological potential of *Trichoderma* strains, in particular their growth activity and antagonistic properties. The influence of PDA, Čapek and barley malt extract (BME) culture media on the growth and development characteristics of strains *T. reesei* 320-TN/L, *T. harzianum* 23/1 and *T. harzianum* 11/16 TM has been established.

Strain *T. reesei* 320-TN/L on PDA formed a colony in the center with a powdery, low-lying conidial sporulation of dark green color, and along the edge of the colony in the form of small white and light grey-green cushions. There was no odor, and the underside of the colony was yellow, as was the surrounding medium. On BME medium, colony growth was observed in the form of large light grey-green cushions throughout the colony. On Čapek medium, growth was very poor. Microscopic examination revealed broad conidiophores with short, compact branches extending at right angles, on which flask-shaped phialides containing narrow-ellipsoidal conidia were formed (Figure 2).

Strain *T. harzianum* 23/1 exhibited similar colony morphology and growth on BME, PDA and Čapek media, regardless of the medium used. On BME and PDA media at a temperature of +25 °C, this strain formed colonies with a fluffy, cobweb-like mycelium. Spreading colonies were observed, with abundant aerial mycelium and sporulation of a pale blue-green colour. Very intense sporulation was observed on the BME medium. The micromycetes grew without a distinct odour and had an unpigmented reverse side. Colony growth on the Čapek medium was slower compared to the others and was characterized by weak sporulation

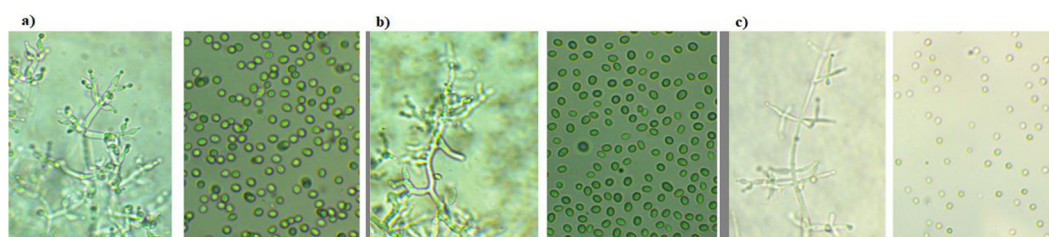


Figure 1. Morphology of conidiophores and conidia of the strains: (a) *Trichoderma viride* 44/13 LM, (b) *T. koningii* 1220-KL, (c) *T. harzianum* 21

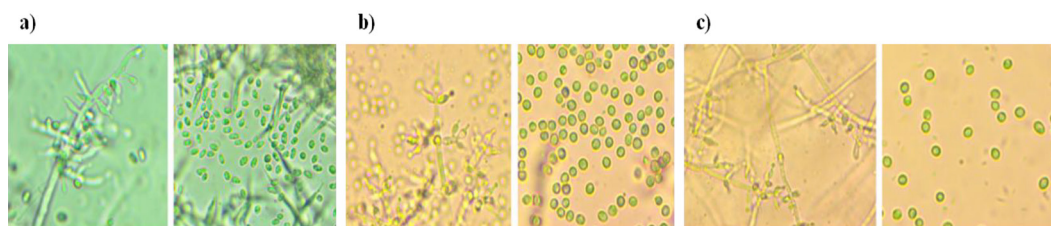


Figure 2. Morphology of conidiophores and conidia of the strains: (a) *Trichoderma koningii* 320-TN/L, (b) *T. harzianum* 23/1, (c) *T. harzianum* 11/16 TM

of a yellow-green color. Conidiophores were tree-like, with branches mainly diverging at right angles, increasing in length from the apex to the base, thus forming a pyramidal shape. Spherical, broadly ellipsoidal conidia and short, flask-shaped phialides were observed.

Strain *T. harzianum* 11/16 TM, like strain 23/1, exhibited similar morphology and colony growth on BME, PDA and Čapek media, regardless of the medium used. On the BME medium, intensive sporulation of *T. harzianum* 11/16 TM, pale blue-green in colour, was observed compared to other culture media. The colonies had a cottony mycelium. Colony growth was observed as a spreading mycelium with weak sporulation, yellow-green in colour on PDA medium and yellow-lemon on Čapek medium. The mycelium had no distinct odor, and the underside of the colony showed no pigmentation.

Antagonistic activity and interaction types of *Trichoderma* spp. against plant pathogens

Based on the results of studies on the interaction between strains of the genus *Trichoderma* and phytopathogen cultures, the antagonism index (AI) was calculated. Table 2

provides a summary of the results of the studied strains inhibited the growth of test cultures of phytopathogenic fungi.

The *T. viride* 44/13 LM strain grew on the mycelium of the pathogens *A. alternata*, *C. herbarum* and *V. lateritium*, forming a zone of growth inhibition (interaction type C_{B2}), and interaction type C_{B1} with *F. culmorum* and *F. verticillioides* – partial overgrowth of the colony with the formation of a zone of growth inhibition. Partial overgrowth of the *B. cinerea* colony was observed, without a zone of growth inhibition. Growth arrest of *D. sorokiniana* occurred prior to contact with strain *T. viride* 44/13 LM, with a zone of inhibition forming.

The *T. koningii* 1220-KL strain completely overgrew the fungi *V. lateritium* and *A. alternata* without forming a zone of growth inhibition; it partially overgrew the mycelium of *F. verticillioides* without forming a zone of growth inhibition; and it overgrew *C. herbarum*, forming a zone of growth inhibition. Growth inhibition of *D. sorokiniana* with the formation of an inhibition zone occurred prior to contact with the *T. koningii* 1220-KL strain. The pathogens *F. culmorum* and *B. cinerea* exhibited interaction types D and E, in which the phytopathogen test culture inhibits *T. koningii* 1220-KL.

