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DEVELOPMENT STRATEGY FOR THE NEW ENVIRONMENTALLY FRIENDLY MULTIFUNCTIONAL BIOFORMULATIONS BASED ON SOIL STREPTOMYCETES

Microorganism biosynthetic activity attracts researchers' attention both in terms of the rules of various biologically active compounds synthesis by bacteria and of the industrially important microbial metabolites application in various industries and agriculture.

*Scientific principles of an integrated approach to selection and study of soil streptomycetes biosynthetic potential have been formulated. Based on the approach the strains *Streptomyces avermitilis* IMV Ac-5015, *S. netropsis* IMV Ac-5025 and *S. violaceus* IMV Ac-5027 with antagonistic activity against phytopathogens and phytonematodes were obtained. Their ability to synthesize a wide range of biologically active metabolites was shown. The latter include antiparasitic antibiotics, free amino and fatty acids, lipids, phytohormones (auxins, cytokinins, gibberellins, brassinosteroids) and others. The strategy of creation of new environmentally friendly multifunctional metabolic bioformulations with a complex mechanism of action, combining such properties as biocontrol of phytopathogens and phytonematodes and the ability to optimize the hormonal balance and enhance plants tolerance has been substantiated theoretically and confirmed experimentally. Under the action of these bioformulations reprogramming of plant genomes occur; turning towards the synthesis induction of small regulatory si/mi RNA with antipathogenic and antiparasitic properties. Under the bioformulations application the microorganisms development in root zone is activated, the overall biological activity of soil increases, the level of plant damage caused by phytopathogens and parasitic nematodes reduces, the yield increases and its quality improves.*

The new bioformulations showed combined biological activity caused both by direct action on various pathogens of different etiology and by phytheregulatory activity or by increasing of plant resistance to biotic and abiotic stresses.

Key words: soil streptomycetes, antagonism, phytopathogens, phytonematodes, lipids, phytohormones, steroid compounds, metabolic bioformulations, si/mi RNA.

Mycelial soil actinobacteria, in particular the representatives of *Streptomyces* genus, which make up from 20 % to 46 % of soil microbiota biodiversity, are the source of biologically active substances, differing in their chemical structure and activity spectrum [1]. From 23000 registered biologically active secondary metabolites, produced by microorganisms, streptomycetes synthesize over 10000 compounds, representing almost 45 % of biologically active metabolites of microbial origin [2]. Most of the synthesized compounds exhibit antibiotic properties [3]. These microorganisms also synthesize other physiologically active metabolites, such as amino acids, enzymes, vitamins, lipids, phytohormones and etc. [4, 5]. Streptomycetes and their metabolites are used to develop bioformulations for medicine and agriculture, including plant protection against phytopathogens and phytonematodes.

The *Streptomyces* genus antagonistic activity spectrum and its biosynthetic capacity are the key issues studied at the Department of General and Soil Microbiology of the Zabolotny Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine (IMV NASU). As a result of a years-long research of soil streptomycetes physiological and biochemical characteristics, one of the largest Ukrainian collections of streptomycetes (over 3000 strains), promising for use in biotechnology, was created (at the Department of General and Soil Microbiology of IMV NASU).

The main objective of the research is to develop scientific backgrounds for an integrated approach to screening and determination of the biosynthetic potential of streptomycetes with antagonistic activity against phytopathogens and phytonematodes. Another aim was to theoretically justify and experimentally confirm the development strategy of new environmentally friendly multifunctional metabolic bioformulations based on soil streptomycetes with a complex mechanism of action.

