

SCIENTIFIC HORIZONS

Journal homepage: <https://sciencehorizon.com.ua>

Scientific Horizons, 28(8), 102-117

UDC 577.2:579.2

DOI: 10.48077/scihor8.2025.102



Molecular-genetic and biotechnological foundations for the development of complex microbial preparations for balanced plant nutrition

Olena Karatieieva

PhD in Agricultural Sciences, Associate Professor
Mykolaiv National Agrarian University
54008, 9 George Gongadze Str., Mykolaiv, Ukraine
<https://orcid.org/0000-0002-0652-1240>

Yevhen Barkar

PhD in Agricultural Sciences, Associate Professor
Mykolaiv National Agrarian University
54008, 9 George Gongadze Str., Mykolaiv, Ukraine
<https://orcid.org/0000-0002-0692-5392>

Olena Yulevich

PhD in Technical Sciences, Associate Professor
Mykolaiv National Agrarian University
54008, 9 George Gongadze Str., Mykolaiv, Ukraine
<https://orcid.org/0000-0003-1594-0700>

Iryna Liuta

Assistant
Mykolaiv National Agrarian University
54008, 9 George Gongadze Str., Mykolaiv, Ukraine
<https://orcid.org/0000-0002-1672-2337>

Vitalii Overchenko*

PhD in Agriculture Sciences, Associate Professor
National University of Life and Environmental Sciences of Ukraine
03041, 15 Heroiv Oborony Str., Kyiv, Ukraine
<https://orcid.org/0009-0009-5435-6815>

Article's History:

Received: 21.01.2025

Revised: 26.07.2025

Accepted: 27.08.2025

Abstract. The purpose of this study was to identify the molecular genetic mechanisms that regulate the functional activity of microbial preparations in the plant rhizosphere. The methodology included analysing the expression of key genes, comparing the effectiveness of different strains of microorganisms in the soil environment, summarising the factors affecting the stability of biological products, and systematising the data to establish the relationship between genetic mechanisms and agronomic indicators. The results confirmed that the expression of key genes responsible for nitrogen fixation, phosphorus mobilisation, and synthesis of biologically active compounds determines the effectiveness of biological products in the soil environment, which was verified

Suggested Citation:

Karatieieva, O., Barkar, Ye., Yulevich, O., Liuta, I., & Overchenko, V. (2025). Molecular-genetic and biotechnological foundations for the development of complex microbial preparations for balanced plant nutrition. *Scientific Horizons*, 28(8), 102-117. doi: 10.48077/scihor8.2025.102.



Copyright © The Author(s). This is an open access article distributed under the terms of the Creative Commons Attribution License 4.0 (<https://creativecommons.org/licenses/by/4.0/>)

*Corresponding author

by genetic analysis of functional markers, microbiological study of strain viability, and analytical determination of available nutrients and plant productivity. The combination of nitrogen-fixing, phosphate-mobilising, and biostimulating microorganisms ensured the development of stable microbial communities capable of increasing the bioavailability of nutrients and crop productivity. When *Azospirillum brasilense* was used on non-legume crops, the concentration of ammonium compounds in the rhizosphere increased by up to 40%, which was accompanied by an increase in nitrogen nutrition. The use of phosphate-mobilising bacteria allowed reducing the rate of phosphate fertiliser application by 25-40% without loss of yield. Overall, the use of biological products on cereals, legumes, and vegetables reduced the need for chemical fertilisers by 20-30% without reducing agronomic efficiency. Stabilisation of microorganisms through microencapsulation ensured increased survival in the soil environment and uniform distribution in the rhizosphere, while spray drying enabled the production of powdered forms of biological products with a long shelf life. The combination of microbial preparations with nanoforms of mineral fertilisers contributed to the uniform release of nutrients, which positively influenced their assimilation, although the exact coefficients of this process were not presented in the study. The practical significance of the findings obtained lies in the possibility of targeted development of biological products with the predicted ability to increase the uptake of nitrogen and phosphorus by plants, reduce the need for mineral fertilisers, and ensure stable crop productivity under intensive farming conditions.

Keywords: biotechnology products; consortium; nitrogen fixation; phosphorus mobilisation; biologically active substances; microencapsulation, gene expression

INTRODUCTION

Recently, modern biotechnology products have been increasingly used in various branches of agriculture. For example, feed additives are widely used in animal husbandry to increase production efficiency (Tsvihun et al., 2025), and in crop production, the technology of using phytopreparations of microbiological origin, such as azo-fixing, phosphate-mobilising bacteria and microorganisms, is becoming increasingly widespread, which leads to increased plant productivity and ensures the production of environmentally friendly food products of animal and plant origin (Panfilova et al., 2025). Balanced plant nutrition is ensured by the interaction of the root system with rhizosphere microorganisms, which affect the availability of nutrients, plant metabolism, and stress resistance. Nitrogen fixers, phosphate-mobilising bacteria, and microorganisms that produce phytohormones and exopolysaccharides promote the development of the root system and improve the absorption of macro- and microelements (Doolotkeldieva & Bobusheva, 2024). Microbial biological products help to control these processes, but their effectiveness depends on the composition, interspecies interaction of strains, and the ability to colonise the rhizosphere. The development of stable consortia requires an understanding of the molecular mechanisms that regulate the adaptation of microorganisms to different soil and climatic conditions.

International organisations, such as the Food and Agriculture Organization (2025) and the Organisation for Economic Co-operation and Development (OECD) (2019), support research into the development of biological products aimed at increasing agricultural production efficiency and reducing dependence on mineral fertilisers. Horizon Europe (n.d.) programmes are aimed at exploring the genetic potential of microorganisms, optimising their biochemical pathways,

and creating stable microbial consortia for diverse types of agroecosystems. Biotechnological research in recent years has opened new opportunities for the development of microbial products with improved properties (Borko et al., 2025). The optimisation of biotechnological processes that ensure the increased viability of beneficial microorganisms and their resistance to external factors has become one of the key areas, as confirmed by Yu.V. Karlash and V.O. Krasinko (2022). The researchers proposed an approach to preserving the functional activity of microorganisms in biological products for a long time. O.P. Tkachuk et al. (2020) presented new ecological aspects of the use of biological products in agriculture, particularly their role in reducing the chemical load and improving the bioavailability of macro- and microelements. The study proved the ability of biological products to maintain the biodiversity of agroecosystems, which changes approaches to their use. Of value is the contribution of L.V. Havryliuk et al. (2022), who substantiated the mechanisms of formation of stable microbial associations in the root zone of plants, which increases their tolerance to phytopathogens. For the first time, the effectiveness of biologically based crop cultivation technologies in different soil and climatic conditions of Ukraine was assessed, which enables the adaptation of microbial consortia to concrete agroecosystems, as demonstrated by O.A. Kovalenko (2021).

V. Sharma et al. (2020) substantiated the role of beneficial microorganisms in stimulating plant growth and protecting them from abiotic stresses, specifically by regulating genetic signalling pathways. The researchers were the first to detail the effects of microbial consortia on the expression of genes responsible for stress adaptation. The analysis of meta-omics studies by B.S. Adel- eke and O.O. Babalola (2022) showed that endophytic

microorganisms form stable associations with plants, improving their nutrition and resistance to pathogens. This study identified genes that determine the effectiveness of microbial interactions in host plants. A significant addition to this topic was the study by M. Fomina and I. Skorochod (2020), which established the mechanisms of interaction between microorganisms and clay minerals, which affects the dynamics of trace elements in the soil. These findings are valuable for the development of biological products with improved adsorption properties that provide a longer-lasting effect in the soil environment.

In addition, studies by N. Nikonchuk and M. Samoilenco (2024) revealed that the use of biological preparations during the growing season of tomatoes contributed to an increase in the above-ground mass and a decrease in the percentage of roots to the total mass of the studied varieties, in addition, the effect of biological preparations on the yield of the studied tomato varieties was proven. P. Vidhyasekaran (2024) proposed innovative approaches to improving plant resistance to bacterial pathogens, showing that the use of biotechnologically modified microbial preparations can activate the innate immunity of plants. This opens prospects for the development of biological products with targeted effects on the signalling molecular cascades of defence. S. Dymytrov and V. Sabluk (2023) introduced a new approach to the biological correction of agro-photosynthetic productivity through the optimisation of microbial processes in the rhizosphere, which confirmed the significance of symbiotic relationships in improving plant metabolism. X. Wei *et al.* (2024) obtained essential data on the use of microbial fertilisers in improving soil health and plant resistance to adverse conditions. This study confirmed the mechanisms that contribute to the development of a favourable microbial environment in the soil, ensuring the long-term effect of the use of biological products in agriculture.

Despite active research in this area, the mechanisms of interspecies interaction of microorganisms in soil communities, which determines the effectiveness of biological products in natural conditions, is still understudied. The purpose of this study was to systematise molecular-genetic and biotechnological approaches to the development of complex microbial preparations for balanced plant nutrition.

MATERIALS AND METHODS

In the course of the theoretical and analytical study, a set of general scientific methods was employed to facilitate a structured analysis of molecular-genetic and biotechnological approaches to the development of microbial preparations for balanced plant nutrition. The use of these methods ensured a comprehensive study of the processes associated with microbial interactions in the rhizosphere, as well as the possibilities of their practical application in agriculture. All generalisations

were made solely based on scientific publications, without the involvement of official statistical or information platforms. The use of the analysis method helped to investigate the molecular genetic mechanisms of microorganisms in soil ecosystems. The impact of bacterial signalling pathways on the expression of genes responsible for nitrogen fixation, phosphate mobilisation, and phytohormone synthesis was examined. The literature review also helped to assess the relationship between the species composition of microbial preparations and their functional efficiency under soil and climatic conditions, including the level of bacterial adaptation to environmental variables. The genetic analysis assessed the level of expression of stress-associated genes (*dnaK*, *groEL*) responsible for molecular chaperonin support and heat shock adaptation of microorganisms. RT-qPCR with specific primers for the corresponding operons was used for quantitative assessment.