Table 2. Type of interaction and antagonism index of *Trichoderma* spp. strains in relation to test cultures of phytopathogens (original culture grown on PDA, max = 35 points)

Test cultures of phytopathogens	Strains of <i>Trichoderma</i> spp.		
	<i>T. viride</i> 44/13 LM	<i>T. koningii</i> 1220-KL	<i>T. harzianum</i> 21
<i>Alternaria alternata</i>	C_{B2}	C_{A2}	C_{A2}
<i>Botrytis cinerea</i>	C_{A1}	D	C_{A1}
<i>Cladosporium herbarum</i>	C_{B2}	C_{B2}	C_{A2}
<i>Drechslera sorokiniana</i>	B	B	B
<i>Fusarium culmorum</i>	C_{B1}	E	C_{A1}
<i>Fusarium verticillioides</i>	C_{B1}	C_{A1}	C_{A1}
<i>Verticillium lateritium</i>	C_{B2}	C_{A2}	C_{A2}
Antagonism index (AI), max=35	28.5	19.5	26.0

Strain *T. harzianum* 21 completely overgrew colonies of *A. alternata*, *C. herbarum* and *V. lateritium*, and partially overgrew *B. cinerea*, *F. verticillioides* and *F. culmorum* without a zone of inhibition. When *T. harzianum* 21 interacted with *D. sorokiniana*, growth inhibition was observed with the formation of an inhibition zone.

Strain *T. viride* 44/13 LM exhibited the highest degree of antagonism towards phytopathogens; its AI was the highest among the three strains studied, amounting to 28.5 points out of a maximum of 35 points. The *T. harzianum* 21 strain also exhibited a high degree of antagonism and had an AI of 26.0 points. The aggressiveness (colony-forming activity on test cultures) of the *Trichoderma* spp. strains was similar and manifested in their ability to grow completely or partially over 6 colonies out of the 7 pathogenic fungal species studied. Strain *T. koningii* 1220-KL grew completely or partially over 4 colonies (AI 19.5 points).

To investigate antagonistic activity against plant pathogens, initial cultures of the strains *T. koningii* 320-TN/L, *T. harzianum* 23/1 and *T. harzianum* 11/16 TM were inoculated onto PDA, BME and Čapek media, and the types of their interaction and the antagonism index were determined (Table 3).

The effect of nutrient media of different compositions on the antifungal activity of strains of the genus *Trichoderma* 23/1 and 11/16 TM was established (Table 4).

The highest degree of antagonism towards the test cultures of phytopathogens was exhibited by the strain *T. harzianum* 11/16 TM, with an AI of 31 points out of a possible 40. Similarly, the strain *T. harzianum* 23/1, showed an antagonism index of 28 points on the PDA medium and 27.5 points. The antagonistic ability of these strains was evident in their ability to fully or partially inhibit growth of 7 and 6 colonies out of 8 studied phytopathogens, respectively.

Studies conducted to determine the influence of the nutrient medium on the AI of the *T. harzianum* 23/1 strain showed high antifungal

activity on these media. Under growth conditions on the Čapek medium, this strain exhibited a 2-fold decrease in aggressiveness. Pathogens showed a more rapid growth and development rate compared to the 23/1 strain. The *T. harzianum* 23/1 strain suppressed the growth and development of the fungi *A. alternata*, *B. cinerea*, *F. verticillioides*, *F. solani*, *F. culmorum*, *R. solani*, *V. lateritium* on the BME and PDA media. Conversely, on Čapek's medium, phytopathogens of the genera *Fusarium* and *Rhizoctonia* suppressed *T. harzianum* 23/1.

The results of determining the influence of the nutrient medium on the AI of the *T. harzianum* 11/16 TM were similar to the results obtained for the *T. harzianum* 23/1. The *T. harzianum* 11/16 TM exhibited antifungal activity against all phytopathogens on BME and PDA. However, on the Čapek medium, *F. verticillioides*, *F. solani*, *F. culmorum* and *R. solani* inhibited *T. harzianum* 11/16 TM.

Thus, the use of different nutrient media allowed for a comprehensive assessment of the biological potential of strains of the genus *Trichoderma*, particularly their growth, sporulation and antagonistic properties. Research into the properties of strains of the genus *Trichoderma* made it possible to select and deposit the best of them and to create the biologics Ecoster® *Trichoderma*.

Figure 3 shows the assessment of the antagonistic potential of the bioagents Ecoster® *Trichoderma* (*Trichoderma harzianum* (BTU, Ukraine)) and a foreign microbial preparation (*T. harzianum*, China) against phytopathogens during 21 days of cultivation revealed significant differences depending on the antagonist strain.

