

ECOLOGY, BIOTECHNOLOGY, AGRICULTURE AND FORESTRY

IN THE 21ST CENTURY

PROBLEMS AND SOLUTIONS



EDITED BY
S. STANKEVYCH, O. MANDYCH

**ECOLOGY, BIOTECHNOLOGY, AGRICULTURE
AND FORESTRY IN THE 21ST CENTURY:
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**Tallinn
Teadmus
2024**

UDC 502:504:630:631:338

Ecology, Biotechnology, Agriculture and Forestry in the 21st century: problems and solutions. Monograph. Edited by S. Stankevych, O. Mandych. – Tallinn: Teadmus OÜ, 2024. 370 p.

ISBN 978-9916-9969-9-7

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The monograph is a collection of the results of scientists' achievements obtained directly in real conditions. The authors are recognized specialists in their fields, as well as young scientists and graduate students of Ukraine. The studies are conceptually grouped in sections: biotechnology, ecology, agriculture, forestry, sustainable development of the economy and the principles of effective agribusiness. The monograph will be of interest to specialists in biotechnology, ecology, breeding, plant protection, agrochemistry, soil science, forestry, agribusiness, etc., researchers, teachers, graduate students and students of specialized specialties of higher educational institutions, as well as everyone who is interested in sustainable development in the agricultural sphere and Green Deal Implementation strategies.

Keywords: sustainable development, modern technologies, agricultural production, biotechnology, ecology, plant protection, forestry, agribusiness.

ISBN 978-9916-9969-9-7

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**APPLICATION OF BIOLOGICAL PREPARATIONS ON THE
CROPS OF PERENNIAL LEGUMINOUS GRASSES AS A MEANS
OF PROVIDING SOILS WITH ATMOSPHERIC NITROGEN**

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The results of our research showed that the use of biological preparations significantly increases the green mass of legumes. The use of preparations contributes to the better development of the plants of alfalfa and clover that is evidenced by the obtained results. It is studied the question of application of biological preparations while growing legumes and the role of nitrogen-fixing microorganisms in increasing the plant productivity. The expediency of using such preparations as Gumat Universal, Azohran, Biocomplex BTU and Emochka on the crops of alfalfa and clover is shown. The purpose of the research is to justify the expediency of using biological preparations as a factor in increasing the green mass of leguminous plants and a source of growing rhizobia on the root system of legumes. The obtained results indicate that the use of bacterial preparations contributes to the increase in the yield of green mass of plants. When analyzing the results of the use of microbiological preparations we see that the increase of green mass on the crops of alfalfa is 1.9 t/ha for Azohran and 3.1 t/ha for Emochka compared to the control option. The use of the preparation Azohran on the crops of clover provided the increase of 1.9 t/ha in the yield of green mass on the unfertilized soil. The increase in the yield of green mass was 2.8 t/ha on the unfertilized soil where the preparation Emochka was applied. The alfalfa rhizosphere has a positive effect on increasing the number and weight of tubers when using biological preparations. The obtained data show that the maximum indicators of the rhizobia increase are noted in the flowering phase of the studied crops. It is studied the question of application of biological preparations while growing legumes and the role of nitrogen-fixing microorganisms for soils. The value and importance of legumes as

predecessors in the accumulation of atmospheric nitrogen is shown. The effect of application of microbial preparations such as Azohran and Emochka on the crops of alfalfa is examined. It is shown that the use of fertilizers significantly increases the weight of root and crop residues of alfalfa. The use of fertilizers contributes to the development of the root system of crops that is evidenced by the results. The comparison of fertilization systems of the crop shows that the use of the recommended norms of fertilizers not only increases the yield of alfalfa, but also enhances its value as a predecessor by accumulating the organic matter. The evaluation of fertilizers as an important factor positively impacting on relationship between rhizobia and crop nutrient is done. It ultimately determines the bottom line of their symbiosis. As we see the environmentally appropriate crop production has a fairly large arsenal of microbiological benefits that will help a lot to reduce the amount of used expensive and environmentally unfavorable nitrogen and phosphate fertilizers. Thus, fertilization is one of the important factors that largely changes the relationship between rhizobia and crop nutrient that ultimately determines the bottom line of their symbiosis. The best results in the accumulation of atmospheric nitrogen have been obtained in the option with the use of the preparation Emochka and $N_{30}P_{60}K_{60}$.