Based on the analysed literature, the study synthesised the data on the specific features of microbial preparations functioning in the rhizosphere, including the patterns of their colonisation, metabolic activity, and interspecific interaction, which form the basis of their agronomic efficiency. This helped to identify the principles of their development, interaction with plant root systems, and potential efficiency in various soil and climatic conditions. Additionally, information on the mechanisms of interaction between microorganisms in biological products was processed, which helped to combine the available information into a single theoretical framework. To develop this model, the study used sources published mainly in 2019–2025, covering the findings of research conducted in the United States, China, the United Kingdom, the Netherlands, France, Nigeria, and Ukraine, which ensured a wide geographical and methodological range of scientific approaches. Induction and deduction methods were employed to logically substantiate the relationship between the physiological and biochemical characteristics of microorganisms and their ability to form stable biological products. The inductive approach helped to compare data from individual experimental studies and formulate general statements about the role of microbial consortia in improving plant mineral nutrition. The deductive method was used to test these statements in the context of known molecular mechanisms of interaction between microorganisms and plants.

The comparison method helped to evaluate the features of distinct approaches to the development of microbial biological products, considering their effectiveness, resistance to external factors, and interaction between strains in consortia. The analysis was conducted at the level of functional groups of bacteria without focusing on commercial products. The functional activity of bacteria expressing nitrogen fixation (*nif*) genes in *Azotobacter*, *Rhizobium*, *Bradyrhizobium*, phosphate mobilisation (*ppk*, *pho*) in *Bacillus*,

Pseudomonas, *Burkholderia*, and phytohormone biosynthesis (*ipd*, *tdc*) in *Pseudomonas*, *Enterobacter*, *Bacillus* was analysed. The selection of strains and target genes was based on the analysis of literature sources, where these microorganisms demonstrated the highest activity in the relevant metabolic processes, the ability to colonise the rhizosphere stably, and adaptability to various soil conditions. The ability of microorganisms to colonise the rhizosphere, form biofilms, and adapt to changing environmental conditions was assessed through the expression of regulatory genes *luxR-luxI* (Quorum Sensing) in *Sinorhizobium*, *Pseudomonas*, *Bacillus*, *rpoN* (nitrogen metabolism) in *Rhizobium*, *Azotobacter*, *Pseudomonas*, and *phoB* (phosphate metabolism) in *Pseudomonas*, *Bacillus*, *Burkholderia*. The comparison helped to analyse the differences between biological products containing single strains of microorganisms and complex consortia, as well as to assess the effects of different formulations of biological products (liquid, granular, microencapsulated) on their stability and efficacy in the field.

The generalisation method was employed to identify the key regularities that explain the functioning of microbial biological products in the soil environment. Particular attention was paid to the processes of rhizosphere colonisation, interspecific competitive interactions of microorganisms, and their ability to synthesise biologically active compounds that determine the effectiveness of consortia. Based on the analysis of these factors, a unified conceptual model describing the integrated effect of microbial preparations in agroecosystems was formed. The potential of genetic technologies, particularly CRISPR-Cas, for targeted modification of metabolic pathways responsible for the synthesis of phytohormones, rhizosphere colonisation, and adaptation to external stressors was analysed separately. The systematisation method was applied to classify modern approaches to the improvement of microbial biological products. This approach analysed genetic, physiological, biochemical, and technological strategies for improving the viability of microorganisms, including encapsulation, freeze-drying, and the use of nanostructured carriers that ensure the stability of consortia in the field. This systematisation helped to substantiate the practical feasibility of each approach depending on the type of crop, soil and climatic conditions, and the target functionality of the preparation.

LibreOffice Calc software (version 7.6) with basic functions of filtering, sorting, and structuring information according to the purpose of this study was used to process literature data, create tables, and logically classify microbiological traits. The integrated application of

these methods provided a comprehensive analysis of the mechanisms of action of microbial biological products, an assessment of their effectiveness in improving nutrient absorption and the development of recommendations for optimising their composition.

RESULTS

Genetic mechanisms of microorganisms functioning in the plant rhizosphere. The functional activity of rhizosphere microorganisms is regulated by complex interactions between genes that control biological nitrogen fixation, phosphate mobilisation, phytohormone synthesis, and adaptation to environmental conditions. The severity of these processes depends on the level of expression of the corresponding genes and the activity of regulatory signalling pathways. It was found that plant root exudates can modulate the expression of key functional genes, changing the metabolic status of bacterial populations (Liu et al., 2022; Horizon Europe, n.d.). The analysis of the expression of *nif* genes responsible for nitrogen fixation showed that their activity is most pronounced in *Rhizobium* and *Azotobacter*, which is explained by their specialisation in biological assimilation of atmospheric nitrogen. In *Rhizobium*, the level of *nif* expression was 1.8 relative units, while in *Azotobacter* it was 1.2, which is consistent with their adaptation to legumes, where they form nodules, and nitrogen-deficient conditions for free-living forms (Hakim et al., 2021).

Phosphate mobilisation in the rhizosphere is controlled by the *pho* and *ppk* genes, which regulate the bioavailability of phosphate compounds. The high activity of *pho* genes in *Pseudomonas* and *Bacillus* indicates their ability to dissolve inorganic phosphorus through the synthesis of phosphatases. In *Pseudomonas*, *pho* expression reached 1.7 units, while *ppk* – 1.5, in *Bacillus* – 1.3 and 1.2, respectively, indicating a strong potential for phosphate mobilisation. In *Burkholderia*, the expression of *ppk* reached 1.0, which confirmed its value in polyphosphate accumulation and long-term nutrition (Kong & Liu, 2022). The synthesis of phytohormones, particularly auxins and cytokinins, is regulated by the *ipd* and *tdc* genes. The greatest levels of expression of these genes were observed in *Pseudomonas* (1.8 for *ipd*, 1.6 for *tdc*) and *Bacillus* (1.4 for *ipd*, 1.5 for *tdc*), which indicated their effectiveness in stimulating plant growth. Studies found that *ipd* expression in *Pseudomonas putida* leads to the accumulation of indole-3-acetic acid, which regulates the development of the root system. The pronounced activity of *tdc* in *Bacillus* confirmed its potential in the synthesis of cytokinins that promote cell proliferation (Table 1).

Table 1. Expression level of key genes in different rhizosphere microorganisms (relative units)

Gene group	Function	Type of microorganisms
Nitrogen fixation genes (<i>nif</i>)	Fixation of atmospheric nitrogen and formation of ammonium compounds	Azotobacter, Rhizobium, Bradyrhizobium

Table 1. Continued

Gene group	Function	Type of microorganisms
Phosphate mobilisation genes (ppk, pho)	Dissolution of inorganic phosphorus and increased availability	Bacillus, Pseudomonas, Burkholderia
Genes for phytohormone biosynthesis (ipd, tdc)	Synthesis of auxins, cytokinins, and other phytohormones	Pseudomonas, Enterobacter, Bacillus
Genes of Quorum Sensing signalling pathways (luxR-luxI)	Regulation of metabolic gene expression in response to the environment	Sinorhizobium, Pseudomonas, Bacillus
Genes for the regulation of nitrogen metabolism (rpoN)	Control over the expression of nitrogen fixation and biofilm formation genes	Rhizobium, Azotobacter, Pseudomonas
Genes regulating phosphate metabolism (phoB)	Regulation of phosphate metabolism and rhizosphere colonisation	Pseudomonas, Bacillus, Burkholderia

Source: compiled by the authors based on S. Hakim et al. (2021), V.V. Gupta and A.K. Sharma (2021), Z. Kong and H. Liu (2022)

The analysis of the structural distribution of key genes among rhizosphere microorganisms revealed the presence of species specialisation in functional areas. Specifically, the highest activity of *nif* genes related to atmospheric nitrogen fixation was found in *Rhizobium* and *Azotobacter*, which are the leading symbiotic and free-living nitrogen fixers, respectively. *Pseudomonas* and *Bacillus* dominate in the mobilisation of hard-to-reach phosphorus, as they can express the *pho* and *ppk* genes responsible for the dissolution of inorganic phosphates and the accumulation of polyphosphates. *Pseudomonas* and *Bacillus*, which contain the *ipd* and *tdc* genes, are considered to be the key organisms in the synthesis of auxin and cytokinin phytohormones. The greatest level of regulatory activity of the *luxR-luxI* signalling complex responsible for Quorum Sensing mechanisms was found in *Sinorhizobium* and *Pseudomonas*. The *rpoN* gene, which regulates the expression of metabolic pathways in response to nitrogen deficiency, is best represented in *Rhizobium* and *Pseudomonas*, while the *phoB* gene, responsible

for rhizosphere colonisation and adaptation to phosphate deficiency, was found in *Pseudomonas*, *Bacillus*, and *Burkholderia*. These observations indicate the predominance of *Pseudomonas* and *Bacillus* as universal candidates for the development of multifunctional biological products with predictable effects.

