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Ecological control of phytopathogenic micromycetes in agrocenoses of Ukraine

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ABSTRACT

The relevance of the problem is determined by the need for organic production of high-quality soybean seeds and increasing the level of biosecurity in agrocenoses. The purpose of the study is to determine effective strategies for controlling the mycelial structures of necrotrophic micromycetes in agrocenoses of soybean varieties Suzir'ya, Vyshyvanka and Kent during the growing season of plants. The work used greenhouse, field and laboratory methods of agroecosystem biocontrol, as well as analytical, comparative and experimental methods. The prevalence and frequency of occurrence of phytopathogenic micromycetes – pathogens of diseases of seeds, vegetative organs and the rhizosphere of soybean plants were determined. The effect of the biological preparations Mycohelp and Phytohelp on the main ecological and trophic groups of microorganisms in the soybean rhizosphere has been established. It has been established that the antifungal activity of exometabolites of soybean plants depends on the bacterial signaling that underlies the biological preparations Phytocid, Mycohelp and Phytohelp. This affects the activity of root exometabolites that control the population of phytopathogenic micromycetes. Soybean plants of the Vyshyvanka, Suzir'ya and Kent varieties, interacting with microorganisms that are part of the biological preparations Mycohelp and Phytocid, produce root exometabolites that differ significantly in their effect on the radial growth rate of the mycelium of the phytopathogenic micromycetes. Root exometabolites of the Suzir'ya and Vyshyvanka soybean varieties exhibit antifungal properties. They are able to inhibit the growth and development of the mycelium *F. solani* and *F. keratoplasticum*. The antifungal effect of plant exometabolites is significantly enhanced when they interact with biological products. This effect is controlled by the antifungal activity of soybean root exometabolites, which depends on the signaling of bacteria that form the basis of biological products. The results obtained make it possible to reduce the number of chemical protection agents in agrocenoses, thereby contributing to an increase in biodiversity, restoration of microbial balance in agrophytocenoses, reduction of cost and improvement of quality of plant raw materials.

Keywords: necrotrophic micromycetes, exometabolites, biofungicides, agrocenoses, mycelial structures, biocontrol, organic production, soybean cultivars.

INTRODUCTION

One of the main crops of the *Fabaceae* family grown in Ukraine is soybean (*Glycine max* Moench.). Among organic producers, more than 4.6% are engaged in growing legumes (Revtyo and Zolin, 2023). There has been a significant increase in the area under cultivation and production volumes of soybeans, and the issue of organic production, and therefore the production of high-quality seeds, is becoming increasingly relevant. Official IFOAM statistical reviews confirm that in 2022 there were about 462 organic farms in Ukraine, and the total area of agricultural land used for

organic production was 263,619 hectares. According to a study of the organic market in 2022 conducted by the Information Center Green Dossier (OrganicInfo.ua), Organic Standard, and the Research Institute of Organic Agriculture (FiBL), Ukraine ranks third (out of 125 countries) in terms of organic product exports to the EU.

Under conditions of anthropogenic stress, namely the irrational use of chemical pesticides, the spread of pathogenic microfungi is accelerating, and resistant forms are forming with increased aggressiveness, which can lead to a loss of disease resistance in plant varieties, including soybeans. Therefore, more and more attention is being paid to organic soybean production, which is based on the regulation of phytopathogenic microfungi in agrocenoses using disease-resistant varieties and biological preparations (Havryliuk et al., 2019; Parfenuk et al., 2021). The effectiveness of such regulation depends on understanding the mechanisms and factors that determine the rate of formation of natural ecotypes of parasitic fungi. After all, the simplification of many agrocenoses affects their optimal functioning and stability, leading to an increase in the level of biological pollution of agroecosystems (Zaimenko et al., 2020). Thus, stimulation of phenotypic variability of phytopathogenic microfungi due to contact with disease-resistant varieties of cultivated plants, especially those created by genetic modification and with highly disease-resistant genotypes, contributes to increased pathogenicity and phytotoxicity of the mycobiome (Turovnik et al., 2020).

In recent years, the number of phytopathogenic microfungi interacting with plants during the growing season has been increasing in soybean agrocenoses. They cause outbreaks of plant diseases, which lead to significant grain yield losses and deteriorate its quality. This increases the biological pollution of agroecosystems and leads to a decrease in the quality and environmental safety of plant products (Drebot and Parfeniuk (Eds.) 2022). At the same time, questions regarding the ecological justification of the allelopathic effect of soybean metabolites as a factor regulating the number of phytopathogenic microfungi-causative agents of plant diseases G. max-remain poorly studied in Ukraine.

The aim of the study was to investigate the exometabolites of soybean varieties Suzirya, Vyshyvanka, and Kent as a factor controlling the mycelial structures of necrotrophic microfungi in plant agrocenoses.

MATERIALS AND METHODS

The objects of the study were exometabolites isolated from the Suziya and Vyshyvanka soybean varieties, bred by the National Scientific Center of the Institute of Agriculture of the National Academy of Agrarian Sciences of Ukraine, and Kent, bred by a Canadian company SAAT-BAULINZ (Austria), biological preparations Fitohelp® (BTU Biotech Company, based on *B. subtilis*, cell titer 4.0 x 10^9 CFU/ml), Mycohelp® (BTU Biotech Company, based on genera and species *Trichoderma, B. subtillis, Azothobacter, Enterococcus, Enterobacter,* biologically active products of the vital activity of microorganisms-producers, total number of viable cells 1.0×10^9 CFU/ml), phytopathogenic fungi *F. solani* and *F. keratoplasticum.* The biological product Fitocid® was used as a comparison standard (BTU Biotech Company, based on *B. subtilis,* total number of viable cells 1.0×10^9 CFU/ml).