In recent years, as a result of soil streptomycetes screening, new highly active antagonists against phytopathogenic micromycetes, bacteria and phytonematodes were selected. According to their morphological, cultural, physiological, biochemical and molecular genetic characteristics, they were identified as *Streptomyces avermitilis* IMV Ac-5015 [6, 7], *S. netropsis* IMV Ac-5025 [8] and *S. violaceus* IMV Ac-5027 [9]. The strains show significant antagonistic activity against phytopathogenic micromycetes of genera *Alternaria*, *Fusarium*, *Nigrospora* (growth inhibition zone was 9.5–27.0 mm). Antagonism against pathogenic bacteria of genera *Pseudomonas*, *Xanthomonas*, *Clavibacter* was confirmed (growth inhibition zone was 12–48 mm) [8, 9].

The research showed that *S. avermitilis* IMV Ac-5015, *S. netropsis* IMV Ac-5025 and *S. violaceus* IMV Ac-5027 had almost no effect on the reference strains of microorganisms conditionally pathogenic to humans [8, 9]. Therefore, the use of the studied streptomycetes strains for medicine is not promising. This fact excludes the possibility of occurrence of multiple resistance in patients to the metabolites of the studied strains.

The selected streptomycetes showed high antiparasitic activity against plant parasitic nematodes (stem, cyst-forming, sedentary (root-knot) [10]. Ethanol extracts from the studied strains biomass induced a 59–83 % *in vitro* death of invasive larvae of root-knot nematode *Meloidogyne incognita* already within half an hour. After 24 hours, death of 100 % individual phytonematodes was observed as a result of action of the aqueous solution of ethanol extracts from *S. avermitilis* and *S. violaceus* biomass, and death of 75 % under the influence of *S. netropsis* biomass extract.

The published data analysis shows that now scientists are actively seeking the streptomycetes with antagonistic activity against plant parasitic nematodes [2, 11, 12]. In Japan *S. lavendulae* SANK 64297, synthesizing a soluble macrocyclic antibiotic, was isolated from soil. The second age *M. incognita* larvae treatment with *S. lavendulae* SANK 64297 for 24 hours completely inhibited their ability to infect plants. It was established that this substance's mechanism of antagonistic action included protein biosynthesis inhibition at the level of transcription [13, 14].

The cultural liquid of *S. sampsonii* KK1024, isolated from crab shells, showed high chitinolytic, protease and lipase activity. On the third day of ac-

tion its 50 % solution resulted death of 82 % root-knot nematode *M. incognita* larvae [15]. Comparison of these results with our data indicates that under the influence of aqueous solutions of ethanol extracts from *S. avermitilis* IMV Ac-5015 and *S. violaceus* IMV Ac-5027 biomass the 100 % death of nematodes *M. incognita* was observed after 24 hours, which proves their higher nematocidal efficiency.

By using modern physical and chemical methods, such as spectrum densitometric thin layer chromatography (TLC), UV- and IR-spectroscopy, high performance liquid and gas chromatography with mass detectors (HPLC LC/MS, GC/MS), matrix-activated laser desorption/ionization time-of-flight spectrometry (MALDI-TOF), nuclear magnetic resonance (^1H NMR) and bioinformatics methods it was determined that the studied streptomycetes synthesized antibiotic substances of different chemical nature. For example, *S. avermitilis* IMV Ac-5015 produces a macrolide antibiotic avermectin, *S. netropsis* IMV-Ac 5025 produces polyenes (tetraen and pentaen) and *S. violaceus* IMV Ac-5027 produces anthracycline antibiotics [9, 10].

Besides antibiotics, such physiologically active substances as amino acids, lipids (including fatty acids), phytohormones, sterol compounds and lectins were found among the streptomycete metabolites [7, 9, 10].

Of all biologically active substances, lipids have a special role. They are cell membranes components and energetically favorable oxidation substrates. They exhibit antioxidant properties and are involved in the antibiotics synthesis [16]. A wide range of lipids was revealed in the biomass of the studied strains, such as free fatty acids, phospholipids, mono- and diglycerides, sterols, triglycerides, sterols ethers and waxes in various proportions. In the biomass of *S. avermitilis* IMV Ac-5015 phospholipids (up to 29 %), sterols (up to 22 %) and triglycerides (up to 14 %) prevailed. In the biomass of *S. netropsis* IMV Ac-5025 and *S. violaceus* IMV Ac-5027 triglycerides (over 50 %) and phospholipids (over 18 %) dominated. When evaluating the possible impact of microbial lipids on plants, attention should be paid to the fact that phospholipids provide fluidity and plastic properties of plant cell membranes and play an important role in energy processes and in the formation of the plants' induced resistance to phytopathogens [17].