Key words: nitrogen, nitrogen fixation, microorganisms, soil, biological preparations, legumes, symbiosis, alfalfa, clover.

Introduction

Soil is a biological environment, the effective use of which can increase the production and improve the quality of grain, fodder, and technical raw materials without unnecessary costs.

In addition to the organic residues of plants and animals, the soil contains many small (micro-), medium (meso-) and larger (macro-) organisms that largely affect vital activity of plants. Microbiota is of the greatest importance for vegetative plants. It prevails on the roots of plants and in the soil adjacent to their rhizosphere, which is located in the soil layer up to 60 cm. This layer is biologically the most active; it is literally the environment of microbiological activity [1-3]. The biological soil activity is supported by deep loosening, plowing organic residues, applying mineral fertilizers, irrigation, keeping the correct crop rotation and their cultivation in mixed, compatible or compacted sowing. The activity of microbiota sharply decreases at the disordered (spontaneous) placement of crops, while the amount of unwanted mesobiota such as nematodes, mite larvae, and

various harmful insects increases. The most active microbiota is near the root system of legumes such as alfalfa, clover, lupine, seradella, hairy vetch, melilot, soybean, etc. [4].

In the rhizosphere of alfalfa, one kilogram of soil contains from 50 to 100 billion bacteria. The live mass of microbiota in the arable layer of the soil reaches more than 10 t/ha. As it deepens, its activity decreases due to deterioration air regime, overmoistening, the content of oxidized forms of iron, aluminum and other elements. As it has already been mentioned, an important agrotechnical measure is a deep (50-70 cm) soil loosening, which significantly increases its biological activity depth. It is of significant importance for increasing the yield of crops at the expense of soil fertility. In addition, mineral and other fertilizers are saved; the ecological condition of the field improves [5].

Decomposing roots, post-harvest stubble residues, manure, siderates, etc., microorganisms increase the content of humus, mobile nitrogen, phosphorus and potassium compounds, as well as the other plant nutrients in the soil. The biological substances such as enzymes, vitamins, free amino acids and auxins, having properties of biocatalysts, are formed in the soil as a result of their activities. These substances activate growth processes in the plant and are an important biological soil factor.

Today, much attention is paid to alternative methods of farming that would provide the maximum productivity and help to obtain ecologically clean plant products [1-3]. An important factor in increasing the productivity of agroecosystems, the potential of which is not used enough now, is an activation of microbial-plant interaction by introducing microbial preparations and plant growth regulators (PGR) of both natural and synthetic origin. They intensify physiological and biochemical processes in plants, increase their resistance to diseases and have a positive effect on soil microorganisms. The production loses at least 10-30% of the harvest without the use of biological preparations for treating the seeds of leguminous crops [1]. When using them the protein content in seeds increases by 2-6%, even when the populations of aboriginal rhizobia are present in the soil [2].

The deficiency of vegetable protein, the orientation of agriculture on ecological clean production, as well as high prices of mineral and organic fertilizers led to the growth of interest in leguminous crops [1, 2]. It is known that legumes, including clover and alfalfa, have great fodder value, because they stand out among other agricultural crops by the protein content in green mass [4]. These plants play an important role in increasing the fertility of

soil thanks to their ability to fix molecular nitrogen in symbiosis with rhizobia. They not only provide themselves with nitrogen, but also contribute to its accumulation in the soil and increasing the productivity of the next crop rotations. Biological nitrogen is the cheapest and most environmentally friendly source of this element for agriculture. The positive effect of rhizobia on the yield of leguminous plants was determined more than 100 years ago, and since then various forms of biological preparations based on active strains of rhizobia have been created [3].