During 2018–2023, samples of soybean varieties grown using organic technology in the Central Forest-Steppe zone of Ukraine were analyzed at the experimental fields of the Skvyrska Research Station organic farming of the Institute of Agroecology and Environmental Management of the National Academy of Agrarian Sciences of Ukraine (IAEM of NAAS). Under the conditions of a vegetation experiment in the laboratory of biocontrol of agroecosystems and organic production of the Department of Agrobiological Resources and Environmentally Safe Technologies of the IAEM of NAAS, laboratory studies were conducted to study the spread and frequency of occurrence of phytopathogenic microfungi on seeds, vegetative organs, and rhizosphere of plants, obtaining root exometabolites of the Suziya, Vyshyvanka, and Kent soybean varieties, and determining their effect on the physiological and biological activity of fungi of the genus *Fusarium*.

The spread and frequency of occurrence of phytopathogenic microfungi were studied on seeds, vegetative organs, and rhizosphere of Kent and Suzirya soybean varieties. To analyze the mycobiota, a biological method for determining the quality of agricultural crop seeds was used. Fifty seeds were selected from soybean plants of the corresponding variety, placed in gauze bags, washed with running water, and sterilized with a 0.5% $KMnO_4$ solution. After that, the seeds were washed in sterile H_2O and dried. The seeds were placed in Petri dishes on Chapek's medium and

germinated in a thermostat at a temperature of 24–25 °C for 7 days. The identification of microfungi species was carried out by microscopy based on the morphology of conidia. The species affiliation of isolates was determined by the morphology of conidia and the cultural and morphological properties of colonies grown on Chapek medium according to the methods (Zaimenko et al., 2014).

Species identification of *Fusarium* spp. was performed using the sequencing method. For this purpose, total DNA was isolated and the ITS sequence fragment was amplified with universal primers. The resulting PCR product was purified and sequenced. For ITS sequence analysis, isolates were grown on PDA nutrient medium. Genomic DNA was isolated using the GeneJet Genomic DNA Purification Kit (ThermoScientific) according to the manufacturer's instructions. Amplification of the 26S rRNA or 18S rRNA gene was performed with ITS1 primers (5'-TCCGTAGGTGAACCTGCGG-3′) ITS4 (5'- TCCTCCGCTTATTGATATGC-3'). PCR was performed on an amplifier Mastercycler Personal 5332 (Eppendorf, Germany). PCR products were separated in 1.7% agarose gel containing 0.01% ethidium bromide. The results were visualized under UV light (Dayarathne et al., 2020). The dendrogram of genetic similarity between the studied strains and other strains of the *Fusarium* genus was constructed using the MEGA 6.0 program (nearest neighbor method, Kimura's 2-parameter model).

Previously isolated exometabolites of soybean plants were added to the Chapek medium at a concentration of 1:4 to study the effect of exometabolites of soybean varieties Suzirya, Vyshyvanka, and Kent on the growth of phytopathogenic fungi (Turovnik et al., 2020).

50 seeds of each variety were selected and sterilized to obtain root exometabolites of Kent, Vyshyvanka, and Suzirya soybean varieties in accordance with DSTU 4138:2002 (2002). The seeds were placed in humid chambers and kept there for 3–5 days until sprouts 2–3 cm long formed. (Parfeniuk et al., 2022). Ten seedlings of each variety were placed in sterile dishes with sterile distilled water and kept for 72 hours in diffused light at a temperature of 22–24 °C to extract root exometabolites. The exudates were washed from the roots and filtered through a microporous bacterial filter (0.02 μm). The obtained exometabolites were used for further research.

Seeds were sown in a universal soil mixture to obtain root exometabolites of soybean plants after interaction with biological preparations. The plants were grown at a temperature of 25–28 °C for 2 days. On the third day, the plants were transferred to a light room. In the phase of the formation of the first trifoliate leaf, the roots of soybean plants were washed from the soil and placed in sterile distilled water to extract exometabolites. After 72 hours, an average sample was obtained from the allocated solutions, which was filtered through a CROMPURE NY 0.22 filter and frozen.

A disc with *F. solani* and *F. keratoplasticum* mycelium (d=0.8 cm) was placed in the center of each Petri dish on the surface of the agarized Chapek medium to determine the radial growth rate under the action of root exometabolites (Patent No. 92066). 3 sterile paper discs place around the perimeter. The solution of the obtained exometabolites was applied to each paper disc. The micromycetes cultivated at a temperature of 24 °C. The radius of the colonies measured in two mutually perpendicular directions every 24 hours. (Turovnik et al., 2020). The radial growth rate of fungus cultures was calculated using the following formula (1):

$$V_r = (r_1 - r_0)\,(t_1 - t_0) \quad (1)$$

where: V_r – radial colony growth rate; r_0 – radius of colonies at the moment t_0; r_1 – radius of colonies at the moment t_1.

For statistical processing of experimental data, statistical and correlation methods of mathematical statistics were used with the application of Microsoft Excel software. To determine the effect of root exometabolites on the growth and development of the isolate, the Newman- Keuls criterion was applied.

RESULTS AND DISCUSSION

Soybean plants interact with pathogens of a number of diseases that affect seeds and vegetative organs of plants during the growing season. This causes significant losses in quality and quantity of the harvest. The most common and harmful diseases of fungal etiology during the growing season of *G. max* plants include: Alternaria blight (*Alternaria alternata*), downy mildew (*Peronospora manshurica*), Septoria leaf spot (*Septoria glycines*), fusarium wilt (*Fusarium* spp.), ascochyta blight (*Ascochyta sojaecola*), anthracnose (*Colletotrichum truncatum*) and cercosporosis (*Cercospora kikuchii*) (Tan and Wang, 2024).

Based on the results of the analysis of the nuclear ribosomal intragenomic spacer (ITS) on soybean seeds, 148 isolates belonging to 13 genera of fungi were identified, among which representatives of the genus Fusarium accounted for 55.0% (Marquez et al., 2019).

It should be noted that the microbiome associated with plants is called the second genome and is crucial in stimulating plant growth and disease resistance (de Vries et al., 2020; Trivedi et al., 2020). These agronomically valuable microorganisms have formed a multifunctional "holobiont" with plants during coevolution, and their colonization of cultivated plants is an important factor in the formation of plant resistance to diseases and pests, which significantly increases the efficiency of organic plant production (Vandenkoornhuyse et al., 2015).