Phytohormones of stimulatory (auxins, cytokinins, gibberellins) and inhibitory (abscisic acid, ethylene) action play a significant role in regulation of plant growth and development and establishment of microbial-plant interactions [18]. The ability of soil microorganisms of *Azotobacteriaceae*, *Bacillaceae*, *Enterobacteriaceae*, *Pseudomonadaceae* families and others to synthesize phytohormones is well known. Representatives of the *Actinomycetaceae* family, including *Streptomyces* genus, are the least studied in this respect.

The data on phytohormones synthesis by the studied species of streptomycetes are almost nonpresent in literature. Our results indicate that *S. avermitilis* IMV Ac-5015, *S. netropsis* IMV Ac-5025 and *S. violaceus* IMV Ac-5027 are able to synthesize phytohormones of stimulating (auxins, cytokinins, gibberellins) and inhibitory (abscisic acid) action (Table 1).

Table 1

Phytohormone content in the streptomycete biomass

Phytohormone		Phytohormone content $\mu\text{g/g ADB}^*$		
		<i>S. avermitilis</i> IMV Ac-5015	<i>S. netropsis</i> IMV Ac-5025	<i>S. violaceus</i> IMB Ac-5027
Auxins	Indole-3-acetic acid	47.09 \pm 0.72	27.85 \pm 1.76	12.22 \pm 1.16
	Indole-3-butyric acid	0.97 \pm 0.10	54.54 \pm 2.46	67.22 \pm 2.73
	Indole-3-acetic acid hydrazide	15.33 \pm 0.41	14.27 \pm 1.26	16.51 \pm 1.35
	Indole-3-carboxyaldehyde	65.13 \pm 0.85	7.54 \pm 0.92	6.71 \pm 0.86
	Indole-3-carbinol	“_”	0.06 \pm 0.03	2.59 \pm 0.54
	Indol-3-carboxylic acid	“_”	2,68 \pm 0,55	8,89 \pm 0,99
	Total auxins	128.52 \pm 3.78	106.94 \pm 3.45	114.14 \pm 3.56
Cytokinins	Zeatin	41.82 \pm 2.16	10.01 \pm 1.05	13.33 \pm 1.22
	Yzopentyl adenine	50.18 \pm 2.36	53.38 \pm 2.44	41.75 \pm 2.15
	Zeatin ribozid	42.80 \pm 2.18	40.28 \pm 2.12	11.03 \pm 1.11
	Izopentanol adenosine	“_”	1.90 \pm 0.46	15.49 \pm 1.31
	Total cytokinins	134.80 \pm 3.87	105.57 \pm 3.42	81.60 \pm 3.01
Gibberellins		22.50 \pm 0.71	8.12 \pm 0.95	13.17 \pm 1.21
Abscisic acid		0.23 \pm 0.16	2.81 \pm 0.56	1.32 \pm 0.38

Note: * ADB – absolutely dry biomass.

“_” – not revealed.

In the biomass of the studied streptomycetes grown in liquid medium a significant amount of stimulating phytohormones is accumulated, including auxins (up to 128 $\mu\text{g/g ADB}$), cytokinins (up to 134.8 $\mu\text{g/g ADB}$) and gibberellins (up to 22.5 $\mu\text{g/g ADB}$), unlike the small quantity of inhibitory phytohormone abscisic acid (up to 2.81 $\mu\text{g/g ADB}$).