The regulation of the expression of these genes is controlled by global signalling systems, including *luxR-luxI*, *rpoN*, and *phoB*. The *lux* gene was activated at 1.4 in *Pseudomonas* and 1.1 in *Bacillus*, demonstrating the ability to coordinate metabolism through Quorum Sensing. The *luxR-luxI* signalling pathway associated with the Quorum Sensing mechanism regulates biofilm formation and coordinates the expression of metabolic genes in response to the population density of microorganisms. The *rpoN* gene is the primary regulator of nitrogen metabolism and is activated in *Rhizobium* and *Azotobacter* during nitrogen deficiency. The expression of *phoB* in *Pseudomonas* and *Bacillus* correlates with the availability of phosphate in the environment, which allows these bacteria to effectively adapt to soil conditions (Fig. 1).

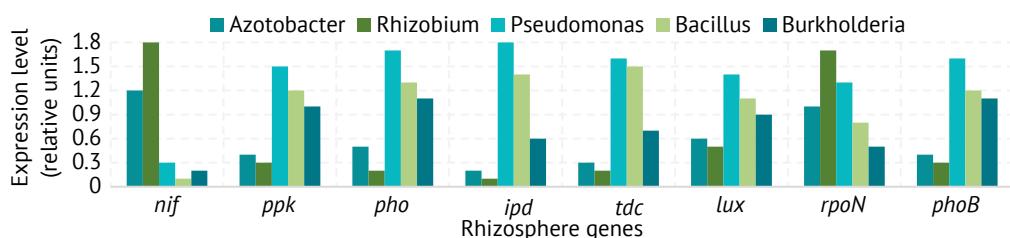


Figure 1. Expression level of key genes in rhizosphere microorganisms (relative units)

Source: compiled by the authors based on J. Ge et al. (2023), Q. Saeed et al. (2021)

The analysis of gene expression levels in different microorganisms indicated differential functional activity of rhizosphere bacteria depending on their ecological specialisation. The greatest level of expression of *nif* genes responsible for biological fixation of atmospheric nitrogen with the development of ammonium was found in *Rhizobium* (1.8 units) and *Azotobacter* (1.2 units), which indicates their ability to effectively provide plants with nitrogen, especially in legumes (for *Rhizobium*) and non-legumes (for *Azotobacter*).

The maximum activity of *pho* genes, which encode phosphatases for dissolving inorganic phosphorus, was recorded in *Pseudomonas* (1.7 units) and *Bacillus* (1.3 units), which ensures increased bioavailability of phosphate in deficient conditions. The *ppk* gene, which regulates polyphosphate metabolism, was the most active in *Pseudomonas* (1.5 units), reflecting the ability to accumulate phosphate reserves for a long time. In the group of phytohormone-associated genes, the expression of *ipd* (indole-3-acetic acid synthesis) in *Pseu-*

domonas reached 1.8 units, while *tdc* (cytokinin pathway) in *Bacillus* reached 1.6 units, which explains the high biostimulatory activity of these strains. The regulatory gene *lux*, which coordinates the expression of other genes in response to quorum sensing, had the greatest activity in *Pseudomonas* (1.4 units), indicating the ability of these bacteria to communicate effectively within consortia. The *rpoN* gene, which is critical for nitrogen metabolism and biofilm formation, showed the greatest expression in *Rhizobium* (1.7 units) and *Pseudomonas* (1.3 units), indicating their resistance to stressful conditions. For the *phoB* gene, which regulates phosphate metabolism, the leading position belonged to *Pseudomonas* (1.6 units), which allows these microorganisms to respond quickly to phosphorus deficiency in the soil.

Root exudates directly affect the expression of bacterial genes. It was proved that in the presence of exudates rich in organic acids, the activity of *pho* genes in *Pseudomonas* increases 3–4 times, which helps to improve phosphorus nutrition of plants. Analogously, phenolic compounds stimulate the expression of *luxR-luxI* in *Sinorhizobium*, which increases the level of colonisation of the root zone (Saeed et al., 2021). The analysis of the data shows that the functional activity of microorganisms in the rhizosphere is determined by the interaction of genetic mechanisms, regulatory pathways, and environmental factors. It was found that the combination of bacteria with different metabolic profiles allows creating more stable microbial consortia. The genetic specialisation of microorganisms in the rhizosphere is of key significance for the development of effective biological products that optimise plant nutrition in different agroecosystems.

Biotechnological approaches to the creation of complex microbial consortia. The development of effective biological products is based on the combination of microorganisms with varying functional characteristics, optimisation of cultivation and stabilisation methods, and the use of modern genetic technologies to improve their properties. Complex microbial consortia demonstrate an advantage over monocultures due to the synergistic effect of distinct groups of microorganisms, which increases their adaptability to soil conditions and the stability of the biological product (Food and Agriculture Organization, 2025). The functional specialisation of bacteria determines their role in biological products. Nitrogen fixers (*Rhizobium*, *Azotobacter*, *Bradyrhizobium*) provide biological fixation of atmospheric nitrogen through the expression of *nif* genes, which is critical for balanced plant nutrition. Phosphate mobilisers (*Pseudomonas*,

Bacillus, *Burkholderia*) expressing *pho* and *ppk* genes contribute to the dissolution of poorly available phosphate compounds, increasing the level of available phosphorus. Microorganisms capable of producing biologically active substances (*Pseudomonas*, *Enterobacter*, *Bacillus*) synthesise auxins and cytokinins using *ipd* and *tdc* genes, which stimulates the development of the root system. The optimum interaction between these groups of bacteria ensures the increased effectiveness of biological products in agroecosystems (Kapoore et al., 2021).

Various methods of cultivation and stabilisation are used to maintain the viability of microorganisms and increase their stability in biological products. One of the most effective approaches is microencapsulation, which involves coating microorganisms with a protective polymeric shell. This technology protects bacteria from unfavourable environmental factors, increases their survival in the soil, and promotes their even distribution in agroecosystems. Encapsulation in biopolymer matrices markedly improves the colonisation of the rhizosphere by microorganisms, which was confirmed by experimental studies (Saber-Riseh et al., 2021). An alternative method of stabilisation is lyophilisation, which allows moisture to be removed by freezing in a vacuum environment. This ensures the long-term preservation of bacterial activity without loss of functionality after reconstitution in an aqueous medium. The addition of cryoprotectants, such as polyethylene glycol and sucrose, improves the survival of microorganisms after drying and long-term storage (Balla et al., 2022). At the stage of mass production of biological products, fermentation cultivation is used to ensure high productivity of bacterial strains under controlled conditions. This method allows producing large volumes of microorganisms with predictable biological properties.

Spray drying is used to increase the shelf life of the products after cultivation, which allows producing the products in powder form with high stability (Ismanzhanov & Tashiev, 2016). Another effective technology is granulation, which involves the incorporation of bacteria into solid carriers, which protects them from degradation and ensures their gradual release into the soil. Granulated biological products are easy to use and ensure high survival of microorganisms in natural conditions. One of the most promising areas of microbial stabilisation is to stimulate the development of biofilms, which increases their resistance to external factors and promotes effective colonisation of the rhizosphere. Bacteria that form biofilms demonstrate better competitiveness and a greater level of biological activity (Table 2).

Table 2. Methods of cultivation and stabilisation of microorganisms in biological products

Method	Working principle	Advantages	Limitations
Microincapsulation	Coating of microorganisms with a protective polymer layer	Protection against stress factors, prolonged activity	High cost, need for special equipment
Lyophilisation	Moisture removal by freezing and vacuum drying	Long-term preservation of bacterial viability	Requires cryoprotectants, possible loss of activity

Table 2. Continued

Method	Working principle	Advantages	Limitations
Fermentation cultivation	Mass cultivation of bacteria in a liquid medium	High performance, controlled conditions	Requires sophisticated equipment, high energy consumption
Spray drying	Conversion of bacterial suspension into dry powder	Long-term storage, easy transport	Loss of some viable cells
Granulation	Production of dry granules with incorporated microorganisms	Easy to apply to the soil, stable in storage	Limited choice of media, risk of reduced activity
Aggregation in biofilms	Stimulation of bacterial biofilm development	Protection against adverse conditions, synergy between strains	Difficult to control in a production environment

Source: compiled by the authors based on N.S. McCarty and R. Ledesma-Amaro (2018), S. Rai and R. Prasad (2023)

The analysis of the above methods of cultivation and stabilisation of microorganisms shows that the choice of approach depends on the specifics of the biological product, its storage conditions, and application features. Microencapsulation and lyophilisation are the most effective methods for preserving the viability of bacteria in commercial biological products. Enzymatic cultivation and spray drying provide high production efficiency, although they may be accompanied by a loss of bacterial viability. Granular biological products are convenient to use, but their effectiveness depends on the composition of the carrier. The aggregation of bacteria in biofilms is a promising area that increases the adaptability of microorganisms, although its large-scale application is limited by technological difficulties.

Modern biotechnological approaches involve the use of genetic engineering methods, such as CRISPR-Cas, to modify microorganisms and increase their effectiveness in biological products. Editing the *nif* genes in *Azotobacter vinelandii* allows increasing the nitrogen fixation, which helps to improve nitrogen nutrition of plants. The use of CRISPR-Cas also opens opportunities to regulate the expression of *pho* genes in *Pseudomonas fluorescens*, which enhances the bacteria's ability to mobilise phosphorus in the soil. The genetic improvement of microorganisms is also aimed at increasing their resistance to stress factors, particularly by modifying *luxR-luxI* in *Bacillus subtilis*, which improves biofilm formation and bacterial survival in the field (Gholizadeh *et al.*, 2020). The combination of various bacterial strains in consortia, optimisation of their preservation methods, and the use of modern molecular genetic technologies allow developing effective biological products that can adapt to diverse soil and climatic conditions. The development of synthetic biology also opens opportunities to create synthetic microbial communities characterised by specified properties that can be used to optimise biological processes in the rhizosphere. The use of the latest biotechnologies in the development of microbial preparations can markedly increase their effectiveness, which is a promising area for the development of sustainable agriculture.