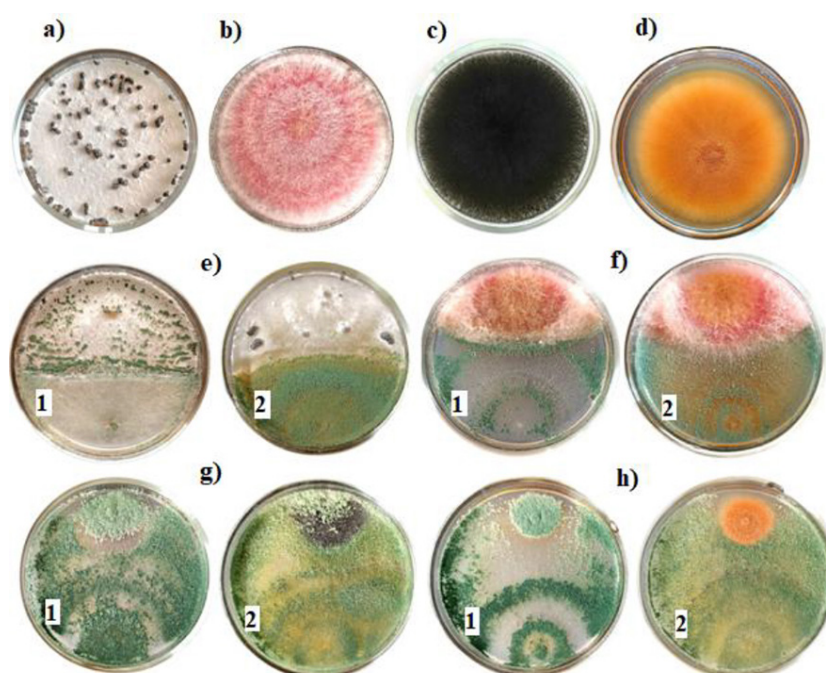
It was found that *T. harzianum* (BTU, Ukraine) exhibits pronounced direct hyperparasitism against *S. sclerotiorum*, which is accompanied by lysis of the pathogen's mycelium and significant inhibition of sclerotia formation. In contrast, the strain *T. harzianum* (China) is characterized mainly by a competitive type of antagonism

Table 3. Type of interaction and antagonism index of *Trichoderma reesei* 320-TN/L in relation to test cultures of phytopathogens (original culture grown on PDA, max = 40 points)

Nutrient media	Test cultures of phytopathogens								Antagonism index (AI)
	<i>Alternaria alternata</i>	<i>Botrytis cinerea</i>	<i>Drechslera sorokiniana</i>	<i>Fusarium culmorum</i>	<i>Fusarium verticillioides</i>	<i>Fusarium solani</i>	<i>Rhizoctonia solani</i>	<i>Verticillium lateritium</i>	
BME	D	A	B	E	A	C _{A1}	E	C _{A1}	11,0
PDA	A	C _{A1}	B	E	D	B	E	B	10,5

Table 4. The influence of the nutrient medium on the type of interaction and antagonism index of the *Trichoderma harzianum* 23/1 and *T. harzianum* 11/16 TM strains in relation to test cultures of phytopathogens

Test cultures of phytopathogens	The medium on which the original strain was grown					
	BME			PDA		
	media for studying the antifungal activity of the strain					
	BME	Čapek	PDA	BME	Čapek	PDA
	<i>T. harzianum</i> 23/1					
<i>A.alternata</i>	B	A	C _{A1}	A	C _{A1}	C _{A1}
<i>B.cinerea</i>	C _{B2}	C _{B2}	C _{B1}	C _{B2}	C _{B2}	C _{B2}
<i>D.sorokiniana</i>	B	B	B	B	B	B
<i>F.culmorum</i>	C _{A1}	E	C _{A1}	C _{A2}	E	C _{A1}
<i>F. verticillioides</i>	C _{A1}	E	C _{B1}	C _{A1}	E	C _{B1}
<i>F. solani</i>	C _{A1}	E	A	C _{A1}	E	A
<i>R. solani</i>	C _{A1}	D	C _{B2}	C _{A1}	D	C _{A2}
<i>V.lateritium</i>	C _{A2}	C _{B1}	C _{A2}	C _{B1}	C _{A1}	C _{A2}
Antagonism index (AI), max=40	27,5	12,0	27,5	27,0	13,0	28,0
	<i>T. harzianum</i> 11/16 TM					
<i>A.alternata</i>	C _{A1}	C _{A1}	C _{A1}	C _{A1}	C _{A1}	C _{A1}
<i>B.cinerea</i>	C _{B2}	C _{B2}	C _{B2}	C _{B2}	C _{B2}	C _{B2}
<i>D.sorokiniana</i>	B	A	B	B	A	B
<i>F.culmorum</i>	C _{A2}	E	C _{A1}	C _{B2}	E	C _{A1}
<i>F. verticillioides</i>	C _{A1}	E	C _{B1}	C _{A1}	E	C _{B1}
<i>F. solani</i>	C _{A1}	E	C _{B1}	C _{A1}	E	C _{B1}
<i>R. solani</i>	C _{A1}	C _{A1}	C _{A2}	C _{A1}	D	C _{A2}
<i>V.lateritium</i>	C _{A2}	C _{A1}	C _{A2}	C _{A2}	C _{A1}	C _{A2}
Antagonism index (AI), max=40	30.0	16.5	31.0	30.5	13.0	31.0

**Figure 3.** Antagonistic activity of (1) *T. harzianum* (BTU, Ukraine) and (2) *T. harzianum*, (China) strains against phytopathogens (on the 21st day of growth): (a), (e) *Sclerotinia sclerotiorum*; (b), (f) *Fusarium boothii*; (c), (g) *Macrophomina phaseolina*; (d), (h) *Verticillium lateritium*

for the nutrient substrate, a low intensity of mycoparasitism, which is manifested in the inhibition of the growth of *S. sclerotiorum* without significant development of hyperparasitism, and the formation of viable sclerotia.