Among the phytohormonal compounds, an important regulatory role in various growth processes of both plants and microorganisms is played by auxins, which are the indole derivatives. They affect different metabolic systems, such as the synthesis of nucleic acids, proteins, carbohydrates, lipids and others. The soil microorganisms need auxins for their growth and development and to establish relations with plants and other microbiota [19].

Among the synthesized auxins, the major active forms were detected: indole-3-acetic acid (IAA) and indole-3-butyric acid. IAA transformation products were also detected, some of which (indole-3-carboxyaldehyde and indole-3-acetic acid hydrazide) can exhibit antimicrobial activity [11].

The total quantity of auxins synthesized by the strains was 106.9–128.5 $\mu\text{g/g ADB}$. This was much higher than in plants that probably can help streptomycetes to establish relations with plants and soil microorganisms. Moreover, it is known that epiphytic bacteria and rhizospheric microorganisms play a primary role in the tryptophan conversion of plant exudates to IAA [19].

It is known from literature that auxins increase the biomass accumulation and sporulation of streptomycetes, intensify certain metabolic processes, including the synthesis of antibiotics, and regulate the biosynthesis of other hormones, when used in optimal concentrations [11, 20].

Among the cytokinins synthesized by streptomycetes, both active (zeatin and izopentyl adenine) and ribosilated (zeatin riboside and izopentyl adenosine) transport forms were identified. The total quantity of synthesized auxins comprised 81.6–134.8 $\mu\text{g/g ADB}$.

Cytokinins can stimulate growth and pigment formation by certain gram-positive bacteria, including streptomycetes, and the atmospheric nitrogen fixation by diazotrophs [19]. Exogenous cytokinins increase the biosynthesis of antibiotics, amino acids and certain enzymes [21] and affect the content of endogenous phytohormones in microorganisms [19]. There is a correlation between phytohormones in a microbial cell: auxins regulate biosynthesis of cytokinins [21]. An increase in the cytokinin quantity in plants enhances resistance against various stresses: abiotic and biotic i.e. to fungi, viruses and pests infections [22].

Compared with auxins and cytokinins, the gibberellin content in the biomass of the studied streptomycetes was lower and ranged within 8.1–22.5 µg/g ADB. It is known that exogenous gibberellins can stimulate the growth of *Azotobacter*, *Pseudomonas*, yeasts and mycelium rhizospheric fungi and enhance the activity of nitrogen fixation some by certain cyanobacteria [19, 23]. The ability of gibberellins to induce expression of amylase and cellulase genes as well as other enzymes synthesis in microorganisms was established [21].

The presence of phytohormones in the producers biomass directly indicates their growth regulatory activity that can be expressed through the stimulation of plant growth and development, as well as through increasing their resistance to phytopathogens.

Lately, some researchers have supported the idea that phytohormones serve not just as stimulators of plant growth and development, but also as the key regulators of the plant immunity formation, in particular, the synthesis of ethylene, salicylic acid, jasmonic acid and others [23]. Hormone-dependent genes of resistance to pathogens and pests were found in *Arabidopsis thaliana* [23].

The hormonal balance and secondary metabolites content, including phytoosterols and others, influence plant resistance to pathogens and to abiotic and biotic environmental factors. It is known that streptomycetes are able to synthesize not only phytohormones but also other isoprenoid compounds of sterol nature that can directly induce systemic plant resistance [23, 24].

There are no data in the literature concerning the ability of *S. avermitilis*, *S. netropsis* and *S. violaceus* to synthesize isoprenoid sterol compounds. For the first time we have shown that sterol compounds are present in the studied producers biomass (Table 2).