Effect of microbial preparations on nutrient uptake by plants. The efficiency of nutrient uptake by plants largely depends on the activity of the rhizosphere

microbiota, which affects the bioavailability of macro- and microelements, modifies soil chemical properties, and creates optimised conditions for plant growth. The use of microbial-based biological products helps to improve mineral nutrition of plants through biochemical and physiological mechanisms that ensure the mobilisation of nutrients in the rhizosphere. Studies confirmed that the use of microbial consortia on cereals, legumes and vegetables substantially reduces the need for chemical fertilisers without losing agronomic efficiency. Specifically, in the field conditions, the use of stabilised strains of *Azospirillum brasiliense* and *Pseudomonas fluorescens* for wheat (*Triticum aestivum*) and soybeans (*Glycine max*) reduced the rate of nitrogen and phosphorus fertiliser application by 25%. On tomatoes (*Solanum lycopersicum*) and cucumbers (*Cucumis sativus*) in greenhouse conditions, the consortium of *Bacillus subtilis* + *Burkholderia cepacia* increased phosphorus uptake by 30% and reduced the need for phosphate fertilisers by 20-25%. For legumes, specifically *Vicia faba*, the use of *Rhizobium leguminosarum* in central black soil provided full coverage of nitrogen fixation needs without supplementing with nitrogen compounds (Das *et al.*, 2022).

One of the key mechanisms for increasing the bioavailability of macro- and microelements is biological nitrogen fixation. Nitrogen-fixing bacteria (*Rhizobium*, *Azotobacter*, *Bradyrhizobium*) express *nif* genes that ensure the conversion of atmospheric nitrogen into forms available for plant uptake. Symbiotic strains of *Rhizobium* that interact with legumes form nodule structures that fix N₂, which markedly reduces the need for chemical nitrogen fertilisers. For non-legumes (wheat, maize, rice), the use of associative nitrogen fixers, such as *Azospirillum brasiliense*, increases the concentration of ammonium compounds in the rhizosphere by up to 40% (Etesami & Adl, 2020). Another essential mechanism is the dissolution of inorganic phosphorus and its mobilisation. Some strains of *Pseudomonas*, *Bacillus*, and *Burkholderia* produce organic acids (gluconic, citric, oxalic) that convert phosphates into a water-soluble form, increasing their availability in the soil. The high expression of *pho* and *ppk* genes in these microorganisms correlates with an increase in the concentration of available phosphorus in the root nutrition zone. The combined use of phosphate-mobilising microorganisms

with mineral fertilisers increases the efficiency of the latter, allowing the rate of phosphorus compounds application to be reduced by 15-25% without losing their availability to plants (Das et al., 2022).

Apart from macronutrients, microorganisms also affect the availability of trace elements such as iron (Fe), zinc (Zn), and manganese (Mn). Some bacteria (*Pseudomonas*, *Enterobacter*) synthesise siderophores, low-molecular-weight compounds that chelate Fe^{3+} ions, converting them into a form accessible to plants. This is crucial for crops growing on carbonate soils with low levels of available iron. The use of siderophore-forming bacteria in crops such as *Zea mays* and *Oryza sativa* can reduce chlorosis symptoms by 30-40%

(Zulfiqar et al., 2019). Microbial consortia also change the structural and functional characteristics of the rhizosphere, which contributes to improved nutrient uptake. The production of exopolysaccharides by bacteria (*Bacillus*, *Sinorhizobium*) contributes to the development of biofilms that regulate the water balance in the soil and protect the root system from stress. Furthermore, microbial biopreparations can increase the permeability of root membranes, stimulating the expression of genes responsible for the transport of nutrients into the plant (Mustafayeva et al., 2011). For example, the treatment of wheat seeds with *Pseudomonas* and *Bacillus* consortia helps to intensify root growth and increase water and nutrient uptake (Table 3).

Table 3. Effect of microbial preparations on nutrient absorption

Group of microorganisms	Function	Resulting effect on plants
Rhizobium, Azotobacter, Bradyrhizobium	Fixation of atmospheric nitrogen (nif genes)	Increased nitrogen levels in the soil, reduced fertiliser requirements
Pseudomonas, Bacillus, Burkholderia	Phosphate dissolution (pho, ppk genes)	Increased phosphorus availability, stimulation of root growth
Pseudomonas, Enterobacter	Synthesis of siderophores for Fe^{3+} chelation	Improved iron absorption, increased resistance to chlorosis
Bacillus, Sinorhizobium	Production of exopolysaccharides, biofilm development	Protection of the root system, improvement of the water regime

Source: compiled by the authors based on F. Zulfiqar et al. (2019), H. Etesami and S.M. Adl (2020), B. Hamid et al. (2021)

A comparative analysis of the effectiveness of distinct groups of microorganisms shows that biological products that combine nitrogen fixers, phosphate mobilisers, and sulphur-forming bacteria have the most comprehensive effects on plant nutrition. The combination of these groups not only reduces dependence on fertilisers but also improves the structure of the rhizosphere and increases plant resistance to adverse conditions. Specifically, nitrogen fixers interact best with phosphate mobilisers, as the application of biological products combining *Rhizobium* and *Pseudomonas* helps to increase the content of available nitrogen and phosphorus in the soil. At the same time, competition between distinct species of microorganisms for nutrients can affect the stability of the biological product. Genetic editing of microorganisms using CRISPR-Cas opens new opportunities to optimise their interactions and improve the effectiveness of biological products.

The use of modern biotechnological products in agriculture helps reduce dependence on chemical fertilisers while supporting the sustainable development of agroecosystems (Bolokhovsky et al., 2024). Combined microbial consortia, including nitrogen fixers, phosphate mobilisers, and bacteria that produce biologically active compounds, help improve plant nutrient uptake and increase yields without significantly depleting soil resources. The inclusion of microorganisms capable of developing biofilms and synthesising siderophores in biological products increases their effectiveness in various soil and climatic conditions. The use of these

technologies in agriculture helps to increase plant productivity and provides environmentally friendly mineral nutrition management strategies.

Optimisation of the composition of microbial preparations to increase efficiency. The effectiveness of microbial biopreparations is determined by the ability of microorganisms to adapt to soil conditions, their competitiveness in the rhizosphere, and the interaction between species within consortia. Optimisation of the composition of biological products is aimed at selecting microorganisms capable of stable colonisation of the root zone, efficient use of available resources, and synergistic interaction to improve plant nutrition. One of the key factors that determines the competitiveness of microorganisms in soil is their ability to quickly adapt to changing environmental conditions, including humidity, pH, macro- and microelements, as well as competition with autochthonous microflora. Studies found that bacterial strains with prominent levels of *rpoN* and *phoB* gene expression demonstrate a better ability to colonise the rhizosphere, as they regulate the synthesis of exopolysaccharides and provide resistance to soil stress factors (Sorbara & Pamer, 2022). Another valuable characteristic of microorganisms is the efficiency of their movement in the soil, which is determined by cell size, hydrophobicity of surface proteins, and the ability to actively chemotaxis. For instance, *Pseudomonas* bacteria, due to their high mobility, reach the root zone faster and have an advantage in competition for nutrients compared to less mobile *Bacillus*

strains (Gammack *et al.*, 2021). In the composition of biological products, special attention is paid to the interaction between microorganisms – both synergistic and antagonistic effects. Some bacteria form mutually beneficial associations, for instance, the combination of *Rhizobium* (nitrogen fixers) and *Pseudomonas* (phosphate mobilisers) helps to increase the availability of nutrients for plants. At the same time, antagonistic interactions between antibiotic-producing bacteria (*Streptomyces*, *Bacillus*) and phytopathogens can increase the bioprotective properties of biological products (Philippot *et al.*, 2023).

Antagonism in microbial consortia can also negatively affect the effectiveness of biological products. For example, *Pseudomonas* bacteria produce toxic metabolites that can inhibit the growth of some beneficial microorganisms, which limits the possibility of their combination in consortia. Controlling antagonistic effects is a crucial task when formulating biological products and can be achieved by selecting strains with minimal negative interactions (Peterson *et al.*, 2020). Strategies

for adapting biological products to diverse soil and climatic conditions include the selection of strains that are resistant to extreme environmental factors, such as drought, salinity, and high acidity. For example, the use of heat- and osmosis-resistant strains of *Bacillus* and *Azospirillum* can increase the effectiveness of biological products in regions with a lack of moisture. Studies show that modification of stress-associated gene expression factors, such as *dnaK* and *groEL*, can increase the survival of microorganisms under adverse conditions (Malik *et al.*, 2019). The use of biological products in different soil types requires adaptation of their composition. In chernozems with a high humus content, beneficial microorganisms colonise the rhizosphere more effectively, while in sandy and infertile soils, the stability of microbial preparations depends on the addition of biopolymer carriers that protect bacteria from dehydration. The application of microorganisms in such soils is often combined with organic fertilisers, which increases the survival rate of bioagents and the efficiency of nutrient uptake (Table 4).