Our own research has demonstrated differences between the phytopathogenic microbiomes of cotyledons, roots, and leaves of Kent and Suziya soybean varieties grown in the Central Forest-Steppe of Ukraine under organic production conditions (Beznosko et al., 2021). Understanding the regulatory processes in the "plant-host-pathogen" interaction reveals ways to create a knowledge base on the dynamics of infectious material accumulation in soybean agrocenoses.

Fungi of the genus *Fusarium* is dominated in the population of micromycete species that inhabit the phyllosphere and rhizosphere of Kent soybean plants. In the mycobiome of Kent soybean seeds, the most common species are micromycetes: *Fusarium solani* (Mart.) Sacc. (57.7%), *Ascochyta sojaecola* Abr (23.1%) and *Peronospora manshurica* (Naumov) Syd. (19.2%) (Figure 1).

The following species: *F. solani* (75.0%), *A. sojaecola* (10.0%), and *P. manshurica* (15.0%) are dominated in the mycobiome of vegetative organs of soybean plants. The rhizosphere mycobiome is significantly dominated by isolates of the species *F. solani*. Their frequency of occurrence reaches 94.1%. The other dominant species is *P. manshurica* (5.9%). The number of isolates of this species also dominates on seeds and vegetative organs (in 3.2 and 2.5 times, respectively). This is explained by the biological characteristics of the development of the microfungus, namely its spread by means of spores that penetrate the plant through stomata or the cuticle, forming hyphae and haustoria in the tissues (Hnatiuk et al., 2019).

In the microbiome of Suziya soybean seeds, the most common species is the fungus *Aspergillus alliaceus* (73.4%). At the same time, the fungi *A. sojaecola* and *F. solani* had the same prevalence spread, which was 13.3%. (Figure 2).

The population of microfungi inhabiting the rhizosphere and vegetative organs of Suziya soybean plants is also dominated by species of the genus *Fusarium*. The rhizosphere mycobiome of

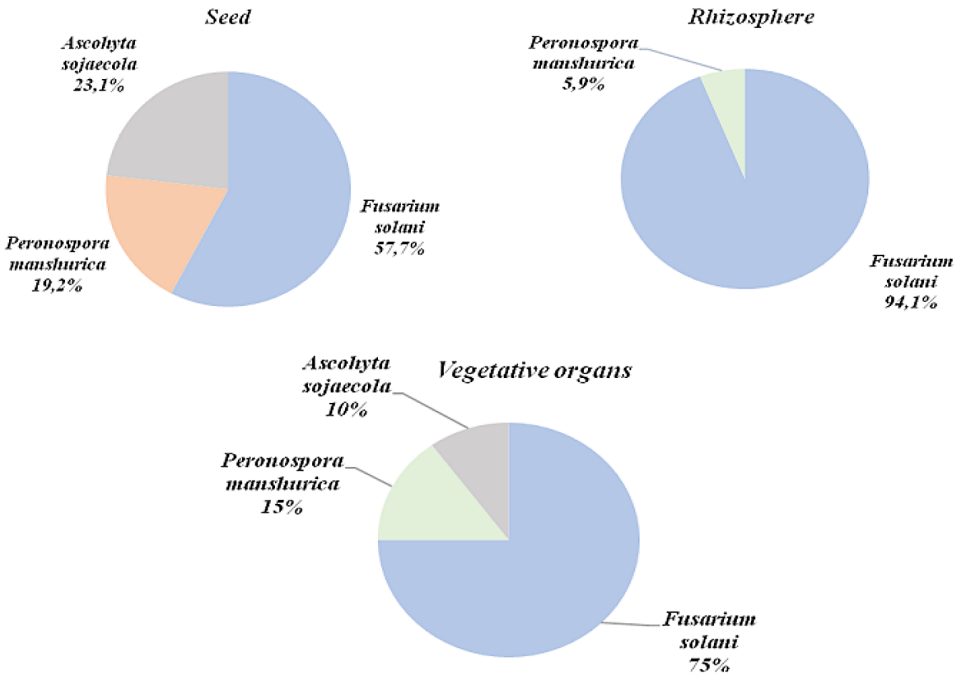

Figure 1. Frequency of occurrence of dominant micromycetes on Kent soybean plants (2020–2022)

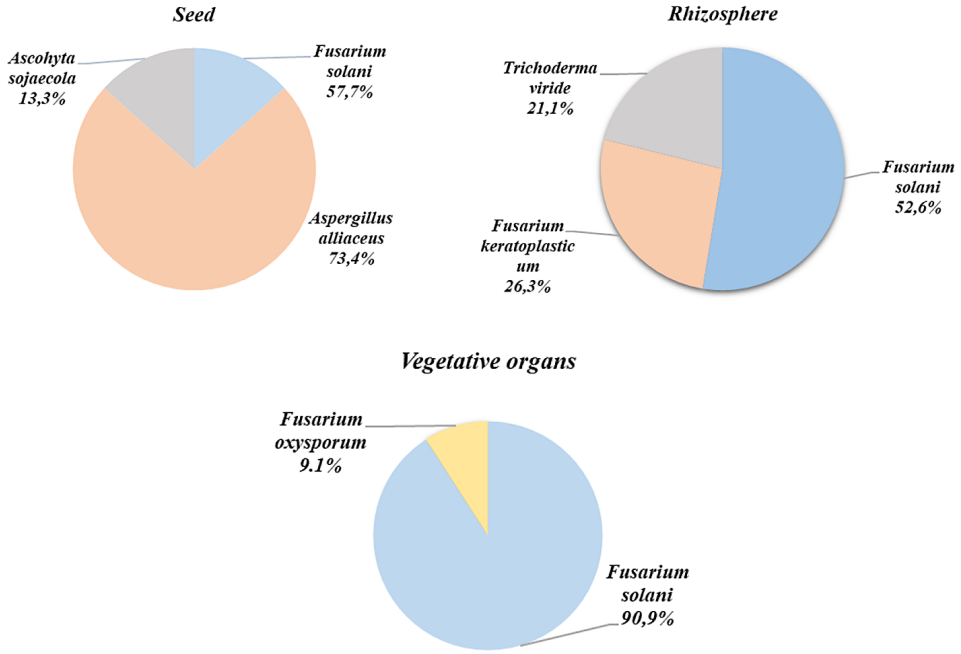

Figure 2. Frequency of occurrence of dominant microfungi on soybean plants of the Suzir'ya variety (2020–2022)