Table 2

Accumulation of sterol compounds in the streptomycete biomass

Sterol compound	The content of steroid compounds, µg/g ADB *		
	<i>S. avermitilis</i> IMV Ac-5015	<i>S. netropsis</i> IMV Ac-5025	<i>S. violaceus</i> IMV Ac-5027
Squalene	“–”	212.13 + 4.85	8.17 + 0.89
Cholesterol	192.6 + 4.63	131.45 + 3.82	4.94 + 0.74
Ergosterol	93.44 + 3.22	398.86 + 6.66	159.23 + 4.21
Sitosterol	0	201.21 + 4.73	25.13 + 1.67
Stigmasterol	0	2.02 + 0.47	5.24 + 0.76
24-epibrassinolide	268.16 + 5.46	8.16 + 0.95	1.49 + 0.41

Note: * ADB – absolutely dry biomass.

“–” – not determined.

S. netropsis IMV Ac-5025 and *S. violaceus* IMV Ac-5027 synthesize cholesterol, ergosterol, sitosterol, stigmasterol, 24-epibrassinolide and their precursor squalene.

The important fact is that *S. avermitilis* IMV Ac-5015 does not synthesize sitosterol, which is a precursor of stigmasterol synthesis. Stigmasterol is abundant in the eggs and females of plant parasitic nematodes, but helminths are not able to synthesize it. Absence of this sterol limits the possibility of existence and reproduction of parasites and decreases the degree of realization of their biological species potential [23]. Absence of sitosterol and stigmasterol also inhibits reproduction of certain pathogenic oomycetes (*Phytophthora* and *Pythium genera*) [23].

According to the literature the ratio of sterols is important for plants, in particular the ratio of sitosterol to stigmasterol, which can change, due to many internal and external factors, especially during the seeds germination and plant tissues aging, under low temperatures, drought, exposure to phytopathogens and pesticides and others [23, 24]. In the conditions of the sitosterol/stigmasterol ratio reduction the degree of plant sensitivity to phytopathogens decreases, thus increasing the accumulation of cholesterol [25]. Therefore, the use of exogenous sterol compounds of microbial origin can improve their balance in a plant, which is important to increase the level of plant resistance to phytopathogens.

Considering the negative environmental impact of pesticides, the bioformulations of the fourth generation for plant protection are spreading more widely. They are based on biologically active substances, aimed not so much at killing phytopathogens as at reducing their harm and increasing plant resistance properties.

Since the studied streptomycetes are promising producers of biologically active substances we have developed multifunctional metabolic bioformulations based on cultural liquid and ethanol extracts from biomass of the following strains: Avercom and Avercom-nova (based on *S. avermitilis* IMV Ac-5015), Phytovit (based on *S. netropsis* IMV Ac-5025) and Violar (based on *S. violaceus* IMV Ac-5027). The bioformulations combine antagonistic activity against phytopathogens and phytonematodes with the properties of growth regulators and plant adaptogens.

A number of bioformulations based on avermectin have been created in other countries and are used as biopesticides to regulate the number of exo- and endoparasites of plants, including nematodes. There are well-known bioformulations, such as Phytoverm, Formatsyn (Aversect 2), Aversectin C, Univerm, Eqvisect (Russia); Ivermectin, Ivomec, Eqvalan, Doramectin, Abamectin, Zimectrin (USA) and others [26]. On the basis of *S. avermitilis* IMV Ac-5015, we have developed a new bioformulation Avercom, which, unlike the existing commercial preparations, was characterized by not only an antiparasitic, but also a phytostimulating effect [7, 26]. Its modification Avercom-nova was developed by adding new substances with elicitor properties (salicylic acid or chitosan) in order to enhance the phytoprotective effect.

Plant resistance enhancement has several advantages over the use of biopesticides, because it is based on induction of the natural defense mechanisms of plants. The resistance is systemic and long enough. Protective systems are lounded before the contact with pathogens (fungi, viruses, bacteria, and phytonematodes). Stimulation of several defense mechanisms makes it unlikely for phytopathogens to adapt to the immunized plants [23].

