# ECONOMIC EFFICIENCY OF GROWING ALFALFA FOR SEEDS BY INOCULATION WITH BACTERIAL PREPARATIONS

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#### Abstract

Research on the impact of seed inoculation with various bacterial preparations on seed productivity of alfalfa varieties, symbiotic nitrogen accumulation, and root system strength was conducted in 2018–2020 at the experimental field of the Institute of Irrigated Agriculture of the National Academy of Sciences of Ukraine. The economic efficiency of growing alfalfa for seeds under inoculation with bacterial preparations was also calculated. The highest seed productivity was obtained after inoculation with a cyanobacterial drug: 456.4 kg/ha in the Unitro variety and 361.1 kg/ha in the Zoryana variety, at a cost of 0.56 c/kg and 0.70 c/kg and conditional net profit of 1571.93 c/ha and 1190.73 c/ha, respectively. The highest root mass (5.76 and 5.80 t/ha) and nitrogen fixation (222 and 228 kg/ha) were noted in the Unitro and Zoryana varieties, respectively, when the cyanobacterial preparation with a cyanobacterial preparation in the Unitro variety 1,894.40 c/ha and in the Zoryana variety 1,516.43 c/ha.

*Key words:* alfalfa, inoculation, nitrogen fixation, root mass, economic efficiency

# **INTRODUCTION**

Alfalfa is a perennial fodder crop that is grown all over the world and is characterized by high biomass productivity, nutritional value and high protein content among fodder legumes. It helps to increase soil fertility [13, 30], protects soils from wind and water erosion [29, 33]. In addition, atmospheric nitrogen fixation makes it an indispensable precursor for other crops. However, the lack of a sufficient amount of seed material, due to the low yield of seeds, does not allow expanding the sown areas of this valuable fodder crop [34].

Increasing the yield of alfalfa seeds is a complex and extremely important problem that can be solved by creating high-yielding varieties and developing improved agricultural technology. Its essence consists in the addition or strengthening of conventional agrotechnical techniques that positively affect the processes of growth and development, the formation of reproductive organs and increase the yield of seeds [11, 32]. Only by creating optimal conditions for the development and growth of plants is it possible to obtain high and stable seed yields.

The root system of crops solves an important task for plants. The powerful architecture of the root system of crops is a guarantee of plant resistance to adverse stress factors, improvement of nutrition, moisture supply and their high yield [9, 26]. This especially applies to alfalfa, which accumulates a large amount of root residues in the soil, improving its meliorational state, humus-forming processes and is a good precursor for the following crops.

The development of the root system depends on many factors, but the main one is the degree of provision of the soil with nutrients. Important factors affecting the development of the root system are moisture availability and nutrient composition of the soil. Low availability of nutrients or moisture reduces or stops cell division and elongation, which leads to the arrest of primary root growth [16, 25].

Plant roots live in close contact with a large number of species of bacteria and fungi in the rhizosphere [2]. Some species, producing substances (ethylene, auxins and cytokinins) can influence the growth and development of roots [3, 23]. Auxin and ethylene regulate the processes that change the architecture of the root system - the elongation of the primary root [1, 28] and the formation of additional root hairs [22].

Bacterial preparations affect the formation of the root structure, stimulating the formation of root hairs and strengthening the branching of lateral roots [21], which increases the surface area [6] and is usually associated with the increased ability of plants to respond to complex environmental conditions [17, 31]. In addition, root structures such as cluster roots [12], nitrogen-fixing nodules [5, 19] and mycorrhizae [2, 27] contribute to the improvement of moisture supply, the transfer of unavailable forms of nutrients into available ones and their absorption by the plant root system. Stimulation of root growth is one of the main markers used to measure the positive effect of bacteria that promote plant growth [11].

The content of organic substances, macro- and microelements, water, oxygen, as well as pH, temperature, and the presence of pathogenic microorganisms in the soil [4, 18] significantly affect the number, physiological and metabolic state of microorganisms. Mineral fertilizers are one of the factors inhibiting the effectiveness of soil microorganisms [8, 24, 35].