plants is significantly dominated by isolates of the species *F. solani* (52.6%) and *F. keratoplasticum* (26.3%). The frequency of their co-occurrence reached 78.9%. The mycobiome of the vegetative organs contained isolates of the species *F. solani* (90.9%) and *F. oxysporum* (9.1%). Plant exometabolites are part of their defense system against biotic factors. Above-ground signaling interactions can be controlled by volatile exometabolites, which include ethylene, methyl jasmonate and salicylate, indole, and several volatile terpenes, which are largely responsible for above-ground signaling interactions between plants. At the beginning of flowering, plants have a large amount of proteins associated with their defense against phytopathogenic microorganisms. Volatile antimicrobial compounds play an important role in the interaction of plants with the microbiome.

Root exudates form a protective complex of chemical compounds in the microbiome. It is known that both low- and high-molecular-weight metabolites, which are components of root exudates, contribute to the protection of plants from soil phytopathogenic microorganisms. Accordingly, for example, overexpression of the *Arabidopsis* spp. gene that regulates the biosynthesis of camalexin and salicylic acid (SA) increases the resistance of soybean plants to nematodes. Activation of these resistance genes as a result of infection activates internal synthesis, accumulation, and secretion of camalexin. At the same time, depression of these genes leads to suppression of camalexin formation and increased damage to plants by pathogens. Strigolactones, present in the rhizosphere during infection by root parasitic plants, participate in the symbiosis of plants with arbuscular mycorrhizal fungi. One of these effects is achieved by interfering with hormonal defense pathways, thereby promoting stress responses induced by plants in the rhizosphere. This is also evidenced by the suppression of the growth of many phytopathogenic fungi in the presence of the synthetic analogue of strigolactone GR24 (Rasmann et al., 2017).

It is known that root exometabolites, such as low-molecular-weight antimicrobial compounds like phytoanticypins and phytoalexins, are important for plant growth, development, and protection against disease (Amb and Ahluwalia, 2016). Phytoalexins protect plant root cells from colonization by pathogenic microorganisms. The spectrum of root exometabolite compounds includes a large number of antimicrobial compounds. For example, rosmarinic acid in the root exudate of sweet basil has an antifungal effect against *Phytophthora cinnamoni*. At the same time, *Lithospermum erythrorhizon* produces pigmented naphthoquinones, which are also

involved in protecting the rhizosphere of plants from pathogenic microorganisms. Colonization of plant root surfaces by motile zoospores of oomycetes, plant pathogens, occurs through the mechanism of electrogenic transport of ions on their surface (Wu al., 2023).

Thus, the frequency of phytopathogenic microfungi in the phyllosphere and rhizosphere of soybean plants can reach 95%. This indicates a high degree of potential ecological risk in the soybean agrocenosis throughout the entire vegetation period. It should be noted that among phytopathogenic microfungi, fungi of the genus *Fusarium* dominate, among which *F. solani* and *F. keratoplasticum* occupy a leading position. Therefore, these species were selected for genetic identification of fungi and study of their interaction with *G. max* plants.

For genetic identification of *Fusarium fungi*, isolates FS1 and FK1 were selected, isolated from the rhizosphere of soybean plants of the Suziya variety. As a result of sequencing, fragments of the ITS sequence with a size of 525 nucleotides were obtained. Comparative analysis of these fragments with sequences in the GenBank database showed that the highest percentage of similarity of isolate FK1 is observed with the typical strain of the species *F. keratoplasticum* and amounts to 99.8%. In the case of strain FS1-KPZB-21, the highest similarity of nucleotide sequences was observed with strains of the species *F. solani*: 98–99.2% (Figure 3).

On the constructed dendrograms of genetic similarity, the studied isolates formed one group with strains of the corresponding species, namely isolate FK1 with *F. keratoplasticum* FRC S-2477, and isolate FS1 with *F. solani* CBS 140079, which confirms the species affiliation of these isolates. Thus, the dominant morphotypes were identified as *F. keratoplasticum* and *F. solani*. Genetic analysis has confirmed the dominance of F. keratoplasticum and F. solani, which infect soybean plants of the Suzirya variety grown at the Skvyrska Research Station organic farming of the IAEM of NAAS. The strains of these species were used to determine the characteristics of the interaction between soybean plants and phytopathogenic microfungi.

The results of the studies have shown that root exometabolites of Kent and Suziya soybean varieties differ significantly in their effect on the rate of radial growth of *F. solani* mycelium (RRGM). The highest antifungal activity is exhibited by plant exometabolites of the Suzirya variety, grown under the influence of the biological products Fitohelp and Mycohelp, which inhibited growth by 0.488-0.458 mm/hour (in the control variant, without the products, growth was 0.512 mm/hour) (Table 1, Figure 4).

It should be noted that exometabolites of Kent variety plants grown with the use of the biological preparations Fitocide and Mycohelp stimulated *F. solani* RRGM (0.499 and 0.513 mm/h, respectively) compared to the control (0.442 mm/h).