The study of certain physiological and molecular genetic mechanisms of

newly designed metabolic bioformulations action was carried out in laboratory. Plot vegetation and field experiments were conducted in the conditions of artificially created invasive background, and in the field with a natural background using crops, such as spring wheat of Early 93, Isolde and Griso cultivars, spring rape of Kalinowski cultivar, potato of Slavyanka and Bellarozza cultivars, cucumber of Nijinsky, Angelina and Gravina cultivars, tomatoes of Sanka cultivar, Chinese cabbage of Michel cultivar.

One of the systemic plant resistance markers is the activity of phenylalanine-ammonia-lyase (PAL) (EC 4.3.1.5) enzyme, which catalyzes deamination of phenylalanine in the beginning of phenyl-propanoid pathway of cell protection compounds synthesis, such as the precursors of lignin, phytoalexin, flavonoids, coumaric and cinnamic acids and others [23].

Under the pre-sowing treatment of seeds of Griso cultivar spring wheat, the studied biological bioformulations significantly stimulated the PAL activity both on the natural background and on the background of artificial infection with *Fusarium oxysporum* n.33. Thus, in the variants to which the bioformulations were applied on the natural background, the PAL activity increased by 1.5–2.6 times, on the artificial infectious background this index increased by 2.0–3.3 times as compared to the control. The chemical fungicide Maxim Star increased the activity of PAL only by 1.3 and 1.5 times, respectively. We believe that stimulation of PAL activity by means of metabolic bioformulations application can be explained by the wide range of physiologically active substances, contained in their composition, including phytohormones and sterol compounds that can act as biogenic elicitors. Increase in PAL activity confirms the induction of plant resistance to phytopathogens when bioformulations are used. Formation of plant protective reactions is related to the inclusion of various signaling systems, which transmit elicitor signals to genome that activates the expression of protective genes [23].

Now the mechanisms of plant resistance to diseases are actively studied at the molecular genetic level. It is known from the literature, that endogenous small noncoding short interfering and micro RNAs (si/mi RNA) are involved in plant defense reactions in response to phytopathogen lesions. Small regulatory si/mi RNA act in two ways. One of them is the blocking of translation of their own mRNA-transcripts of genes, which control plant growth and development, the expression of which is induced by pathogens. Another way is the interaction of plant si/mi RNA with molecular targets of mRNA of pathogenic and parasitic organisms and the destruction of them by enzymes. Si/mi RNA play an important role in the regulation of plant immune protection taking part in the transcriptional gene silencing or TGS (by means of DNA methylation at the target locus) or in the post-transcriptional gene silencing or PTGS (by means of blocking the target mRNA translation or by promoting its degradation) [28].

Microbial PR/PGP inducers cause reprogramming of plant genome, i.e. induce the plant cells synthesis of immune-protective si/miRNA with antisense sequences both to the plant mRNA and to the nematode mRNA.

In a series of experiments with spring rape, spring wheat, potato and Chinese cabbage the influence of the developed metabolic bioformulations on the synthesis of endogenous si/mi RNAs and their silencing activity have been studied [29, 30]. Using the method of DOT blot hybridization a significant increase in the difference of homology degree (up to 27–49 %) was deter-

mined between cytoplasmic mRNAs from the unaffected control plants and si/mi RNAs from the experimental plants, infected with cyst-forming nematode *Heterodera schachtii* Schmidt and the ones, treated with the metabolic bioformulations. A significant increase in silencing activity (up to 25–45 %) of si/mi RNAs, isolated from cells of studied plants, infected and treated with bioformulations, against the mRNA from cells of nematodes *H. schachtii* larvae, has been shown. The obtained molecular-genetic indexes testify that bio-protective effect of microbial PR/PGP inducers occurs through the stimulation of the plant cells synthesis of small regulatory si/miRNA with immune-protective against nematode *H. schachtii* Schmidt properties. As a result the plant resistance to this pest is increased.

This conclusion was confirmed in experiments at the whole plant organism level. It was revealed, that the pre-sowing bioformulations application to the seeds contributed to the reduction of plants infection with phytohelminths and phytopathogens.