During the action of *T. harzianum* (BTU, Ukraine) on *F. boothii*, pronounced hyperparasitism was observed on *F. boothii*, active colonization, lysis and inhibition of the growth of the pathogen's mycelium. In contrast, the Chinese-bred *T. harzianum* strain realizes its biocontrol potential mainly through competition for the substrate (due to a higher growth rate), without significant signs of direct mycoparasitic influence. The differentiation of the mechanisms of action indicates a higher aggressiveness of the domestic bioagent against the *Fusarium* spp. pathogen, which is a critical factor for the biological control of Fusarium diseases of plants in agroecosystems.

Analysis of the mycoparasitic activity of the tested strains against *M. phaseolina* indicates the dominance of the direct hyperparasitism strategy in both studied bioagents. The strain *T. harzianum* (BTU, Ukraine) showed a high level of hyperparasitic activity against *M. phaseolina*, characterized by rapid growth and complete colonization of the pathogen mycelium with subsequent suppression. However, the intensity of substrate colonization and the rate of linear growth of *T. harzianum* (China) were lower compared to the BTU isolate. The pronounced strain-specificity of the hyperparasitic activity of *T. harzianum* may determine the effectiveness of biocontrol of coal rot, suppressing of the pathogen at the early stages of its development in agroecosystems.

The study of the interspecific interaction of bioagents with *V. lateritium* revealed similar patterns. The strain *T. harzianum* (BTU)

Table 5. Antifungal activity of the biologicals Ecoster® Trichoderma (gel form) against phytopathogenic fungi

Experiment option	Growth inhibition of phytopathogens relative to control, %							
	<i>D.sorokiniana</i>	<i>F.verticillioides</i>	<i>F.sporotrichio-ides</i>	<i>R. solani</i>	<i>A.alter-nata</i>	<i>B.cinerea</i>	<i>S.sclerotiorum</i>	<i>V.late-ritium</i>
after 1 month of storage								
1:10	88	84	90	88	87	100	86	88
1:100	81	79	86	79	87	100	94	92
1:500	81	76	83	76	88	100	94	92
after 3 month of storage								
1:10	89	85	86	83	91	100	90	90
1:100	86	78	86	83	91	100	94	93
1:500	86	76	81	84	89	100	94	93

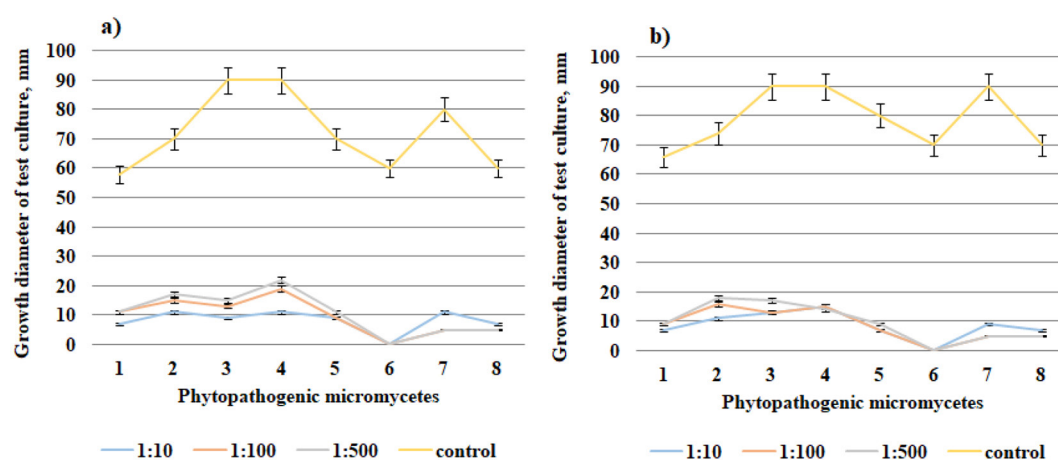


Figure 4. Antifungal effect of Ecoster® Trichoderma (in dilutions of 1:10, 1:100, 1:500) by on the growth and development of test cultures of phytopathogenic micromycetes: (a) after 1 month of storage; (b) after 3 month of storage; (1) control, (2) *D. sorokiniana*, (3) *F. verticillioides*, (4) *F. sporotrichio-ides*, (5) *R. solani*, (6) *A. alternata*, (7) *B. cinerea*, (8) *S. sclerotiorum*, (9) *V. lateritium*

demonstrated a high level of hyperparasitic activity against the pathogen, intensive growth, complete colonization of the pathogen mycelium, and subsequent suppression. At the same time, *T. harzianum* (China) was characterized by a predominantly competitive type of antagonism, without clearly expressed signs of mycoparasitism. Thus, a pronounced strain-specificity of the mechanisms of antagonistic activity was established, in particular, the BTU isolate had a higher potential as a biocontrol agent due to the dominance of direct parasitism mechanisms.

Antifungal activity of the biological Ecostern® Trichoderma (gel form) against phytopathogenic fungi, the stability of it's titre

The stability of the titer of the gel form of the biological preparation Ecostern® Trichoderma was established during 3 months of storage, which was within $1.3\text{--}3.0 \times 10^7$ CFU/cm³, as well as the preservation of high antifungal activity (on average – 79.9–100%) of the biological preparation regardless of its dilution (1:10, 1:100, 1:500) (Table 5). The preparation actively suppressed *B. cinerea* in all dilutions, *V. lateritium* and *S. sclerotiorum* in dilutions of 1:100 and 1:500 by an average of 92–100% during 3 months of storage. The biological preparation at different dilutions had almost the same effectiveness against *A. alternata* – 87–91%, and against the remaining phytopathogens, the highest activity was found at a concentration of 1:10 (85–90%).