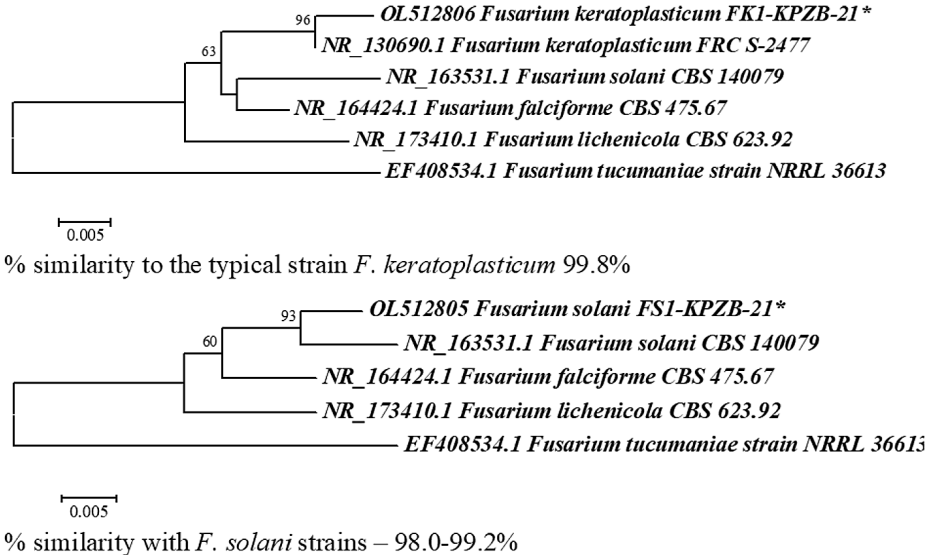

Figure 3. Dendrogram of phylogenetic interaction between strains *F. solani* CBS 140079, *F. keratoplasticum* FRC S-2477, and typical strains of the genus Fusarium, constructed on the basis of ITS sequences using the Neighbour-Joining method and Kimura's 2-parameter model (the strains under study are marked with *)

Table 1. Diameter of *F. solani* mycelium colonies under the influence of root exometabolites of Kent and Suzira soybean varieties

Option	Growth zone (d, mm)	
	24 hour	48 hour
Control (without exometabolites)	10.7±1.09	23.0±1.58
Exometabolites of the Kent variety		
Control (without biopreparations)	9.7±0.54	20.3±1.65
Fitocid	7.7±0.91	22.1±1.71
Fitohelp	11.0±1.06	21.3±1.08
Mycohelp	10.7±1.71	23.0±1.87
Exometabolites of the Suziya variety		
Control (without biopreparations)	9.3±0.22	22.0±1.19
Fitocid	11.7±1.27	24.0±1.82
Fitohelp	12.3±1.66	24.0±1.49
Mycohelp	11.7±1.28	22.7±1.46

Under the action of Fitohelp (based on *B. subtilis*, cell titer 4.0 x 10^9 CFU/ml), inhibition of *F. solani* RRGM (0.429 mm/hour) was observed. At the same time, under the action of Mycohelp, *F. solani* RRGM was at the control level. Thus, the difference in the action of exometabolites of the Canadian Kent variety and the domestic Suzirya variety can be explained by the manifestations of adaptive resistance of varieties to indigenous strains of *F. solani*.

It is known that the total amount of root exudates can reach 5–10% of the mass of the entire plant organism. According to the literature, during the analysis of root secretions of various legume species grown in vegetative or laboratory conditions, organic acids (oxalic, tartaric, citric, malonic, malic), carbohydrates (glucose, raffinose, sucrose, arabinose, rhamnose), enzymes (catalase, tyrosinase, phenolase, asparaginase, urease, invertase, amylase, cellulase, protease, lipase, phosphatase), amino acids (serine, glycine, alanine, aspartic and glutamic acids, leucine, threonine, valine, tryptophan), amides (asparagine and glutamine) (Yusnawan et al., 2019). In addition, various vitamins have been found in plant root exometabolites, including biotin, thiamine (B1), pantothenic and nicotinic acids, riboflavin (B2), and pyridoxine (B6) (Tan et al., 2013).

The use of biological products is gradually becoming an alternative strategy for protecting plants against soybean root rot. Han et al. (2021) isolated *B. subtilis* HSY21 from soybean rhizosphere soil, which had a level of inhibition of *F. oxysporum* fungus development: in laboratory conditions - 81.30 ± 0.15% ($P < 0.05$), and in greenhouse and field conditions - 63.83% and 57.07% ($P < 0.05$), respectively. RNA-seq analysis of *F. oxysporum* after treatment with strain HSY21 revealed a decrease in the expression of 1.445 genes and an increase in 1.561 genes involved in mycelium growth, metabolism regulation, and enzymes associated with disease-causing ability. The activity of cellulase, β-glucosidase, α-amylase, and pectin methyl-galacturonase, as well as the levels of oxalic acid and ergosterol in *F. oxysporum*,

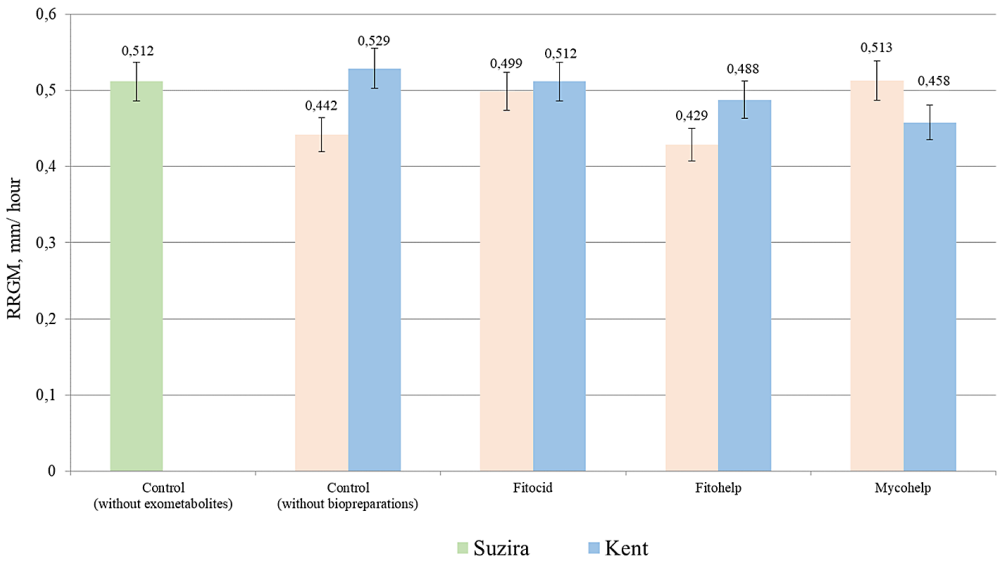

Figure 4. The rate of radial growth of *F. solani* under the influence of root exometabolites of Kent and Suzir'ya soybean varieties