Table 4. Factors affecting the effectiveness of microbial preparations

Factor	Impact on the effectiveness of biological products	Examples of microorganisms
Competitiveness	Regulates the ability to colonise the rhizosphere	<i>Pseudomonas</i> , <i>Bacillus</i> , <i>Azotobacter</i>
Mobility in soil	Determines the speed of reaching the root zone	<i>Pseudomonas</i> , <i>Rhizobium</i>
Interaction of microorganisms	Synergy or antagonism between species in consortia	<i>Rhizobium</i> + <i>Pseudomonas</i> (synergy), <i>Pseudomonas</i> + <i>Bacillus</i> (an antagonism)
Stress resistance	Ability to survive in adverse conditions	<i>Bacillus</i> , <i>Azospirillum</i>
Soil conditions	Affects the stability of microorganisms	Black soils: high colonisation, sandy soils: dependence on carriers

Source: compiled by the authors based on A.A. Malik *et al.* (2019), S.B. Peterson *et al.* (2020)

The data analysis shows that the combination of microorganisms with complementary functions ensures stable colonisation of the root zone and maximum use of available nutrients. The use of strains with high mobility in the soil, resistance to stress factors, and minimal antagonistic effects is a crucial criterion for the development of biological products. Optimisation of the composition of biological products may also include the use of genetically modified strains with enhanced expression of genes responsible for stress resistance and rhizosphere colonisation. For example, modification of the *rpoN* gene in *Pseudomonas fluorescens* can increase the level of biofilm development, which improves the bacteria's competitiveness in soil conditions. Further improvement of biopreparations involves the selection of microbial consortia based on soil and climatic characteristics and the potential for interaction between strains. The use of adapted microbial preparations helps to increase the efficiency of nutrient uptake,

reduce dependence on chemical fertilisers, and improve the environmental sustainability of agricultural systems. The development of new consortia that combine microorganisms with different functional characteristics is a key approach to creating highly effective biological products for agricultural production.

Environmental aspects of the use of microbial preparations and environmental protection. The use of microbial biological products in modern agriculture contributes to the preservation of soil biodiversity, reduces dependence on mineral fertilisers, and improves crop adaptation to changing environmental conditions. Due to their ability to improve the physical and chemical properties of soil and maintain the stability of agroecosystems, biological products are considered a promising tool for increasing the sustainability of agricultural technologies (Organisation for Economic Co-operation and Development, 2019; Horizon Europe, n.d.). One of the major environmental effects of

biological products is their influence on the biodiversity of soil microbial communities. The introduction of beneficial bacteria helps to increase the number of microorganisms that perform important ecosystem functions, including decomposition of organic matter, nitrogen fixation, and mobilisation of mineral compounds (Kuts *et al.*, 2023). For instance, the use of *Bacillus subtilis* and *Pseudomonas fluorescens* stimulates the development of microorganisms that produce enzymes to decompose organic residues, improving humus development and soil structure. At the same time, excessive use of biological products can cause the displacement of autochthonous microbiota, which can alter the ecological balance of soil microorganisms and cause effects analogous to those of intensive agrochemical use (Elnahal *et al.*, 2022).

Another significant aspect is the reduced need for mineral fertilisers due to the ability of microorganisms to improve nutrient uptake. Nitrogen fixers (*Rhizobium*, *Azotobacter*, *Bradyrhizobium*) provide fixation of atmospheric nitrogen, which reduces the need for synthetic nitrogen fertilisers. Phosphate-mobilising bacteria (*Pseudomonas*, *Bacillus*, *Burkholderia*) help convert insoluble forms of phosphorus into a form available to plants, which reduces the use of phosphate fertilisers by 25-40% without losing crop productivity. This not only reduces farmers' costs but also minimises water pollution and prevents eutrophication processes that occur when mineral fertilisers are overused (Castiglione *et al.*, 2021). Microbial biopreparations also reduce the environmental impact of agriculture, as they do not accumulate in the soil and do not cause long-term degradation of soil ecosystems. Unlike mineral fertilisers, which can leach out of the soil and into groundwater, microorganisms can develop stable colonies in the rhizosphere, providing a prolonged effect. The use of microbial consortia enables a long-lasting effect of improved nutrient uptake, which ensures a more balanced use of resources (Kosovska *et al.*, 2022).

Practical aspects of introducing biological products into modern agricultural technologies include the selection of optimum application methods, adaptation of the composition of biological products to specific soil and climatic conditions, and introduction of microbial products into integrated fertilisation systems. In practice, biological products can be applied by treating seeds, inoculating the root system, or applying them to the soil as part of tank mixtures, which enables the most efficient use of their effect (Zakharchuk *et al.*, 2019). Studies show that the use of biological products in combination with organic fertilisers can increase crop yields by 15-30% by improving mineral nutrition and stimulating root development (Bonaterra *et al.*, 2022). Another essential element of the successful introduction of biological products is cost-effectiveness. Initial costs for the production and use of biological products may be greater than for mineral fertilisers, but

their long-term effects, improved soil properties, and reduced need for repeated application compensate for these costs. The use of biological products in organic farming is becoming the primary method of ensuring effective mineral nutrition for plants, as it eliminates the use of synthetic fertilisers and pesticides. Organic producers are actively introducing microbial preparations as an alternative to chemical growth stimulants, which confirms their valuable role in the development of sustainable agriculture (Tiwari, 2022).

The widespread use of biological products also contributes to increasing the resistance of crops to stress factors. Inoculant bacteria can increase plant resistance to drought, salinity, and low temperatures through the synthesis of osmoprotectants, auxins, and polyamines. For example, consortia of *Bacillus* and *Pseudomonas* bacteria increase the drought resistance of wheat and soybeans, which helps reduce the negative impact of climate change on yields. The inclusion of microorganisms in agrobiotechnological crop management programmes is becoming an indispensable tool for increasing the adaptive potential of crops in changing environmental conditions (Kundius, 2021). The use of microbial biological products in agriculture helps to reduce the adverse environmental impact of agricultural production, improve soil microbiota, and increase the efficiency of nutrient uptake by plants. Their integration into agrotechnology allows creating more environmentally friendly and cost-effective fertilisation systems that ensure sustainable agricultural production and maintain a balance in agroecosystems.

DISCUSSION

The analysis of the obtained findings helped to assess the mechanisms of influence of microbial biological products on nutrient uptake and plant adaptation to soil conditions. The study found that the effectiveness of biological products is determined by the combination of functionally distinct microorganisms, their competitiveness, and ability to colonise the rhizosphere. Comparison with other studies allows identifying shared patterns and differences in the mechanisms of action of microbial consortia. The obtained findings confirmed the significant role of functional genes of microorganisms in improving nutrient uptake by plants and the formation of stable microbial consortia in the rhizosphere. The high level of *nif* gene expression in *Rhizobium* and *Azotobacter* contributed to the increase in the efficiency of biological nitrogen fixation, which is consistent with the findings of Q. Liu *et al.* (2022), who found a correlation between the activity of these genes and an increase in the production of ammonium compounds. Analysis of the expression of *ppk* and *pho* genes in *Pseudomonas* and *Bacillus* showed their key role in the mobilisation of inorganic phosphates, which was confirmed by S. Hakim *et al.* (2021), who described the intensive involvement of phosphate

pathways in microbial rhizosphere metabolism. The study of *ipd* and *tdc* genes in *Pseudomonas* demonstrated their valuable function in the synthesis of auxins and cytokinins, which is consistent with the results of Z. Kong and H. Liu (2022), who noted the significant impact of microbial phytohormone biosynthesis on plant adaptation to stressful conditions.

Regulatory mechanisms of microbial signalling pathways also played a crucial role in the stability of consortia in the rhizosphere. The study found that the expression of *rpoN* and *phoB* in *Pseudomonas* and *Rhizobium* contributed to the adaptation of microorganisms to changing environmental conditions and stimulated the development of stable microbial communities, which is consistent with the findings of J. Ge *et al.* (2023), who pointed out the criticality of regulatory pathways in ensuring the viability of microorganisms in soil. The combination of microbial strains with diverse functions in consortia improved complex nutrient uptake, which was supported by the findings of Q. Saeed *et al.* (2021), who reported that nitrogen fixers, phosphate mobilisers, and biostimulatory bacteria interact effectively to improve plant growth characteristics. The findings highlighted the value of an integrated approach to the development of microbial biological products, factoring in the molecular genetic characteristics of bacteria and their ecological interaction in soil ecosystems.

The findings demonstrated the significance of an integrated approach to the formation of microbial consortia in biological products. It was confirmed that the combination of nitrogen fixers, phosphate mobilisers, and producers of biologically active compounds provides a synergistic effect that increases the efficiency of nutrient uptake by plants. This is in line with the findings of R.V. Kapoore *et al.* (2021), who proved that the cultivation of microorganisms with different functional characteristics contributes to the development of stable microbial communities that adapt to soil conditions. The pronounced level of rhizosphere colonisation depended on the ability of bacteria to develop biofilms and interact through signalling molecules, which is consistent with the data from R. Saberi-Riseh *et al.* (2021), who described the positive effect of microencapsulated microorganisms on the stability of biological products. The present study also confirmed that the bioencapsulation of *Pseudomonas* and *Bacillus* bacteria contributed to their survival in adverse conditions and prolonged action in agroecosystems, which is consistent with the findings reported by A. Balla *et al.* (2022).