A significant inhibition of the diameter of the colonies of test cultures was observed (Figure 4).

After one and three months of storage, Ecostern® Trichoderma completely inhibited the growth of *B. cinerea* at different concentrations, *S. sclerotiorum* and *V. lateritium* – 12–16 times at concentrations of 1:100 and 1:500 and on average 5.1–8.6 times the growth of other pathogens after a month, respectively 14–18 times after a month and 50.1–11.4 times after 3 months of storage. After a month, on average, it was 87.5%, after 3 – 88.7%.

The influence of the preparation Ecostern® Trichoderma on the soil mycobiota

Phytopathological analysis of soil samples under rapeseed in Western Ukraine revealed (Figure 5) that samples treated with the biopreparation Ecostern® Trichoderma had 5.3 times fewer pathogenic fungi compared to the control variant.

Additionally, the proportion of saprotrophic mycobiota increased to 97.9% from 88.9% in the control. Figure 6 shows that the number of fungi of the genus *Trichoderma* increased in the soil under the influence of the Ecostern® Trichoderma preparation – 44.4% versus 27.8% compared to the control variant.

Among saprotrophic fungi, species from the genus *Penicillium* (*P. simplicissimum* (Oudem.) Thom., *P. funiculosum* Thom., *P. solitum* Westling, *P. waksmanii* Zaleski, *P. brevicompactum* Dierckx, *P. canescens* Sopp); from the genus *Rhizopus* (*R. stolonifer* (Ehrenberg: Fries) Vuill.); from the genus *Cladosporium* (*C. cladosporioides*

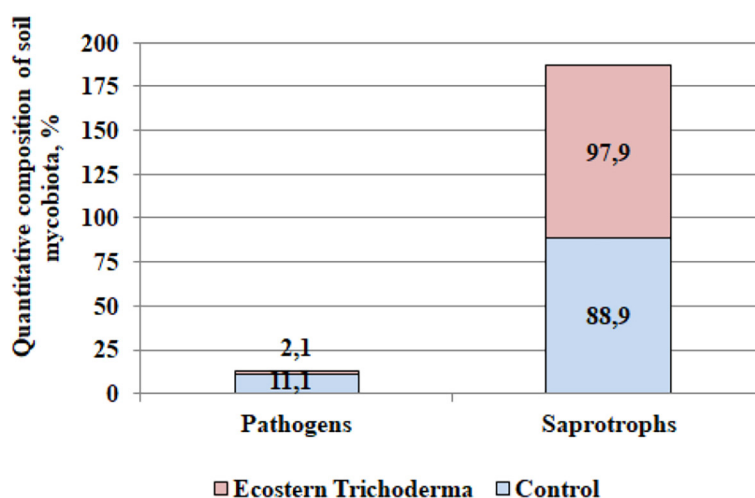


Figure 5. Quantitative composition of soil mycobiota under the influence of the preparation Ecostern® Trichoderma, %

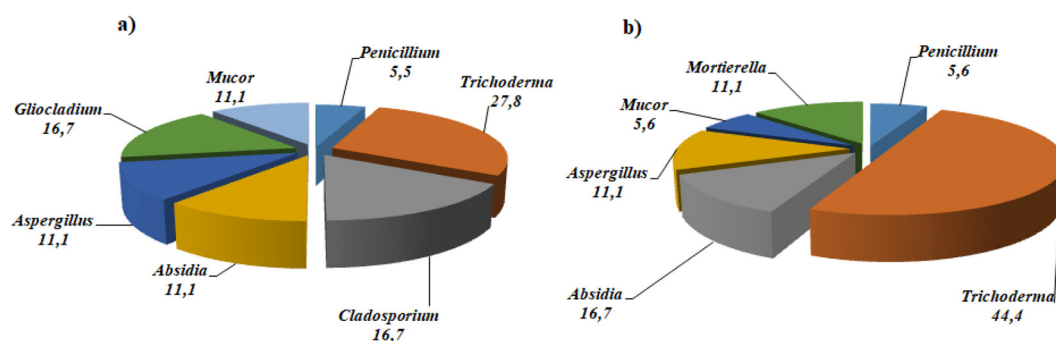


Figure 6. Genus ratio of saprotrophic mycobiota under the influence of the preparation Ecostern® *Trichoderma*, %

(Fresen.) G.A. de Vries, *C. herbarum* Fr.); from the genus *Mucor* (*M. hiemalis* Wehmer, *M. circinelloides* v. Tiegh, *M. mucedo* L.); from the genus *Absidia* (*A. butleri* Lendn., *A. glauca* Hagem.); from the genus *Gliocladium* (*G. catenulatum* J.C. Gilman & E.V. Abbott, *G. roseum* Bainier); from the genus *Mortierella* (*M. alpina* Peyronel, *M. polycephala* Coem); from the genus *Aspergillus* (*A. candidus* Link, *A. terreus* Thom); from the genus *Trichoderma* (*T. viride* Pers., *T. koningii* Oudemans, *T. harzianum* Rifai).

Consequently, our field data confirm that the strategic introduction of *Trichoderma* isolates serves as a dual-action ecological tool: it acts as a robust biocontrol agent via pathogen exclusion while simultaneously driving organic matter recycling and enhancing the systemic biological fertility of agricultural soils.

Thus, the biotechnological evaluation of strains of the genus *Trichoderma*, which are the basis of the Ecostern® line of biodestructors and soil improvement preparations, showed their high antifungal activity, stability of their viability and activity during storage. The results of our own research and analysis of scientific literature sources confirm the feasibility of their use in field conditions.