In recent years a number of microbial preparations for many agricultural crops have been developed at the Institute of Agricultural Microbiology of the Ukrainian Academy of Sciences. However, the research on selection of new effective strains is conducted constantly. Their use allows to increase the yield of leguminous plants and the protein content in them. An economic substantiation of the expediency of using biological preparations during the cultivation of agricultural crops, and in particular legumes, is relevant in the modern conditions of increasing in the price of traditional resources [13].

Analysis of recent research and publications

The symbiotic nitrogen fixation is carried out by rhizobia, which are in the close symbiotic relationship with leguminous plants. Symbiosis also helps increasing both the soil fertility and the yield of next crop rotations, since root and stubble residues rich in nitrogen, phosphorus, calcium, potassium and other macro- and microelements remain in the soil [6].

In this regard, the works of M. Mishustin, P. Vavilov, S. Posypanov, O. Berestetskyi, Yu. Vozniakov, L. Dorosynskyi, A. Babich, V. Patyka, I. Tykhonovych, D. Kuk, P. Ambrus and other scientists are of great importance. The Institute of Plant Physiology and Genetics of NAS of Ukraine, the Institute of Microbiology and Virology of NAS of Ukraine, the Institute of Agricultural Microbiology of NAAS of Ukraine and the Institute of Agroecology of NAAS of Ukraine deal with this issue.

Biologically active preparations play a significant role in the process of forming crop yields, when they are used in the modern agricultural technologies.

The ability of alfalfa and clover, as legumes, to absorb nitrogen from atmosphere is one of the main biological features of these crops.

L. Dorosynskyi [5], analyzing the scale of symbiotic nitrogen fixation, cites the data of D. Pryanyshnykov (“when providing them phosphorus and potassium, various types of leguminous plants can fix the following

amounts of nitrogen per hectare per year: clover – 150-160 kg, lupine – 160 kg, alfalfa – 300 kg”). As we can see, the level of nitrogen supply is recognized as high enough. Bielfe (quoted by L. Dorosynskiy, 1965) gives even larger figures: the maximum amount of atmospheric nitrogen assimilated per year per 1 ha in Sweden is 150-200 kg for annual leguminous crops (peas, vetch, etc.), and for perennials (clover and alfalfa) - 300-400 kg. At the same time, the researcher notes that the average indicators of the amount of fixed nitrogen are 50-100 kg, respectively, for annual and perennial plants. A lot of attention was paid to this issue by a large number of Ukrainian scientists, among whom V. Patyka, I. Tykhonovych, V. Volkohon, V. Morhun, S. Kots and other scientists of the Institute of Plant Physiology and Genetics of NAS of Ukraine, the Institute of Microbiology and Virology of NAS of Ukraine, the Institute of Agricultural Microbiology of NAAS of Ukraine and the Institute of Agroecology of NAAS of Ukraine.

Conditions and methods of research

Microbiological preparations play an increasingly important role in the process of forming crop yields, when they are used in the modern agricultural technologies. The bacteria inhabiting roots form a kind of biological “cover” – the rhizosphere and are trophic mediators between the soil and the plant. The microorganisms are responsible for transformation of a number of complex compounds in space, which are available for plant nutrition. Soil microorganisms are irreplaceable and integral component in the system soil–microorganisms–plant. Therefore, the plant surrounded by a complete complex of microorganisms receives the necessary root nutrition and, as a result, implements its own genetic potential for yield [7].

Today, most soils have individual microorganisms that have always been an indicator of fertility, are on the verge of extinction. Their place is taken by bacteria improper for the soil-forming process. At the same time, the young roots are settled by non-specific microorganisms that perform atypical functions, instead of “feeding” plants with nutrients and providing them with biologically active substances compounds, they themselves compete with them for nutrients. The consequences are known: agricultural crops do not provide a full harvest, even with sufficient mineral nutrition. Similar conditions are developed during the introduction of new types of cultivated plants.