The optimisation of biological products also included the use of synthetic biology and genetic engineering methods to improve the properties of microorganisms. The use of CRISPR-Cas allowed modifying the expression of genes regulating nitrogen fixation, phosphorus mobilisation, and synthesis of signalling molecules, as confirmed by N.S. McCarty and R. Ledesma-Amaro (2018). Genetically modified strains demonstrated

increased competitiveness in the rhizosphere, providing more efficient nutrient uptake, which is consistent with the findings of S. Rai and R. Prasad (2023). Editing of *luxR-luxI* in *Sinorhizobium* and *Bacillus* increased the level of biofilm formation, ensuring stable soil colonisation, which was also confirmed in the study by P. Gholizadeh *et al.* (2020). Microbial biopreparations ensured efficient absorption of nutrients by plants due to the interaction of diverse groups of microorganisms. The use of nitrogen fixers, phosphate mobilisers, and biostimulating bacteria increased the bioavailability of macro- and microelements, as described by P.P. Das *et al.* (2022), who emphasised the significance of a three-way interaction between plants, soil, and microorganisms to improve mineral nutrition. As reported by H. Etesami and S.M. Adl (2020), rhizobacteria that stimulate plant growth can change the metabolism of the root system, activating the mechanisms of efficient nutrient uptake. The combination of biological products with mineral fertiliser nanoparticles ensured the gradual release of nutrients into the rhizosphere, which increased their effectiveness, as reported by F. Zulfiqar *et al.* (2019). Biostimulatory bacteria also played a significant role in increasing plant resistance to abiotic stresses through the synthesis of phytohormones and stimulation of root development. Analogous mechanisms were considered by B. Hamid *et al.* (2021), who pointed out the major impact of microbial consortia on plant adaptation to adverse conditions. The obtained findings confirmed the prospects of using microbial biological products to increase the efficiency of mineral nutrition and the productivity of agroecosystems.

Optimisation of the composition of microbial biological products required considering the competitiveness of microorganisms in soil conditions and their ability to effectively colonise the rhizosphere. M.T. Sorbara and E.G. Pamer (2022) confirmed that the successful adaptation of microorganisms to agroecosystems depends on their interaction with the autochthonous microbiota and the ability to form stable communities. It was found that the mobility of microorganisms in the soil, their penetration into the rhizosphere, and their fixation in the root zone were determined by the physical and chemical properties of the soil and the type of microbial interactions, as reported by S.M. Gam-mack *et al.* (2021). Apart from the physical barriers, microbial competition and antagonism played a significant role, affecting the survival of strains in biological products. L. Philippot *et al.* (2023) showed that the active functioning of microbial consortia depended on their interaction with soil minerals and the availability of organic material. A prominent aspect was also the ability of microorganisms to produce antimicrobial compounds, which allowed them to suppress competitive species, as reported by S.B. Peterson *et al.* (2020). Consideration of these factors in the development of biological products allowed increasing their

effectiveness by optimising the ratio between cooperative and antagonistic interactions in microbial consortia. A.A. Malik *et al.* (2019) also pointed to the significance of environmental specialisation of strains, which determined their ability to function for a long time in various soil and climatic conditions.

The use of microbial biological products in agriculture contributed not only to improving nutrient uptake but also to maintaining the ecological balance of agro-ecosystems. A.S.M. Elnahal *et al.* (2022) confirmed that biological inoculants play a key role in stimulating plant growth and protecting against phytopathogens, which reduces the need for synthetic fertilisers and protection products. The use of microbial biostimulants contributed to the accumulation of biologically active compounds that positively influenced plant metabolism, as described by A. Castiglione *et al.* (2021). The interaction of biological products with soil microbial communities was crucial for maintaining soil fertility. N. Kosovska *et al.* (2022) found that the use of biological products in combination with nanofertilisers changed the composition and functional activity of the microbiome, which ensured optimum plant nutrition.

The use of biological products in plant protection against pathogens was considered an effective method of reducing the chemical burden on agro-ecosystems. A. Bonaterra *et al.* (2022) confirmed that bacterial biological products can inhibit the growth of pathogens through the production of antimicrobial compounds and competition for nutrients. Additionally, the study found that the integration of biological products into organic production contributed to the efficiency of agro-technologies, which is consistent with the findings of V. Kundius (2021), who proved that biointensive technologies can markedly improve yields without harming the environment. The further development of organic farming is based on the introduction of biotechnological solutions that minimise the dependence on chemical fertilisers, as discussed in V. Kundius's study. The findings confirmed the pronounced efficiency of microbial biological products in increasing crop productivity and maintaining the ecological stability of agroecosystems. The study confirmed that the effectiveness of microbial biological products depends on their composition, properties of microorganisms, and soil conditions. The combination of nitrogen fixers, phosphate mobilisers, and biostimulants improved nutrient uptake and plant resistance. The use of genetic engineering and microencapsulation increased the viability of microorganisms and their effectiveness in various agroecosystems. The interaction of biological products with the rhizosphere microbiome ensured the development of stable consortia and improved soil fertility. The data obtained confirmed the prospects of improving the composition of biological products for their widespread use in sustainable agriculture.

CONCLUSIONS

The study summarised the mechanisms of action of microbial biological products on plant nutrition, establishing that their effectiveness depends on the composition of consortia, the level of expression of key genes, and the ability of microorganisms to colonise the rhizosphere in a stable manner. The study found that biological products containing nitrogen-fixing (*Rhizobium*, *Azotobacter*), phosphate-mobilising (*Pseudomonas*, *Bacillus*) and biostimulating microorganisms (*Burkholderia*, *Enterobacter*) contribute to a comprehensive improvement in the uptake of macro- and microelements by plants. The integration of microorganisms with distinct functional properties enables a synergistic effect that promotes more efficient nutrient uptake and increased plant stress resistance. Analysis of the genetic mechanisms of rhizosphere microorganisms confirmed that the effectiveness of microbial biological products is determined by the expression of genes that control nitrogen fixation (*nif*), phosphate mobilisation (*pho*, *ppk*) and phytohormone synthesis (*ipd*, *tdc*). The greatest activity of *nif* genes was observed in *Rhizobium* and *Azotobacter*, which was explained by their adaptation to nitrogen fixation, especially in symbiotic interactions with legumes. The study found that plant root exudates modulate the expression of functional genes, which affects the metabolic status of bacterial populations and their competitiveness in the rhizosphere. *Pseudomonas* and *Bacillus* demonstrated the greatest activity of *pho* and *ppk* genes, which contributed to the mobilisation of phosphorus and improved its bioavailability for plants.

The optimisation of biological products involved the use of microencapsulation and lyophilisation, which prolonged the viability of microorganisms and ensured their stability in soil conditions. The combination of microbial biological products with nanoparticles of mineral fertilisers contributed to the gradual release of nutrients, which improved their absorption and reduced the loss of elements to the environment. The study found that the integration of phosphate-mobilising bacteria *Pseudomonas fluorescens* with bioavailable phosphates increased the efficiency of phosphorus uptake, which is crucial for improving plant nutrition. The data obtained suggest that the combination of microbiological, genetic, and technological approaches enables the creation of more stable and effective biological products for crops.

The present study was theoretical and based on the generalisation of scientific data, which may limit its application in practical agricultural technologies. The findings obtained require experimental confirmation in real field conditions to assess the effectiveness of the proposed biological products in various agroecosystems. Further research is recommended to focus on the experimental verification of theoretical conclusions through field trials in diverse soil and climatic conditions. A prominent area is to evaluate the effectiveness

of microbial biological products in the long term, particularly their impact on the structure of the soil microbiome, plant productivity, and nutrient bioavailability. Additionally, it is advisable to investigate the genetic optimisation of microorganisms, specifically their competitiveness and resistance to stress factors, which will increase the stability and effectiveness of biological products in real agroecosystems.

ACKNOWLEDGEMENTS

None.

FUNDING

None.

CONFLICT OF INTEREST

None.