were significantly reduced after treatment with HSY21. Thus, the differentiation in the inhibition or suppression of pathogens by root exometabolites of soybean varieties against the background of biological products is associated with plant metabolism and the composition of exudates. The results of the studies have proven that biological products affect the metabolism of soybean plants. Root exometabolites of plants, when interacting with biological products, slightly inhibited the rate of radial growth of *F. keratoplasticum* fungal mycelium. As shown in Figure 5, the lowest RRGM *F. keratoplasticum* was in the variant of treating soybean plants of the Suzirya variety with the Mycohep biological product and amounted to 0.0212 mm/year, which is 1.2 times less than in the control variant (without the product) – 0.0255 mm/year.

Under the action of the biological product Fitocide, colony growth was within the range of 0.0214–0.0224 mm/hour, and under the action of the product Fitohelp, it was 0.0236–0.0241 mm/hour. In the control variant (without exometabolites), this indicator was 0.0247 mm/hour. The most active inhibition of RRGM was observed in the root exometabolites of Vyshyvanka soybean plants grown under the action of the biological product Mycohelp – 0.0243 mm/hour. This may be due to the action of metabolites of *Trichoderma* fungi, which are part of Mycohelp. Thus, Xu et al. (Xu et al., 2022) claim that *T. harzianum* induces protective reactions in soybean root cells, increasing resistance to fusarium wilt. According to the authors, treatment of soybean seeds with *T. harzianum* TN-35 significantly reduces the development of fusarium wilt caused by *F. Oxysporum*. In variants with other biological products, RRGM in both soybean varieties ranged from 0.0226 to 0.0236 mm/hour. The highest RRGM values for the *F. keratoplasticum* isolate were observed in the control variants (without exometabolites – 0.0258 mm/year) and with exometabolites of varieties (without biopreparations 0.0248-0.0256 mm/hour) (Figure 6).

Due to the action of microorganisms that are components of biopreparations and as a result of interaction with phytopathogenic microfungi, substances are synthesized in soybean plants that can actively participate in plant defense mechanisms. For example, ABC transporter proteins, which bind ATP and are located in root cells, are involved in the extracellular secretion of proteins that are components of root exometabolites. In turn, inhibition of ABC transporters leads to increased susceptibility of roots to pathogenic microorganisms. This is due to reduced secretion of antifungal compounds such as diterpene sklareol (Jiao et al., 2017). It has been found that biotic stress in *Medicago truncatula* plants inhibits ABC transporter synthesis, which reduces medicarpin synthesis in the phenylpropanoid pathway. This leads to an increase in isoflavonoid levels in plants. ABC transporters modulate the synthesis and exudation of

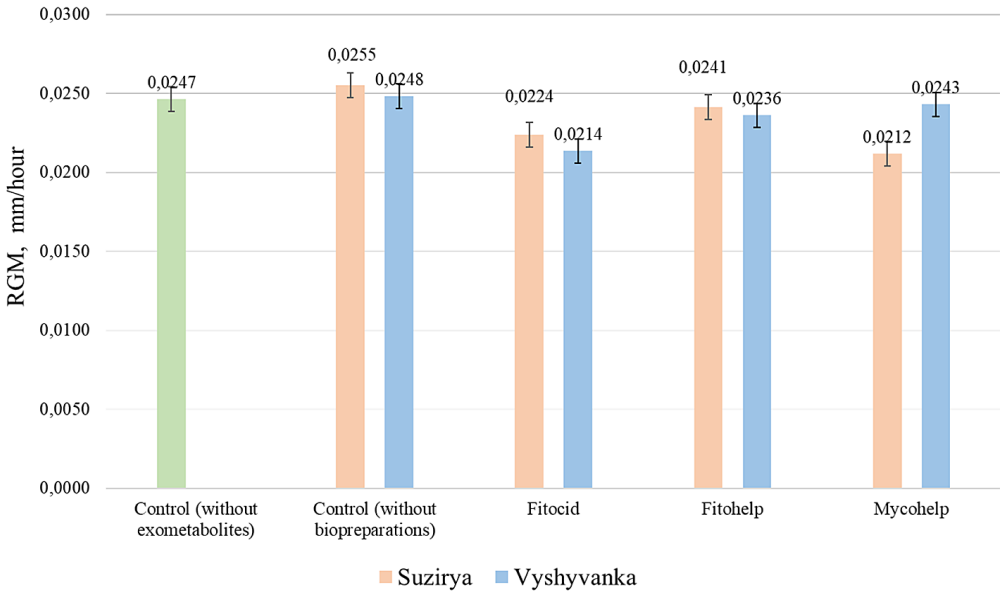

Figure 5. Radial growth rate of *F. keratoplasticum* isolate under the influence of root exometabolites of soybean plants grown under the influence of biological preparations (4 days of cultivation)