For example, alfalfa, when growing in the traditional soil and climatic conditions, creates active nitrogen-fixing symbioses with rhizobia, forming morphologically expressed structures on the roots – bubbles, in which the

binding such a necessary element for plant development as nitrogen is carried out from the atmospheric air.

In such conditions the absence of the necessary nitrogen-fixing bacteria reduces the value of this leguminous crop as a nitrogen accumulator to the level of nitrogen loss.

In this connection, there is a need to apply agricultural techniques aimed at increasing the number of agronomically valuable microorganisms in soils. One of these methods is the use of pre-sowing inoculation of leguminous crops [8].

Currently, an essential increasing in the production of legumes, which are the main source of environmentally friendly protein balanced in terms of amino acid composition and content, remains one of the main tasks of the agrarian sector of the economy of Ukraine in the development of agricultural production [9].

Today a number of microbiological preparations have been created on the basis of nitrogen-fixing bacteria for most types of agricultural crops. The conditions for their effective use have been defined. The drugs have undergone the production inspection and a significant amount of them is recommended for use in the agricultural production [14].

Biological preparations have a complex effect on the growth and development of plants, as well as on the condition of agrocenoses. First of all, this is the enzymatic binding of atmospheric nitrogen. The industrial production of nitrogen fertilizers due to binding of atmospheric nitrogen is extremely energy-intensive, as it requires about 500 degrees and the excess pressure at 300 atm, whereas bacteria are capable of fixing nitrogen under the normal atmospheric conditions. In association or symbiosis with plants, bacteria bind a large amount of nitrogen. The dimensions of nitrogen accumulation may be sufficient to ensure full development of certain leguminous crops and replenish the nitrogen fund of soils [14].

The most important reserve for overcoming nitrogen deficiency in agriculture of Ukraine is expanding the use of bacterial fertilizers, i.e. preparations, the basis of which are nitrogen-fixing bacteria.

The efficiency of using microbiological preparations affects not only on the growth and development of plants, but also contributes to the additional formation of harvest elements by inoculated plants [10-12].

The economic efficiency of bacterization is quite high, considering the indicators of yield increase, as well as the saving of nitrogen fertilizers. The pre-sowing bacterization of seeds does not violate the technologies of growing these crops, but it is only an additional ecologically and

economically justified measure to increase its productivity. Taking into account the optimization of plant nutrition, the need to use microbial preparations in crop production is beyond doubt [9].

Research results

Azohran is a combined bacterial preparation of complex action. It is produced in the liquid or granular form. Azohran provides an increase in the yield of grain, vegetable and industrial crops by 15-40%. The composition of the preparation includes bacteria of the strain *Bacillus subtilis*, which has antagonistic properties to a wide range of causative agents of root rot, as well as fungal and bacterial diseases. It has the ability to mobilize phosphate (ensures the transition of immobile soil phosphorus compounds into forms available to plants), which is equivalent to the application of more than 100 kg of phosphorus fertilizers in physical weight. It also increases the ratio of mineral fertilizers assimilation [11-12].

The *Azotobacter vinelandii* strain has nitrogen-fixing properties (transforms nitrogen from the air into ammonium compounds available to plants). Its action is equivalent to the application of 100 kg of ammonium nitrate and does not lead to soil acidification.

Emochka is a complex of several dozen strains of useful microorganisms such as lactic acid bacteria, yeast, photosynthesizing bacteria, radioactive and fermentative fungi. It is enriched with a wide range of trace elements, necessary for the optimal development of microorganisms. It is effective for the prevention of diseases due to the displacement of pathogens and creating conditions that make their development impossible or at least difficult. In turn, the wealth of trace elements and photosynthetic bacteria is an ideal source of nutrition for plants in the technology of natural growth biostimulation. The wide spectrum of action of the preparation allows to use it in agriculture, as well as to improve the ecological condition.