REFERENCES

- [1] Adeleke, B.S., & Babalola, O.O. (2022). Meta-omics of endophytic microbes in agricultural biotechnology. *Biocatalysis and Agricultural Biotechnology*, 42, article number 102332. [doi: 10.1016/j.bcab.2022.102332](https://doi.org/10.1016/j.bcab.2022.102332).
- [2] Balla, A., Silini, A., Cherif-Silini, H., Bouket, A.C., Alenezi, F.N., & Belbahri, L. (2022). Recent advances in encapsulation techniques of plant growth-promoting microorganisms and their prospects in the sustainable agriculture. *Applied Sciences*, 12(18), article number 9020. [doi: 10.3390/app12189020](https://doi.org/10.3390/app12189020).
- [3] Bolokhovsky, V., Bolokhovska, V., Khomenko, T., Datsko, A., & Litvinova, O. (2024). Optimisation of plant nutrition under the influence of biopreparations in integrated sunflower cultivation technologies. *Plant and Soil Science*, 15(4), 64-75. [doi: 10.31548/plant4.2024.64](https://doi.org/10.31548/plant4.2024.64).
- [4] Bonaterra, A., Badosa, E., Daranas, N., Francés, J., Roselló, G., & Montesinos, E. (2022). Bacteria as biological control agents of plant diseases. *Microorganisms*, 10(9), article number 1759. [doi: 10.3390/microorganisms10091759](https://doi.org/10.3390/microorganisms10091759).
- [5] Borko, Y., Bolokhovskyi, V., Datsko, A., Lungul, A., & Zhurba, M. (2025). Microbiological activity of soil and its impact on maize productivity when applying biologics. *Biological Systems: Theory and Innovation*, 16(1), 58-71. [doi: 10.31548/biologiya/1.2025.62](https://doi.org/10.31548/biologiya/1.2025.62).
- [6] Castiglione, A.M., Mannino, G., Contartese, V., Berte, C.M., & Ertani, A. (2021). Microbial biostimulants as response to modern agriculture needs: Composition, role and application of these innovative products. *Plants*, 10(8), article number 1533. [doi: 10.3390/plants10081533](https://doi.org/10.3390/plants10081533).
- [7] Das, P.P., Singh, K.R., Nagpure, G., Mansoori, A., Singh, R.P., Ghazi, I.A., Kumar, A., & Singh, J. (2022). Plant-soil-microbes: A tripartite interaction for nutrient acquisition and better plant growth for sustainable agricultural practices. *Environmental Research*, 214, article number 113821. [doi: 10.1016/j.envres.2022.113821](https://doi.org/10.1016/j.envres.2022.113821).
- [8] Doolotkeldieva, T., & Bobusheva, S. (2024). [Biofungicides and bioinoculants for sustainable agriculture: Rhizosphere streptomyces bacteria for protecting seeds and plants from phytopathogens and stimulating their growth](#). *Bulletin of the Kyrgyz National Agrarian University*, 22(5), 89-103.
- [9] Dymytrov, S., & Sabluk, V. (2023). Effectiveness of seed treatment with biopreparations for increasing soybean (*Glycine max* (L.) Merr.) productivity. *International Science Journal of Engineering & Agriculture*, 2(3), 67-81. [doi: 10.46299/ijisjea.20230203.07](https://doi.org/10.46299/ijisjea.20230203.07).
- [10] Elnahal, A.S.M., El-Saadony, M.T., Saad, A.M., Desoky, E.M., El-Tahan, A.M., Rady, M.M., AbuQamar, S.F., & El-Tarabily, K.A. (2022). The use of microbial inoculants for biological control, plant growth promotion, and sustainable agriculture: A review. *European Journal of Plant Pathology*, 162(4), 759-792. [doi: 10.1007/s10658-021-02393-7](https://doi.org/10.1007/s10658-021-02393-7).
- [11] Etesami, H., & Adl, S.M. (2020). Plant growth-promoting rhizobacteria (PGPR) and their action mechanisms in availability of nutrients to plants. In M. Kumar, V. Kumar & R. Prasad (Eds.), *Phyto-microbiome in stress regulation* (pp. 147-203). Singapore: Springer. [doi: 10.1007/978-981-15-2576-6_9](https://doi.org/10.1007/978-981-15-2576-6_9).
- [12] Fomina, M., & Skorochod, I. (2020). Microbial interaction with clay minerals and its environmental and biotechnological implications. *Minerals*, 10(10), article number 861. [doi: 10.3390/min10100861](https://doi.org/10.3390/min10100861).
- [13] Food and Agriculture Organization. (2025). *NSP – agriculture and soil biodiversity*. Retrieved from <https://www.fao.org/agriculture/crops/thematic-sitemap/theme/spi/soil-biodiversity/agriculture-and-soil-biodiversity/en/>.
- [14] Gammack, S.M., Paterson, E.R.I.C., Kemp, J.S., Cresser, M.S., & Killham, K. (2021). Factors affecting the movement of microorganisms in soils. In J.-M. Bollag & G. Stotzky (Eds.), *Soil biochemistry* (pp. 263-305). Bora Raton: CRC Press. [doi: 10.1201/9781003210207](https://doi.org/10.1201/9781003210207).
- [15] Ge, J., Li, D., Ding, J., Xiao, X., & Liang, Y. (2023). Microbial coexistence in the rhizosphere and the promotion of plant stress resistance: A review. *Environmental Research*, 222, article number 115298. [doi: 10.1016/j.envres.2023.115298](https://doi.org/10.1016/j.envres.2023.115298).
- [16] Gholizadeh, P., Köse, S., Dao, S., Ganbarov, K., Tanomand, A., Dal, T., Aghazadeh, M., Ghotaslou, R., Ahangarzadeh Rezaee, M., Yousefi, B., & Samadi Kafil, H. (2020). How CRISPR-cas system could be used to combat antimicrobial resistance. *Infection and Drug Resistance*, 13, 1111-1121. [doi: 10.2147/IDR.S247271](https://doi.org/10.2147/IDR.S247271).
- [17] Gupta, V.V., & Sharma, A.K. (2021). *Rhizosphere biology: Interactions between microbes and plants*. Singapore: Springer. [doi: 10.1007/978-981-15-6125-2](https://doi.org/10.1007/978-981-15-6125-2).

[18] Hakim, S., Naqqash, T., Nawaz, M.S., Laraib, I., Siddique, M.J., Zia, R., Mirza, M.S., & Imran, A. (2021). Rhizosphere engineering with plant growth-promoting microorganisms for agriculture and ecological sustainability. *Frontiers in Sustainable Food Systems*, 5, article number 617157. [doi: 10.3389/fsufs.2021.617157](https://doi.org/10.3389/fsufs.2021.617157).

[19] Hamid, B., Zaman, M., Farooq, S., Fatima, S., Sayyed, R.Z., Baba, Z.A., Sheikh, T.A., Reddy, M.S., Enshasy, H.E., Gafur, A., & Suriani, N.L. (2021). Bacterial plant biostimulants: A sustainable way towards improving growth, productivity, and health of crops. *Sustainability*, 13(5), article number 2856. [doi: 10.3390/su13052856](https://doi.org/10.3390/su13052856).

[20] Havryliuk, L.V., Kichigina, O.O., & Turovnik, Yu.A. (2022). Biopreparations as an agroecological factor for increasing biosafety in agroecosystems. *Balanced Nature Management*, 4, 105-111. [doi: 10.33730/2310-4678.4.2022.275037](https://doi.org/10.33730/2310-4678.4.2022.275037).

[21] Horizon Europe. (n.d.). *Horizon Europe in Ukraine*. Retrieved from <https://horizon-europe.org.ua/en/heo-in-ua/>.

[22] Ismanzhanov, A.I., & Tashiev, N.M. (2016). Development and research of the technology for powdering agricultural products using solar energy. *Applied Solar Energy (English translation of Geliotekhnika)*, 52(4), 256-258. [doi: 10.3103/S0003701X16040101](https://doi.org/10.3103/S0003701X16040101).

[23] Kapoore, R.V., Padmaperuma, G., Maneein, S., & Vaidyanathan, S. (2021). Co-culturing microbial consortia: Approaches for applications in biomanufacturing and bioprocessing. *Critical Reviews in Biotechnology*, 42(1), 46-72. [doi: 10.1080/07388551.2021.1921691](https://doi.org/10.1080/07388551.2021.1921691).

[24] Karlash, Yu.V., & Krasinko, V.O. (2022). *Fundamentals of biotechnological production design*. Kyiv: National University of Food Technologies.

[25] Kong, Z., & Liu, H. (2022). Modification of rhizosphere microbial communities: A possible mechanism of plant growth promoting rhizobacteria enhancing plant growth and fitness. *Frontiers in Plant Science*, 13, article number 910813. [doi: 10.3389/fpls.2022.920813](https://doi.org/10.3389/fpls.2022.920813).

[26] Kosovska, N., Makarenko, N., Bondar, V., Matviikiv, A., & Symochko, L. (2022). Soil microbiome under the influence of nano and biopreparations. *International Journal of Ecosystems and Ecology Science*, 12(3), 1-8. [doi: 10.31407/ijees12.301](https://doi.org/10.31407/ijees12.301).

[27] Kovalenko, O.A. (2021). *Agroecological justification and development of elements of biologized technologies for growing agricultural crops in the South of Ukraine*. Kherson: Kherson State Agrarian and Economic University.

[28] Kundius, V. (2021). Justification of the concept of development of modern organic agriculture on the basis of biointensive technologies. *SHS Web of Conferences*, 101, article number 02031. [doi: 10.1051/shsconf/202110102031](https://doi.org/10.1051/shsconf/202110102031).

[29] Kuts, O., Onyschenko, O., Chayuk, O., Konovalenko, K., & Ilinova, Ye. (2023). Use of microbial preparations and humics fertilizer for eggplant growing in film greenhouses. *Scientific Reports of the National University of Life and Environmental Sciences of Ukraine*, 19(6). [doi: 10.31548/dopovid6\(106\).2023.008](https://doi.org/10.31548/dopovid6(106).2023.008).

[30] Liu, Q., Cheng, L., Nian, H., Jin, J., & Lian, T. (2022). Linking plant functional genes to rhizosphere microbes: A review. *Plant Biotechnology Journal*, 21(5), 902-917. [doi: 10.1111/pbi.13950](https://doi.org/10.1111/pbi.13950).

[31] Malik, A.A., Martiny, J.B.H., Brodie, E.L., Martiny, A.C., Treseder, K.K., & Allison, S.D. (2019). Defining trait-based microbial strategies with consequences for soil carbon cycling under climate change. *ISME Journal*, 14(1), 1-9. [doi: 10.1038/s41396-019-0510-0](https://doi.org/10.1038/s41396-019-0510-0).

[32] McCarty, N.S., & Ledesma-Amaro, R. (2018). Synthetic biology tools to engineer microbial communities for biotechnology. *Trends in Biotechnology*, 37(2), 181-197. [doi: 10.1016/j.tibtech.2018.11.002](https://doi.org/10.1016/j.tibtech.2018.11.002).

[33] Mustafayeva, K., Mahiou-Leddet, V., Suleymanov, T., Kerimov, Y., Ollivier, E., & Elias, R. (2011). Chemical constituents from the roots of Cephalaria kotschy. *Chemistry of Natural Compounds*, 47(5), 839-842. [doi: 10.1007/s10600-011-0079-y](https://doi.org/10.1007/s10600-011-0079-y).