DISCUSSION

Analysis of cultural and morphological characteristics is a key stage of the study, which ensures accurate taxonomic identification and serves as the basis for targeted selection of the most active strains. The biotechnological assessment of *Trichoderma* strains requires systematic screening of their morphocultural stability and growth kinetics as precursors to efficacy testing under production conditions. In our study, phenotypic evaluation on PDA

revealed distinct macromorphological profiles and growth rates among the isolates, reflecting varying metabolic adaptations. These variations in mycelial differentiation, sporulation intensity, and pigmentation underscore the high morphogenetic plasticity of the strains, which directly influences their competitive space-occupation and overall biocontrol potential. Similarly, in a study by Surma et al. (2025), characterization of 24 apple *Trichoderma* strains revealed substantial morphocultural diversity. The isolates were divided into four groups corresponding to specific species complexes: *T. hamatum* (n = 10), *T. viride* (n = 6), *T. harzianum* (n = 4), and *T. koningiopsis* (n = 4).

The research results of Voloshchuk et al. (2024) reported that rapid growth is critical for the physical suppression of toxigenic strains, our isolates displayed marked variability in colony development on PDA. While *T. viride* 44/13 LM and *T. harzianum* 21 demonstrated rapid spatial occupation, *T. koningii* 1220-KL exhibited the slowest growth rate. This kinetic disparity suggests that these strains deploy different strategies during competitive interactions in the rhizosphere.

The morphocultural evaluation across different substrates revealed two distinct phenotypic strategies among the tested isolates. While *T. reesei* 320-TN/L demonstrated high macromorphological plasticity, both *T. harzianum* strains (23/1 and 11/16 TM) exhibited relative structural homeostasis, maintaining consistent colony growth patterns regardless of the nutrient medium used. Specifically, *T. reesei* 320-TN/L modulated its biomass allocation and colony architecture in response to nutrient availability, transitioning from a central powdery, dark green sporulation flanked by light grey-green tufts on PDA to expansive grey-green tufts on BME. Conversely, the *T. harzianum* isolates maintained a stable cottony-to-fluffy mycelial phenotype

across BME, PDA, and Čapek media. Despite this macro-morphological stability, nutrient-rich substrates—particularly BME—significantly enhanced the intensity of pale blue-green conidiogenesis in both *T. harzianum* 23/1 and 11/16 TM, confirming that complex carbohydrate and amino acid profiles stimulate reproductive differentiation. The research results of Kurchenko et al. (2021) and Valente et al. (2026) reported that substrate-specific limitations modulate strain-specific metabolic and antagonistic profiles. In our study, growth on the synthetic Čapek medium, however, was restricted by its limited nutritional content and specific C:N ratio.

In our experiments, spatial competition and direct mycoparasitism were the dominant strategies against several foliar and root pathogens. Specifically, *T. harzianum* 21 demonstrated a highly aggressive competitive phenotype, completely overgrowing *Alternaria alternata*, *Cladosporium herbarum* and *Verticillium lateritium*, and partially overgrowing *Botrytis cinerea*, *Fusarium verticillioides*, and *Fusarium culmorum* without forming inhibition zones. Similarly, *T. koningii* 1220-KL completely overgrew *V. lateritium* and *A. alternata*, and partially covered *F. verticillioides*. However, the susceptibility of *T. koningii* 1220-KL to inhibition by *F. culmorum* and *B. cinerea* (interaction types D and E) underscores that its competitive efficiency is highly pathogen-specific. Conversely, antimycotic activity via the secretion of diffusible metabolites played a critical role in interactions where growth arrest occurred prior to physical contact. This was most pronounced against *D. sorokiniana*, which was consistently restricted by all three tested *Trichoderma* strains through the formation of distinct inhibition zones. A similar mixed mechanism was observed for *T. viride* 44/13 LM, which formed clear zones of inhibition (type C_{B2}) against *A. alternata*, *C. herbarum*, and *V. lateritium*, and a combination of partial overgrowth and inhibition zones (type C_{B1}) against *F. culmorum* and *F. verticillioides*. These varying interaction profiles demonstrate that while rapid spatial occupation is a primary factor in *Trichoderma* biocontrol potential, the secretion of antifungal compounds remains a crucial prerequisite for suppressing more resilient phytopathogens. The different interaction profiles observed in our study strongly align with the biocontrol model described by Liu et al. (2024) for *Trichoderma asperellum* CMT10 against *Neopestalotiopsis clavispora* CMGF3.

These authors reported that effective pathogen suppression by *Trichoderma* spp. is mediated by a synchronized combination of competitive, antimycotic, and hyperparasitic activities (the rapid surface colonization and high inhibition rates - 65.49% after 7 days).

The observed phenotypic diversity and varying biocontrol efficacy among the tested strains are deeply rooted in their metabolic physiology. According to Mukhopadhyay and Kumar (2020) and Valente et al. (2026), the distinct behavior of *Trichoderma* spp. strains under different nutritional conditions stems from genetically determined carbon/nitrogen assimilation pathways and variations in secondary metabolism regulation, which directly dictate the intensity of antifungal compound synthesis. This explains why strains like *T. koningii* 1220-KL, despite slower radial growth, activated alternative metabolic pathways leading to pigment secretion, whereas *T. viride* 44/13 LM and *T. harzianum* 21 relied primarily on rapid morphogenetic reactions and aggressive spatial occupation.