- It increases the biological activity of the soil and improves its structure.
- It stimulates the growth and development of plants.
- It increases the absorption of nutrients by the roots.
- It has an antiseptic and antioxidant effect.

The research was carried out on the experimental plots of the research farm “Agronomichne” of Vinnytsia National Agrarian University in 2020-2022. The results of our research showed that the use of biological preparations significantly increases the green mass of leguminous plants.

The use of preparations contributes to the better development of alfalfa and meadow clover plants that is evidenced by the obtained results (Tables 1, 2).

Table 1

**Productivity of green mass and yield of dry matter of alfalfa
depending on foliar fertilization for 2020-2022**

Preparations	The main fertilization	Green mass, t/ha	Dry matter, t/ha
Control	without fertilizers	16.3	3.2
	N ₃₀ P ₃₀ K ₃₀	16.8	3.8
	N ₃₀ P ₆₀ K ₆₀	17.0	3.9
Gumat Universal	without fertilizers	17.1	4.1
	N ₃₀ P ₃₀ K ₃₀	17.5	4.4
	N ₃₀ P ₆₀ K ₆₀	17.9	4.6
Azohran	without fertilizers	18.2	4.8
	N ₃₀ P ₃₀ K ₃₀	18.5	5.0
	N ₃₀ P ₆₀ K ₆₀	18.7	5.2
Biocomplex BTU	without fertilizers	17.9	4.6
	N ₃₀ P ₃₀ K ₃₀	18.3	4.9
	N ₃₀ P ₆₀ K ₆₀	18.5	5.0
Emochka	without fertilizers	19.1	5.3
	N ₃₀ P ₃₀ K ₃₀	19.4	5.5
	N ₃₀ P ₆₀ K ₆₀	19.6	5.8

As it can be seen from Table 1, the best increase in green mass was observed when using the preparations Azohran and Emochka. According to the results of the use of the preparation Emochka on sowing alfalfa crops, it can be concluded that its use has a positive effect on the yield indicators of green mass.

The analysis of the results of the use of microbiological preparations (Table 2) shows that the increase in green mass is significantly higher on meadow clover crops compared to the control option. As well as on alfalfa crops, the best results were obtained on the options with preparations Azohran and Emochka.

The use of the preparation Azohran provided the increase of 1.9 t/ha in the yield of green mass on the unfertilized soil. The increase in the yield of green mass was 2.8 t/ha on the unfertilized soil where the preparation Emochka was applied.

When studying the action of microbiological preparations, it is important to examine them not only as a nitrogen fixer, but also to research their impact on the soil rhizosphere (Tables 3, 4).

Table 2

Productivity of green mass and yield of dry matter of meadow clover depending on foliar fertilization for 2020-2022

Preparations	The main fertilization	Green mass, t/ha	Dry matter, t/ha
Control	without fertilizers	16.2	3.1
	N ₃₀ P ₃₀ K ₃₀	16.7	3.7
	N ₃₀ P ₆₀ K ₆₀	16.9	3.8
Gumat Universal	without fertilizers	17.0	4.0
	N ₃₀ P ₃₀ K ₃₀	17.4	4.3
	N ₃₀ P ₆₀ K ₆₀	17.8	4.5
Azohran	without fertilizers	18.1	4.7
	N ₃₀ P ₃₀ K ₃₀	18.4	4.9
	N ₃₀ P ₆₀ K ₆₀	18.6	5.1
Biocomplex BTU	without fertilizers	17.8	4.5
	N ₃₀ P ₃₀ K ₃₀	18.2	4.8
	N ₃₀ P ₆₀ K ₆₀	18.4	4.9
Emochka	without fertilizers	19.0	5.2
	N ₃₀ P ₃₀ K ₃₀	19.3	5.4
	N ₃₀ P ₆₀ K ₆₀	19.5	5.7