In the field experiments with rape on artificially created with *H. schachtii* invasive background the population density of parasitic nematodes complex on the roots of plants was significantly reduced under the Avercom action by 9.8 times, and under the Avercom-nova and Violar action – twice [29].

According to the results of many authors' research it can be affirmed, that bioformulations, which are used in plant cultivation, can influence a number of natural associations of soil microorganisms, their biochemical activity, initiate synthesis of biologically active compounds by these microorganisms, increase the ability of microbiota to produce antibiotic substances against phytopathogens and enhance the resistance of soil microbiota to unfavorable environmental factors, caused by anthropogenic influence [23, 24, 25, 26].

According to the field experiments results in the root zone of plants treated with bioformulations the overall soil biological activity increased up to 12–54 % as compared to the control. At the same time, the application of chemical fungicide Konfidor Maxi depressed the respiration activity up to 42 % in comparison with the control variant [29].

The study of soil microbial communities in the root zone of rape plants has shown an increase in the number of microorganisms of the major ecological and functional groups (such as nitrogen fixing, phosphorus mobilizing, ammonifying and amylolytic) by 1.2–2.2 times in the conditions of bioformulations application [29]. Konfidor Maxi negatively affected the growth of microorganisms, which number was lower, compared to the control variant by 1.2–1.7 times.

We can presume that the treatment of plant seeds with bioformulations positively affects the development of microbial-plant system. This creates a precondition for the microbiological control of soil phytopathogens because the natural microbiota acts as a stabilizing factor, hindering the development of phytopathogens in soil [26]. An important result of the bioformulations using is a positive effect on soil microbiota, which plays an important role in soil fertility [18, 23, 26].

Efficiency of the developed new multifunctional metabolic bioformulations has been confirmed in the field on a number of crops, such as wheat, spring rape, potatoes, cucumbers, tomatoes, Chinese cabbage [7, 26, 27, 29, 30, 31, 32].

Due to presence of biologically active substances complex, metabolic bioformulations exhibit phytoprotective, growth regulatory and adaptogenic

effect on plants, resulting an increased yield (up to 10–33 %) and improved biochemical quality characteristics of the final product [7, 26, 27, 29, 30, 31, 32]. Thus, in the dry matter of tomato fruits the content of sugars (by 1.6 times), vitamin C, β -carotene (by 2 times) increased and the content of nitrates reduced (by 2–3 times) [31]. The category of wheat grain is increased, which indicates an improvement of such important characteristics as gluten content, glass, nature and others.

Thus, we have developed scientific principles of an integrated approach to selection and determination of biologically active complex of soil streptomycetes. We have proved theoretically and confirmed experimentally the strategy of ecologically friendly multifunctional metabolitic bioformulations development in order to improve the formation and functioning of soil microbial cenoses and highly productive microbial-plant systems. Biological preparations Avercom, Avercom nova, Violar and Phytovit, based on metabolitic complexes of *S. avermitilis* IMV Ac-5015, *S. netropsis* IMV Ac-5025, and *S. violaceus* IMV Ac-5027, showed combined biological activity caused both by direct action on various pathogens of different etiology and indirectly through phyto regulatory activity or by increasing the plant resistance to biotic and abiotic stresses.

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СТРАТЕГІЯ СТВОРЕННЯ НОВИХ ЕКОЛОГІЧНО БЕЗПЕЧНИХ ПОЛІФУНКЦІОНАЛЬНИХ БІОПРЕПАРАТІВ НА ОСНОВІ ҐРУНТОВИХ СТРЕПТОМІЦЕТІВ

Резюме

Вивчення біосинтетичної активності мікроорганізмів привертає увагу дослідників як з точки зору пізнання закономірностей утворення бактеріальною клітиною різноманітних біологічно активних сполук, так і з точки зору використання промислово важливих мікробних метаболітів для різних галузей промисловості й сільського господарства.