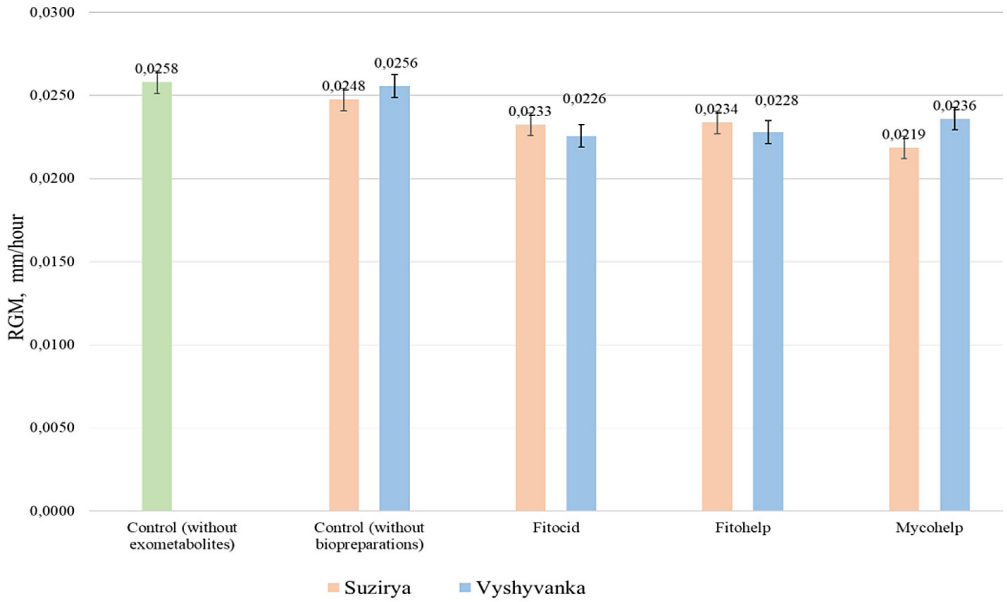

Figure 6. Radial growth rate of *F. keratoplasticum* isolate under the influence of root exometabolites of soybeans grown under the influence of biopreparations (on the 6th day of cultivation)

protective phytochemicals, which can be modified by the microbiome. Thus, a decrease in the expression of the MtABCG10 gene, which encodes the ABC protein in *M. truncatula*, leads to an increase in root infection by the fungus *F. oxysporum* (Jiao et al., 2017).

The antifungal activity of soybean plant exometabolites probably depends on the signaling of bacteria, which are the basis of the biological products Fitocide, Mycohelp, and Fitohelp. According to literature sources, metabolites of *B. subtilis* bacteria produce lipopeptides with antifungal activity (surfactins, iturins, fungicins, and other compounds), which trigger the mechanism of acquired induced resistance in plants. This, in turn, affects the activity of plant root exometabolites.

CONCLUSIONS

The results of the conducted research indicate that fungal diseases dominate the soybean (*G. max*) agrocoenoses throughout the growing season in the Central Forest-Steppe of Ukraine. These include: Alternaria leaf spot (*Alternaria alternata*), downy mildew (*Peronospora manshurica*), Septoria brown spot (*Septoria glycines*), Fusarium wilt (*Fusarium* spp.), Ascochyta blight (*Ascochyta sojaecola*), anthracnose (*Colletotrichum truncatum*), and Cercospora leaf spot (*Cercospora kikuchii*).

Soybean varieties (Vyshyvanka, Suzir'ya, and Kent), when interacting with the microorganisms contained in the biopreparations MycoHelp and Phytocid, produce root exudates (exometabolites). These exudates differ significantly in their antifungal activity, which is likely dependent on the signaling mechanisms of the bacteria that form the basis of the biopreparations (Phytocid, MycoHelp, and PhytoHelp). The mycobiome of *G. max* plants in the Central Forest-Steppe of Ukraine is dominated by fungi of the genus *Fusarium*, with *F. solani* and *F. keratoplasticum* being the most prominent species. The root exudates produced by soybean varieties Vyshyvanka, Suzir'ya, and Kent – both with and without interaction with the microorganisms from the biopreparations MycoHelp and Phytocid – exhibit distinct differences in their influence on the radial growth rate of phytopathogenic fungal mycelium.

Root exudates from the Suzir'ya and Vyshyvanka soybean varieties, regardless of biopreparation application, possess inherent antifungal properties. They are capable of inhibiting the growth and development of *F. solani* and *F. keratoplasticum* mycelia. Crucially, when interacting with the biopreparations, the antifungal effect of the exudates significantly increases and is controlled by the antifungal activity of the root exometabolites, which depends on the bacterial signaling embedded in the biopreparations.