Table 3

The effect of biological preparations on increasing rhizobia in the root system of alfalfa (average for 2020-2022)

Research option	Phases of alfalfa development					
	Budding		Flowering		Fruit and seed ripening	
	number of active nodules, pcs/plant	mass of active nodules, g/plant	number of active nodules, pcs/plant	mass of active nodules, g/plant	number of active nodules, pcs/plant	mass of active nodules, g/plant
Control	16.0	0.14	16.3	0.16	16.2	0.15
Gumat Universal	17.0	0.15	18.5	0.17	18.3	0.19
Azohran	16.6	0.17	17.5	0.21	17.3	0.19
Biocomplex x BTU	14.6	0.15	16.7	0.19	15.1	0.17
Emochka	17.6	0.18	19.7	0.21	18.8	0.19

Analyzing the data in Table 3, it can be concluded that the alfalfa rhizosphere has a positive effect on increasing the number and weight of nodules when using microbial preparations. The obtained data indicate that the maximum increasing rates of rhizobia are observed in the flowering phase. When using Gumat Universal and Emochka, the indicators of the number of active nodules are 18.5 and 19.7 pcs/plant, respectively.

Table 4

The effect of biological preparations on increasing rhizobia in the root system of meadow clover (average for 2020-2022)

Research option	Phases of alfalfa development					
	Budding		Flowering		Fruit and seed ripening	
	number of active nodules, pcs/plant	mass of active nodules, g/plant	number of active nodules, pcs/plant	mass of active nodules, g/plant	number of active nodules, pcs/plant	mass of active nodules, g/plant
Control	15.0	0.13	15.7	0.15	15.1	0.14
Gumat Universal	16.0	0.14	17.5	0.19	17.3	0.17
Azohran	16.4	0.15	16.5	0.15	16.3	0.14
Biocomplex BTU	16.1	0.14	16.6	0.15	16.1	0.14
Emochka	15.3	0.14	15.8	0.16	15.7	0.17

Therefore, the results of the experiment show that the use of these preparations has a positive effect on the growth processes of the studied crops. The maximum productivity of the leguminous-rhizobial complex is ensured by the use of Gumat Universal and Biocomplex BTU in the flowering phase of meadow clover. The conducted calculations of the number of nodules proved that there were 17.5 pcs/plant when using Gumat Universal, while 16.6 pcs/plant when using Biocomplex BTU, which was by 0.9-1.8 pcs/plant more compared to the control.

In the conditions of modern intensive agricultural production, which involves a significant removal of nitrogen from the soil by crops, there is an urgent need to improve nitrogen nutrition of plants both by increasing the mobilization of soil resources and by additional sources of this element.

From 1-3 t/ha (tomatoes, potatoes, oats, barley, fodder beets, cabbage) to 7-9 and even 12 t/ha (alfalfa) of plant residues per year enters the soil of

agrocenoses [7, 9]. Most of the plant residues of the soil are transformed by microorganisms and representatives of the soil fauna within 1-2 years.

The final products of transformation are mineral compounds and humus. Thanks to the arrival of plant residues and their transformation into soil humus, organic matter of the soil is continuously regenerated. The organic matter of the soil contains thousands of compounds, the average lifetime of which varies from a day to hundreds and thousands of years. The organic matter of the soil is in the following forms:

- 1) almost not decomposed or slightly decomposed residues;
- 2) organic residues in the stage of deep transformation, which appear in the form of a homogeneous loose black mass of humus to the naked eye of the observer;
- 3) no traces of plant tissue are observed under the microscope, but a specific soil organic formation - humus is visible. These are amorphous, transparent and weakly colored yellow-brown formations; poorly transparent ones have a darker color, they cement and glue mineral particles of the soil [8].