[34] Nikonchuk, N., & Samoilenko, M. (2024). The influence of biopreparations on the growth and development of tomatoes under biological cultivation. *Ecological Engineering & Environmental Technology*, 25(8), 37-46. <https://doi.org/10.12912/27197050/189236>

[35] Organisation for Economic Co-operation and Development. (2019). *Impacts of agricultural policies on productivity and sustainability performance in agriculture: A literature review*. Retrieved from https://www.oecd.org/en/publications/impacts-of-agricultural-policies-on-productivity-and-sustainability-performance-in-agriculture-a-literature-review_6bc916e7-en.html.

[36] Panfilova, A., Korkhova, M., Domaratskiy, Y., & Kozlova, O. (2025). Development of winter wheat productivity under the influence of biopreparations and different moisture conditions in the steppe zone. *Ecological Engineering & Environmental Technology*, 26(3), 255-264. [doi: 10.12912/27197050/200245](https://doi.org/10.12912/27197050/200245).

[37] Peterson, S.B., Bertolli, S.K., & Mougous, J.D. (2020). The central role of interbacterial antagonism in bacterial life. *Current Biology*, 30(19), R1203-R1214. [doi: 10.1016/j.cub.2020.06.103](https://doi.org/10.1016/j.cub.2020.06.103).

[38] Philippot, L., Chenu, C., Kappler, A., Rillig, M.C., & Fierer, N. (2023). The interplay between microbial communities and soil properties. *Nature Reviews Microbiology*, 22(4), 226-239. [doi: 10.1038/s41579-023-00980-5](https://doi.org/10.1038/s41579-023-00980-5).

[39] Rai, S., & Prasad, R. (2023). *New and future developments in microbial biotechnology and bioengineering: Trichoderma for biotechnological applications: Current insight and future prospects*. Amsterdam: Elsevier. [doi: 10.1016/C2021-0-00448-X](https://doi.org/10.1016/C2021-0-00448-X).

- [40] Saberi-Riseh, R., Moradi-Pour, M., Mohammadinejad, R., & Thakur, V.K. (2021). Biopolymers for biological control of plant pathogens: Advances in microencapsulation of beneficial microorganisms. *Polymers*, 13(12), article number 1938. [doi: 10.3390/polym13121938](https://doi.org/10.3390/polym13121938).
- [41] Saeed, Q., et al. (2021). Rhizosphere bacteria in plant growth promotion, biocontrol, and bioremediation of contaminated sites: A comprehensive review of effects and mechanisms. *International Journal of Molecular Sciences*, 22(19), article number 10529. [doi: 10.3390/ijms221910529](https://doi.org/10.3390/ijms221910529).
- [42] Sharma, V., Salwan, R., & Al-Ani, L.K.T. (2020). *Molecular aspects of plant beneficial microbes in agriculture*. Amsterdam: Academic Press. [doi: 10.1016/C2018-0-03869-4](https://doi.org/10.1016/C2018-0-03869-4).
- [43] Sorbara, M.T., & Pamer, E.G. (2022). Microbiome-based therapeutics. *Nature Reviews Microbiology*, 20(6), 365-380. [doi: 10.1038/s41579-021-00667-9](https://doi.org/10.1038/s41579-021-00667-9).
- [44] Tiwari, A.K. (2022). Assessing the real productivity of organic farming systems in contemporary. *Plant Science Archives*, 7(4). [doi: 10.51470/psa.2022.7.4.01](https://doi.org/10.51470/psa.2022.7.4.01).
- [45] Tkachuk, O.P., Shkatula, Yu.M., & Titarenko, O.M. (2020). *Agricultural ecology*. Vinnytsia: Vinnytsia National Agrarian University.
- [46] Tsvihun, A., Karatieveva, O., Ponko, L., Yakovchuk, V., Yulevich, O., & Ponichtera, P. (2025). Effect of probiotic supplement on growing lambs of. *Online Journal of Animal and Feed Research*, 15(3), 150-158. [doi: 10.51227/ojafr.2025.18](https://doi.org/10.51227/ojafr.2025.18).
- [47] Vidhyasekaran, P. (2024). *Bacterial disease resistance in plants: Molecular biology and biotechnological applications*. Boca Raton: CRC Press. [doi: 10.1201/9781003578550](https://doi.org/10.1201/9781003578550).
- [48] Wei, X., Xie, B., Wan, C., Song, R., Zhong, W., Xin, S., & Song, K. (2024). Enhancing soil health and plant growth through microbial fertilizers: Mechanisms, benefits, and sustainable agricultural practices. *Agronomy*, 14(3), article number 609. [doi: 10.3390/agronomy14030609](https://doi.org/10.3390/agronomy14030609).
- [49] Zakharchuk, O.V., Lupenko, Y.O., Hutorov, A.O., & Dorokhov, O.V. (2019). Economics of development of the seed-growing in Ukraine. *Bulletin of the Transilvania University of Brasov Series II Forestry Wood Industry Agricultural Food Engineering*, 12(2), 127-136. [doi: 10.31926/but.fwiafe.2019.12.61.2.11](https://doi.org/10.31926/but.fwiafe.2019.12.61.2.11).
- [50] Zulfiqar, F., Navarro, M., Ashraf, M., Akram, N.A., & Munné-Bosch, S. (2019). Nanoertilizer use for sustainable agriculture: Advantages and limitations. *Plant Science*, 289, article number 110270. [doi: 10.1016/j.plantsci.2019.110270](https://doi.org/10.1016/j.plantsci.2019.110270).

Молекулярно-генетичні та біотехнологічні основи створення комплексних мікробних препаратів для збалансованого живлення рослин

Олена Каратеєва

Кандидат сільськогосподарських наук, доцент
Миколаївський національний аграрний університет
54008, вул. Георгія Гонгадзе, 9, м. Миколаїв, Україна
<https://orcid.org/0000-0002-0652-1240>

Євген Баркарь

Кандидат сільськогосподарських наук, доцент
Миколаївський національний аграрний університет
54008, вул. Георгія Гонгадзе, 9, м. Миколаїв, Україна
<https://orcid.org/0000-0002-0692-5392>

Олена Юлевич

Кандидат технічних наук, доцент
Миколаївський національний аграрний університет
54008, вул. Георгія Гонгадзе, 9, м. Миколаїв, Україна
<https://orcid.org/0000-0003-1594-0700>

Ірина Лята

Асистент
Миколаївський національний аграрний Університет
54008, вул. Георгія Гонгадзе, 9, м. Миколаїв, Україна
<https://orcid.org/0000-0002-1672-2337>

Віталій Оверченко

Кандидат сільськогосподарських наук, доцент
Національний університет біоресурсів і природокористування України
03041, вул. Героїв Оборони, 15, м. Київ, Україна
<https://orcid.org/0009-0009-5435-6815>

Анотація. Дослідження спрямоване на визначення молекулярно-генетичних механізмів, що регулюють функціональну активність мікробних препаратів у ризосфері рослин. Методологія включала аналіз експресії ключових генів, порівняння ефективності різних штамів мікроорганізмів у ґрунтовому середовищі, узагальнення факторів, що впливають на стабільність біопрепаратів, та систематизацію даних для встановлення взаємозв'язку між генетичними механізмами та агрономічними показниками. Отримані результати підтвердили, що експресія ключових генів, відповідальних за фіксацію азоту, мобілізацію фосфору та синтез біологічно активних сполук, визначає ефективність біологічних препаратів у ґрунтовому середовищі, що було верифіковано шляхом генетичного аналізу функціональних маркерів, мікробіологічного дослідження життєздатності штамів і аналітичного визначення вмісту доступних елементів живлення та продуктивності рослин. Комбінування азотфіксуючих, фосфатмобілізуючих і біостимулюючих мікроорганізмів забезпечувало формування стабільних мікробних спільнот, здатних підвищувати біодоступність елементів живлення та продуктивність культур. При застосуванні *Azospirillum brasiliense* на небобових культурах концентрація амонійних сполук у ризосфері зростала до 40 %, що супроводжувалося посиленням азотного живлення. Використання фосфатмобілізуючих бактерій дозволяло зменшити норму внесення фосфорних добрив на 25-40 % без втрати врожайності. Загалом, застосування біопрепаратів на зернових, бобових та овочевих культурах забезпечувало скорочення потреби у хімічних добривах на 20-30 % без зниження агрономічної ефективності. Стабілізація мікроорганізмів шляхом мікроінкапсуляції забезпечувала підвищену виживаність у ґрунтовому середовищі та рівномірне поширення в зоні ризосфери, тоді як спрей-сушіння дозволяло отримати порошкові форми біопрепаратів із тривалим строком зберігання. Поєднання мікробних препаратів із наноформами мінеральних добрив сприяло рівномірному вивільненню елементів живлення, що позитивно впливало на їх засвоєння, хоча точні коефіцієнти цього процесу у роботі не наведені. Практичне значення отриманих результатів полягає у можливості цілеспрямованої розробки біологічних препаратів із прогнозованою здатністю підвищувати засвоєння азоту та фосфору рослинами, знижувати потребу в мінеральних добривах та забезпечувати стабільну продуктивність сільськогосподарських культур в умовах інтенсивного землеробства

Ключові слова: продукти біотехнології; консорціум; фіксація азоту; мобілізація фосфору; біологічно активні речовини; мікроінкапсуляція експресія генів