High phenotypic aggressiveness and elevated AI scores do not automatically imply safe biocontrol efficacy, particularly when dealing with mycotoxigenic pathogens. This critical nuance is supported by Ren et al. (2025), who reported that while *Trichoderma* isolates effectively halted the growth of *Fusarium graminearum* and suppressed trichothecene (DON, 3-ADON, and 15-ADON) synthesis by 95.3–99.6%, certain volatile and non-volatile metabolites paradoxically upregulated the expression of *tri4* and *tri5* genes, thereby stimulating mycotoxigenesis. The dependency of biocontrol mechanisms on the specific target pathogen is further corroborated by Voloshchuk et al. (2024), who demonstrated that *Trichoderma* spp. suppress *Aspergillus flavus* primarily via antibiosis, whereas *Aspergillus parasiticus* is restricted via direct mycoparasitism. Furthermore, Voloshchuk et al. (2024) highlighted the role of volatile organic compounds (VOCs) and diffusible metabolites from strains like *T. asperellum* BTU in simultaneously reducing mycelial growth and suppressing aflatoxin B₁ production. Given that our highly active strains, *T. viride* 44/13 LM and *T. harzianum* 21, interacted directly with *Fusarium* spp. and other toxigenic templates, their high AI scores must be interpreted with caution. The potential for sub-lethal metabolic stress or specific chemical signaling to trigger defensive mycotoxigenesis—as observed by Ren

et al. (2025)—or to shift the dominance between antibiosis and mycoparasitism (Voloshchuk et al., 2024), underscores the critical need for additional evaluation of *Trichoderma* spp. strains regarding their potential stimulating effects on mycotoxigenesis before field deployment.

The nutritional profile of the substrate directly modulates the biocontrol efficacy and competitive success of *T. harzianum* 11/16 TM and *T. harzianum* 23/1. On nutrient-rich media (PDA and BME), both isolates demonstrated strong antifungal activity, achieving high AI scores and suppressing up to 7 out of 8 tested phytopathogens, including resilient *Fusarium* and *Rhizoctonia* species. However, on the synthetic Čapek medium, *T. harzianum* 23/1 experienced a two-fold decrease in aggressiveness, allowing faster-growing phytopathogens to outcompete and suppress it. A similar reversal of dominance was observed for strain 11/16 TM, which was actively inhibited by *F. verticillioides*, *F. solani*, *F. culmorum*, and *R. solani* under nutrient-limiting conditions. These substrate-dependent shifts in competitive dynamics closely mirror the patterns observed by Kim et al. (2019) for the *T. harzianum* complex and Iqbal et al. (2024) for *T. viride*, who noted that *Trichoderma* growth, sporulation, and overall biocontrol fitness are strictly constrained by the availability of easily assimilable nutrients. Furthermore, the metabolic stress induced by standard synthetic media can be mitigated by complex organic supplements, as evidenced by Kim et al. (2019), who reported enhanced sporulation and fitness when Čapek-Dox medium was modified with yeast extract. Our results confirm that while both *T. harzianum* strains possess high intrinsic antagonistic potential, their capacity to dominate the shared ecological niche is conditional upon substrate composition, and nutrient-limiting conditions severely compromising their competitive advantage against aggressive soil-borne pathogens.

The comparative analysis of *T. harzianum* strains from different geographic origins reveals distinct, pathogen-independent biocontrol strategies. The domestic isolate *T. harzianum* (BTU, Ukraine) consistently deployed a strategy of direct, aggressive hyperparasitism. Across tests with *S. sclerotiorum*, *F. boothii*, *M. phaseolina*, and *V. lateritium*, the BTU strain demonstrated rapid linear growth, complete mycelial colonization, and subsequent lysis of the target pathogens, notably restricting the formation

of *S. sclerotiorum* sclerotia. This destructive phenotype implies a high intensity of exocellular hydrolase secretion (Mukhopadhyay et al., 2020; Vinale et al., 2020). Conversely, the commercial Chinese strain of *T. harzianum* primarily relied on competitive exclusion (spatial competition) driven by fast initial substrate colonization, displaying significantly lower mycoparasitic activity and failing to prevent the development of viable sclerotia or to induce active lysis.

These divergent behavioral profiles underscore a pronounced strain-specificity in *Trichoderma* biocontrol mechanisms, which is regulated at the molecular level. According to Mukhopadhyay et al. (2020), the activation of core conserved signaling networks—specifically the heterotrimeric G-protein-dependent, MAP kinase, and cAMP-dependent pathways—directly governs the differentiation of infectious structures (e.g., hyphal coiling and appressoria formation) and the induction of secondary antifungal metabolites. The enhanced aggressiveness and active hyphal wrapping observed in the *T. harzianum* BTU strain suggest a more efficient upregulation of these signaling cascades in response to host-derived chemical cues. Consequently, while both strains possess biocontrol potential, the dominance of direct mycoparasitism renders the domestic BTU isolate a more formidable candidate for the early-stage suppression and long-term control of resilient soil-borne and foliar pathogens within agricultural ecosystems.

The phytopathological and structural shifts observed in the soil mycobiota under rapeseed upon application of the *Trichoderma*-based biopreparation demonstrate its high ecological adaptability and suppressive capacity, the successful colonization of the soil niche. These findings closely align with the models proposed by Ji et al. (2021) and Hang et al. (2022), where the inoculation of *Trichoderma*-based biofertilizers consistently restructured the soil microbiome by suppressing phytopathogens and promoting beneficial saprotrophic communities.