Alfalfa is a reliable means of increasing soil fertility in field crop rotations. When penetrating into the lower layers of the soil with its powerful root system, it takes out phosphorus, potassium and trace elements from there, enriching the arable layer of the soil with these elements. After harvesting alfalfa, a significant amount of root and crop residues, containing up to 200 kg/ha of nitrogen, remains in the field [9]. The more biomass alfalfa produces, the more powerful its development is, the more nitrogen is contained in the biological mass.

It is not by chance that a lot of attention has recently been paid to the new methods of farming, which involve the wide application of biological preparations in the cultivation of crops and the partial rejection of chemicals in agriculture. One of these measures is the use of biological preparations based on associative nitrogen-fixing microorganisms, which, in addition to fixing molecular nitrogen increase the rate of using nutrients from the soil. It can significantly improve nitrogen nutrition of plants, reduce the use of mineral fertilizers, and thus improve the quality of the obtained products and the state of the environment [10].

The results of our research showed that the mass of root and crop residues increases significantly with the use of fertilizers. The use of fertilizers contributes to the better development of the alfalfa root system, as evidenced by the obtained results. The analytical determination of nitrogen in structural parts of the biological crop showed that the amount of

nitrogen in the root and crop residues of the control option of the experiment without fertilizers is 102.7 kg/ha; it is 117.5 kg/ha when applying $N_{30}P_{30}K_{30}$ and 121.7 kg/ha when applying $N_{30}P_{60}K_{60}$. When alfalfa seeds are treated with Azohran without fertilizers, the nitrogen content in root and crop residues is 136.0 kg/ha; it is 145.5 kg/ha when applying $N_{30}P_{30}K_{30}$ and 169.7 kg/ha when applying $N_{30}P_{60}K_{60}$ (Table 5).

The highest nitrogen content in root and crop residues is observed when seeds are treated with the preparation Emochka. Thus, its use without fertilizers contributes to the accumulation of nitrogen in root and crop residues and amounts to 174.3 kg/ha; it is 187.7 kg/ha when applying $N_{30}P_{30}K_{30}$ and 198.9 kg/ha when applying $N_{30}P_{60}K_{60}$.

The same trend was noted in relation to above-ground biomass. The amount of nitrogen in the control option without fertilizers is 2.06%; it increases by 2.08% when $N_{30}P_{30}K_{30}$ is applied and by 2.11% when $N_{30}P_{60}K_{60}$ is applied.

Table 5

The influence of the after-effect of fertilizers on the accumulation of biologically bound nitrogen in alfalfa crops

Research options		Nitrogen content in					
		root and crop residues		above-ground mass		biomass	biologically bound
Factor A	Factor B	%	kg/ha	%	kg/ha	kg/ha	kg/ha
Control	without fertilizers	2.15	102.7	2.06	109.0	210.8	159.7
	$N_{30}P_{30}K_{30}$	2.19	117.5	2.08	123.1	221.4	207.3
	$N_{30}P_{60}K_{60}$	2.21	121.7	2.11	137.9	237.5	212.4
Azohran	without fertilizers	2.29	136.0	2.19	149.3	228.7	227.4
	$N_{30}P_{30}K_{30}$	2.34	145.5	2.22	156.6	245.6	246.5
	$N_{30}P_{60}K_{60}$	2.41	169.7	2.27	163.8	251.3	260.5
Emochka	without fertilizers	2.21	174.3	2.18	175.6	231.7	271.3
	$N_{30}P_{30}K_{30}$	2.36	187.7	2.24	182.8	259.3	283.4
	$N_{30}P_{60}K_{60}$	2.49	198.9	2.26	196.9	270.4	297.4

In the research option with Azohran, an increase in percentage is also observed: it is 2.19% without fertilizers, while it increases by 2.22% with the application of $N_{30}P_{30}K_{30}$ and by 2.27% with the application of $N_{30}P_{60}K_{60}$.