Розроблено наукові засади комплексного підходу до селекції і вивчення біосинтетичного потенціалу ґрунтових стрептоміцетів, на основі якого отримані штамми антагоністів фітопатогенів і фітонематод *Streptomyces avermitilis* IMB Ac-5015, *S. netropsis* IMB Ac-5025 і *S. violaceus* IMB Ac-5027. Показана їхня здатність до синтезу широкого спектру біологічно активних метаболітів: антипаразитарних антибіотиків, вільних аміно- і жирних кислот, ліпідів, фітогормонів (ауксинів, цитокінінів, гіберелінів, брасиностероїдів) та ін. Теоретично обґрунтовано та експериментально підтверджено стратегію створення на основі виділених продуцентів нових екологічно безпечних поліфункціональних метаболічних біопрепаратів комплексної дії, які поєднують властивості біоконтролю фітопатогенів і фітонематод із здатністю оптимізувати гормональний баланс і активізувати захисні системи рослин. За дії біопрепаратів відбувається репрограмування геному рослин у напрямку індукції синтезу малих регуляторних si/mi РНК з антипатогенними та антипаразитарними властивостями. За умов застосування біопрепаратів активізується розвиток мікроорганізмів кореневої зони, підвищується загальна біологічна активність ґрунту, зменшується

рівень ураження рослин фітопатогенами і паразитичними нематодами, підвищується урожай та поліпшується його якість.

Ключові слова: ґрунтові стрептоміцети, антагонізм, фітопатогени, фітонематоди, ліпіди, фітогормони, стероїдні сполуки, метаболічні біопрепарати, si/mi РНК.

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СТРАТЕГИЯ СОЗДАНИЯ НОВЫХ ЭКОЛОГИЧЕСКИ БЕЗОПАСНЫХ ПОЛИФУНКЦИОНАЛЬНЫХ БИОПРЕПАРАТОВ НА ОСНОВЕ ПОЧВЕННЫХ СТРЕПТОМИЦЕТОВ

Резюме

Изучение биосинтетической активности микроорганизмов привлекает внимание исследователей как с точки зрения познания закономерностей образования бактериальной клеткой различных биологически активных соединений, так и с точки зрения использования промышленно важных микробных метаболитов для различных отраслей промышленности и сельского хозяйства.

Разработаны научные основы комплексного подхода к селекции и изучению биосинтетического потенциала почвенных стрептомицетов, на основе которого получены штаммы антагонистов фитопатогенов и фитонематод *Streptomyces avermitilis* IMB Ac-5015, *S. netropsis* IMB Ac-5025 и *S. violaceus* IMB Ac-5027. Показана их способность к синтезу широкого спектра биологически активных метаболитов: антипаразитарных антибиотиков, свободных аминокислот и жирных кислот, липидов, фитогормонов (ауксинов, цитокининов, гиббереллинов, брассиностероидов) и др. Теоретически обоснована и экспериментально подтверждена стратегия создания на основе выделенных продуцентов новых экологически безопасных полифункциональных метаболитных биопрепаратов комплексного действия, которые сочетают свойства биоконтроля фитопатогенов и фитонематод со способностью оптимизировать гормональный баланс и активизировать защитные системы растений. При действии биопрепаратов происходит репрограммирование генома растений в направлении индукции синтеза малых регуляторных si/mi РНК с антипатогенными и антипаразитарными свойствами. В условиях применения биопрепаратов активизируется развитие микроорганизмов корневой зоны, повышается общая биологическая активность почвы, уменьшается уровень поражения растений фитопатогенами и паразитическими нематодами, повышается урожай и улучшается его качество.

Ключевые слова: почвенные стрептомицеты, антагонизм, фитопатогены, фитонематоды, липиды, фитогормоны, стероидные соединения, метаболитные биопрепараты, si/mi РНК.

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