Notably, this microbial restructuring stimulated the development of the order *Mortierellales*. Representatives of this group are recognized not only as highly efficient decomposers of complex plant polymers (cellulose, hemicellulose, and lignin) but also as producers of plant growth-promoting phytohormones like indole-3-acetic acid (Ozimek et al., 2021). The stimulation of these

native functional groups explains the broader agrochemical and yield benefits observed for biodestructors within Ukrainian agroecosystems. Specifically, our results complement the data of Lungul et al. (2024), who reported that such microbial inoculants (based on increased *Trichoderma* spp.) increased labile soil carbon, enhanced the organic matter transformation coefficient, and elevated the ecophysiological diversity index, ultimately translating into increased crop yields.

In the context of the widespread trophic degradation of soil resources in Ukraine (Veremeyenko, 2019), the recycling of post-harvest crop residues via targeted biological intervention is critical. This is further supported by the work of Pavlenko et al. (2021), Dudchenko et al. (2021), and Korsun et al. (2024), who demonstrated that combining crop residues (such as straw) with aggressive, cellulolytic *Trichoderma* or multi-strain bacterial-fungal complexes optimizes soil nutrient regimes, accelerates residue mineralization, and enhances crop performance (e.g., in corn, wheat, and soybean).

Despite the high intrinsic potential of *Trichoderma*-based biocontrol agents, their field efficacy is frequently constrained by a combination of abiotic fluctuations, target-pathogen specificity, and biotic resistance from native soil microbial communities (Alfiky and Weisskopf, 2021). Furthermore, the field performance of *Trichoderma*-based inoculants is dependent on the temporal and technical parameters of colonization. Bandara and Kang (2024) reported that the biocontrol efficiency of *T. virens* was strongly modulated by the time and mode of application, noting that the highest density of viable propagules did not necessarily correlate with the most pronounced plant growth-promoting effects. Their study revealed no significant structural changes in the native rhizosphere bacterial and fungal populations regardless of the product concentration applied. This stable microbial background aligns with the findings of Rigobello et al. (2024), who observed that while combined treatments of *T. harzianum* and *Bacillus subtilis* significantly enhanced soybean growth parameters in specific application variants, they exerted no substantial influence on the root microbiome. Furthermore, recent studies highlight that substrate nutrient limitations can trigger a reversal of competitive dominance, severely compromising *Trichoderma* survival and colonization potential under such conditions (Kim et al., 2019; Valente et al., 2026). A critical

emerging limitation is the potential for sub-lethal metabolic stress induced by *Trichoderma* metabolites to paradoxically upregulate mycotoxin biosynthetic genes (such as *tri4* and *tri5*) in resilient pathogens like *Fusarium* spp., underscoring the need for rigorous strain-specific risk assessments before wide-scale agricultural deployment (Ren et al., 2025).

While acknowledging these limitations, the biotechnological evaluation of the tested *Trichoderma* strains—which form the active basis of advanced microbial biodestructors and soil amendments—demonstrated high intrinsic antifungal activity, along with robust stability of their viability and metabolic performance during storage. Taken together, our experimental findings and the analysis of current scientific literature confirm the ecological feasibility and practical efficacy of deploying these specific isolates under field conditions.

CONCLUSIONS

A comprehensive biotechnological evaluation of the strains *Trichoderma viride* 44/13 LM, *T. koningii* 1220 KL, *T. reesei* 320 TN/L and *T. harzianum* 21, 23/1, 11/16 TM was carried out, which are the basic components of biodestructors for soil improvement and restoration of the Ecoster® line of preparations.

Strain-specific differences in growth, cultural morphological characteristics and mechanisms of antagonistic action of the studied *Trichoderma* spp. isolates were established, which were manifested in the type of interaction and values of the antagonism index against phytopathogenic micromycetes. All studied *Trichoderma* spp. strains demonstrated pronounced antifungal activity against a wide range of phytopathogens, including *Cladosporium herbarum*, *Botrytis cinerea*, *Alternaria alternata*, *Drechslera sorokiniana*, *Fusarium* spp., *Rhizoctonia solani* and *Sclerotinia sclerotiorum*.

The highest antagonism index values are characteristic of strains *T. harzianum* 11/16 TM, *T. viride* 44/13 LM and *T. harzianum* 21. The maximum level of inhibition of phytopathogens was achieved on media with barley malt extract and potato glucose agar, while on Čapek medium the biological activity decreased by an average of 2.2 times, which indicates the crucial role of carbon nutrition in the formation of antifungal potential.

The dominance of mechanisms of direct mycoparasitism was revealed in the strain *T. harzianum* (BTU), which exceeded its foreign analogue in terms of biocontrol potential, in particular against *Sclerotinia sclerotiorum*, *Fusarium boothii*, *Macrophomina phaseolina* and *Verticillium lateritium*. Mycoparasitic activity was manifested by intensive colonization, lysis of pathogen mycelium and inhibition of sclerotia formation. The biopreparation Ecosterne® Trichoderma (gel form) was characterized by high technological stability: the titer of viable cells was maintained at $1.3\text{--}3.0 \times 10^7$ CFU/cm³, and the antifungal activity was 79.9–100% during 3 months of storage, regardless of dilution. Extending the shelf life of the preparation to 3 months did not lead to a decrease in its antifungal efficiency, which indicates the stability of biologically active components in gel form.

The use of the preparation in the soil ensured a 5.3-fold decrease in the number of pathogenic mycobiota and the formation of the dominance of saprotrophic mycobiota, which is a prerequisite for increasing the destruction of plant residues and restoring the biological activity of the soils.

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