The highest nitrogen content of the above-ground mass in the research option with the preparation Emochka is 2.18% without fertilizers, 2.24% with the application of N₃₀P₃₀K₃₀ and 2.26% with the application of N₃₀P₆₀K₆₀.

The nitrogen content in biomass is also observed to be the highest when using the preparation Emochka and amounts to 270.4 kg/ha, while the content of biologically bound nitrogen is 297.4 kg/ha. A comparison of fertilization systems in the first year of the crop's life shows that the application of recommended rates of fertilizers not only increases the yield of alfalfa, but also enhances its value as a predecessor. Thus, the increase is 13.8 compared to the control when applying N₃₀P₃₀K₃₀, while it is 16.4 when applying N₃₀P₆₀K₆₀ in the control option of the experiment. When alfalfa seeds are treated with Azohran without fertilizers, the increase is 18.6 compared to the control; it is 19.8 when applying N₃₀P₃₀K₃₀ and 20.7 when applying N₃₀P₆₀K₆₀ (Table 6).

Table 6

The influence of the after-effect of fertilizers on the accumulation of root and crop residues of alfalfa in the first year of use

Research options		Root mass, q/ha	Nutrient residues, q/ha	Amount of residues, q/ha	Increase to the control
Control	without fertilizers	37.7	10.2	47.8	-
	N ₃₀ P ₃₀ K ₃₀	39.2	13.8	53.4	13.8
	N ₃₀ P ₆₀ K ₆₀	40.1	14.2	59.1	16.4
Azohran	without fertilizers	41.2	15.8	65.4	18.6
	N ₃₀ P ₃₀ K ₃₀	45.6	16.8	68.7	19.8
	N ₃₀ P ₆₀ K ₆₀	47.3	17.1	69.8	20.7
Emochka	without fertilizers	49.4	17.2	71.2	21.8
	N ₃₀ P ₃₀ K ₃₀	51.6	18.4	73.4	22.7
	N ₃₀ P ₆₀ K ₆₀	54.3	19.7	73.5	24.3

It can be seen from Table 6 that the greatest increase compared to the control is observed in the research option, where the seeds were treated with the preparation Emochka before sowing. So, the increase is 21.8 without fertilizers; it is 22.7 when applying N₃₀P₃₀K₃₀ and 24.3 when applying

N₃₀P₆₀K₆₀. Therefore, the fertilization system in compliance with the recommended norms creates optimal conditions for the activity of natural groups of nitrogen-fixing microorganisms.

Conclusions

Based on the research results, the feasibility of using biological preparations such as Gumat Universal, Azohran, Biocomplex BTU and Emochka on leguminous crops is justified. It has been determined that the increase of green mass in alfalfa crops is significantly higher compared to the control variant, it is 1.9 t/ha when applying Azohran and 3.1 t/ha when applying Emochka.

The use of the preparation Azohran on the crops of clover provided the increase of 1.9 t/ha in the yield of green mass on the unfertilized soil. When using the preparation Emochka the increase in the yield of green mass was 2.8 t/ha on the unfertilized soil. The maximum indicators of the rhizobia increase are noted in the flowering phase of the leguminous crops. When using Humat Universal and Emochka, the indicators of the number of active nodules are 18.5 and 19.7 pcs/plant, respectively. The conducted calculations of the number of nodules proved that there were 17.5 pcs/plant when using Gumat Universal, while 16.6 pcs/plant when using Biocomplex BTU, which was by 0.9-1.8 pcs/plant more compared to the control.

As we can see, the ecologically appropriate crop production has a rather large microbiological arsenal, the use of which will help to greatly reduce the amount of expensive and environmentally unfavorable nitrogen and phosphorus mineral fertilizers. Thus, the application of mineral fertilizers is one of the important factors that greatly changes the relationship between rhizobia and the plant nutrient and ultimately determines the practical result of their symbiosis. The best results on the accumulation of atmospheric nitrogen were obtained in the option when using the preparation Emochka and applying N₃₀P₆₀K₆₀.

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