REFERENCES

1. Amb, M. K., Ahluwalia, A. S. (2016). Allelopathy: potential role to achieve new milestones in rice cultivation. *Rice Science*, *23*(4), 165–183. https://doi.org/10.1016/j.rsci.2016.06.001
2. Beznosko, I. V., Horgan, T. M., Havryliuk, L. V. Ya., Turovnik, Yu. A., Kosovska, N. A. (2021). Pathogenic mycobiome of seeds of cultivated plant varieties. *Agroecological Journal*, (1), 81–87 (in Ukrainian). https://doi.org/10.33730/2077-4893.1.2021.227242
3. Dayarathne, M. C., Mridha, A. U., Wang, Y. (2020). Diagnosis of fungal plant pathogens using conventional and molecular approaches. In *Diagnostics of plant diseases*. IntechOpen. https://doi.org/10.5772/intechopen.94980
4. De Vries, F. T., Griffiths, R. I., Knight, C. G., Nicolitch, O., Williams, A. (2020). Harnessing rhizosphere microbiomes for drought-resilient crop production. *Science*, *368*(6488), 270–274. https://doi.org/10.1126/science.aaz5192
5. Drebot, O. I., Parfeniuk, A. I. (Eds.). (2022). *Ecological and biological safety of Ukraine: Collective monograph*. Kyiv: NULES of Ukraine Publishing House. (in Ukrainian).
6. DSTU 4138:2002. Seeds of agricultural crops: Methods for quality determination. [In force since 2004-01-01]. Kyiv: Derzhstandart of Ukraine, 2002. Official web-site (in Ukrainian). URL: https://online.budstandart.com/ua/catalog/doc-page.html?id_doc=91465
7. Han, S., Chen, J., Zhao, Y., Cai, H., Guo, C. (2021). *Bacillus subtilis* HSY21 can reduce soybean root rot and inhibit the expression of genes related to the pathogenicity of *Fusarium oxysporum*. *Pesticide biochemistry and physiology*, *178*, 104916. https://doi.org/10.1016/j.pestbp.2021.104916
8. Havryliuk, L. V. Ya., Kosovska, N. A., Parfeniuk, A. I., Mostoviak, I. I. (2019). The effect of exometabolites of plants of different soybean cultivars on the radial growth rate of *Fusarium graminearum*. *Agroecological Journal*, (4), 55–59 (in Ukrainian). https://doi.org/10.33730/2077-4893.4.2019.189454
9. Hnatiuk, T. T., Zhytkevych, N. V., Petrychenko, V. F., Kalinichenko, A. V., Patyka, V. P. (2019). Diseases of soybean under the influence of phytopathogenic bacteria of the genus *Pseudomonas*. *Microbiological Journal*, *81*(3), 68–83 (in Ukrainian). https://doi.org/10.15407/microbiolj81.03.068
10. Jiao, Y., Wang, E., Chen, W., Smith, D. L. (2017). Complex interactions in legume/cereal intercropping system: role of root exudates in root-to-root communication. *BioRXiv*, 097584. https://doi.org/10.1101/097584
11. Marquez, N., Giachero, M. L., Gallou, A., Debat, H. J., Declerck, S., Ducasse, D. A. (2019). Transcriptome analysis of mycorrhizal and nonmycorrhizal soybean plantlets upon infection with Fusarium virguliforme, one causal agent of sudden death syndrome. *Plant Pathology*, 68(3), 470–480. https://doi.org/10.1111/ppa.12964
12. Parfeniuk, A. I., Horgan, T. M., Sahanovska, V. I., Horgan, N. A. (2014). *Method for determining the effect of volatile fractions of phytoncides of onion cultivars on micromycete spores* (Ukraine Patent No. 92066). Institute of Agroecology and Environmental Management NAAS (in Ukrainian).
13. Parfeniuk, A. I., Kosovska, N. A., Borodai, V. V., Turovnik, Yu. A. (2022). Root exometabolites as an environmental factor in the interaction of cultivated plants with soil microorganisms. *Agroecological Journal*, (3), 62–74 (in Ukrainian). https://doi.org/10.33730/2077-4893.3.2022.266410
14. Parfenuk, A. I., Havryliuk, L. V., Beznosko, I. V., Pasichnik, L. P., Turovnik, Y. A., Ternovyi, Y. V. (2021). Influence of Filazonit biopreparation on soybean seed quality. *Ukrainian Journal of Ecology*, *11*(3), 86–89. https://doi.org/10.15421/2021_148
15. Rasmann, S., Bennett, A., Biere, A., Karley, A., Guerrieri, E. (2017). Root symbionts: Powerful drivers of plant above-and belowground indirect defenses. *Insect science*, *24*(6), 947–960. https://doi.org/10.1111/1744-7917.12464
16. Revtyo, O. Ya., Zolin, O. O. (2023). Peculiarities of soybean cultivation under climate change conditions. *Taurian Scientific Bulletin*, *133*, 105–112. https://doi.org/10.32782/2226-0099.2023.133.15
17. Tan, S., Yang, C., Mei, X., Shen, S., Raza, W., Shen, Q., Xu, Y. (2013). The effect of organic acids from tomato root exudates on rhizosphere colonization of Bacillus amyloliquefaciens T-5. *Applied Soil Ecology*, *64*, 15–22. https://doi.org/10.1016/j.apsoil.2012.10.011
18. Tan, Y., Wang, Z. (2024). Research on influencing factors of soybean yield in China's northeast black soil region based on PLS-SEM. *Frontiers in Sustainable Food Systems*, *8*, 1436224. https://doi.org/10.3389/fsufs.2024.1436224
19. Trivedi, P., Leach, J. E., Tringe, S. G., Sa, T., Singh, B. K. (2020). Plant–microbiome interactions: from community assembly to plant health. *Nature reviews microbiology*, *18*(11), 607–621. https://doi.org/10.1038/s41579-020-0412-1
20. Turovnik, Yu., Parfeniuk, A., Demianiuk, O., Beznosko, I. (2020). Root exometabolites of sunflower plants as an influencing factor on the viability of the phytopathogenic fungus *Alternaria alternata* (Fr.) Keiss. *Balanced Nature Management*, (1), 102–107. (in Ukrainian). https://doi.org/10.33730/2310-4678.1.2020.203936
21. Vandenkoornhuyse, P., Quaiser, A., Duhamel, M.,

Le Van, A., Dufresne, A. (2015). The importance of the microbiome of the plant holobiont. *New Phytologist*, *206*(4), 1196–1206. https://doi.org/10.1111/nph.13312.

22. Wu, L., Weston, L. A., Zhu, S., Zhou, X. (2023). Rhizosphere interactions: root exudates and the rhizosphere microbiome. *Frontiers in plant science*, *14*, 1281010. https://doi.org/10.3389/fpls.2023.1281010
23. Xu, H., Yan, L., Zhang, M., Chang, X., Zhu, D., Wei, D., ..., Yang, W. (2022). Changes in the density and composition of rhizosphere pathogenic Fusarium and beneficial Trichoderma contributing to reduced root rot of intercropped soybean. *Pathogens*, *11*(4), 478. https://doi.org/10.3390/pathogens11040478
24. Yusnawan, E., Inayati, A., Baliadi, Y. (2019). Isolation of antagonistic fungi from rhizospheres and its biocontrol activity against different isolates of soil borne fungal pathogens infected legumes. *Biodiversitas*, *20*(7), 2048–2054. https://doi.org/10.13057/biodiv/d200735
25. Zaimenko, N. V., Didyk, N. P., Dzyuba, O. I., Zakrasov, O. V., Rositska, N. V., Viter, A. V. (2014). Enhancement of drought resistance in wheat and corn by nanoparticles of natural mineral analcite. *Ecologia Balkanica*, *6*(1), 1–10.
26. Zaimenko, N. V., Pavliuchenko, N. A., Ellanska, N. E., Ivanytska, B. O., Kharytonova, I. P., Yunosheva, O. P., ..., Tian, L. (2020). Comparative analysis of agrochemical, allelopathic and microbiological characteristics of the soil environment for *Actinidia arguta* (Siebold et Zucc.) Planch. ex Miq. cultivated in Ukraine and two provinces of China. *Plant introduction*, (85/86), 3–14. https://doi.org/10.46341/PI2020002