Therefore, studying the effectiveness of the inoculation of seeds with bacterial fertilizers to increase the volume of the root mass, activate symbiotic processes, increase the resistance of plants to environmental stress factors is a very important condition for increasing the seed productivity of alfalfa. Bacterial preparations that improve the growth and development of plants, stimulate an increase in yield, accumulation of organic matter (in the form of root residues) and symbiotic nitrogen, while reducing the application of mineral fertilizers for subsequent crops without polluting the environment is very important.

Study of the effect of bacterial preparations on seed productivity, nitrogen-fixing capacity, accumulation of root mass in the second year of alfalfa life and give an economic assessment of the effectiveness of their use.

# MATERIALS AND METHODS

Research was conducted at the experimental field of the Institute of Irrigated Agriculture of the NAAS, located in the steppe zone of the Ingulets irrigated massif, during 2018–2020. The method of establishing a field experiment is split plots. The main areas (factor A) are alfalfa varieties (Unitro and Zoryana); subplots (factor B) - treatment of seeds with bacterial preparations at the rate of 1% of the weight of seeds without dilution with water on the day of sowing: 1 - control (no treatment); 2 – Rhizobophyte (on a specific highly effective strain of nodule bacteria 404b); 3 -Complex of biological preparations (CBP) includes symbiotic nitrogen-fixing, phosphate-mobilizing and bioprotective microorganisms with the functional properties of biological preparations: Rhizobophyte, Phosphoenterin and Biopolicid (1:1:1), which are characterized by a complex effect on crops; Cyanobacterial leguminous 4 \_ consortium (CBC) homogenate based on growth-stimulating cyanobacteria strain 144 and nodule bacteria strain 404b with primary secondary metabolites; and 5 Cyanobacterial drug (CBP) drug based on growth-stimulating cyanobacteria strain 144 and nodule bacteria strain 404<sup>b</sup> with primary metabolites. Sowing period is early spring. Wide-row sowing with 70 cm row spacing. The area of the sowing area is  $60 \text{ m}^2$ , the area of the accounting area is  $40 \text{ m}^2$ , repetition 3 times.

Correlation analysis was performed using Microsoft ® Excel 2013/XLSTAT © -Pro (v. 2015.6.01.23953, 2015, Addinsoft, Inc., Brooklyn, New York, USA). AgroSTAT, Statistica (v. 13), carried out statistical processing of experimental data.

# **RESULTS AND DISCUSSIONS**

The obtained experimental data indicate a different reaction of alfalfa varieties to biological preparations. Seed yield when monoinoculated with nodule bacteria was 16.0-20.0% (Rhizobophyte) higher compared to the control and amounted to 271.8 kg/ha in the Zoryana variety and 361.9 kg/ha in the Unitro variety. However, the effect of the monoculture (Rhizobophyte) on seed productivity differed significantly and was lower than the three-component associations (CBP) based on Rhizobophyte, Phosphoenterin and Biopolycide with a seed yield of 303.6 kg/ha (Zoryana variety) and 398.8 kg/ha (Unitro variety) (Table 1).

The data of some authors indicate the stimulation of plant growth and development under the influence of artificial consortia of various cyanobacteria and species of Rhizobium, which helps to increase their yield [10, 11, 15, 20].

The analysis of the obtained results shows that cvanorhizobial the use of consortia contributes to a sharp increase in seed productivity compared to both control and monoinoculation.

Table 1. Economic assessment of growing alfalfa seeds in the second year of life depending on the variety and the use of bacterial preparations (2018–2020)

Variety (factor A)	Application of bacterial preparations (factor B)	Seed yield, kg/ha	The cost of the obtained products, $\epsilon$ /ha	Costs per 1 ha, $\epsilon$	Cost of 1 kg, $\epsilon$	Conditionally net profit, €/ha	Profitability level, %	
	Control 1 (no inoculation)	307.5	1,230.00	243.10	0.79	986.90	406	
	Rhizobophyte	369.1	1,476.40	247.47	0.67	1,228.93	497	
Unitro	CBP	398.8	1,595.20	249.60	0.63	1,345.60	539	
Uni	CBC	424.6	1,698.40	251.47	0.59	1,446.93	575	
	CBP	456.4	1,825.60	253.67	0.56	1,571.93	620	
	average	391.3	1,565.12	249.06	0.65	1,316.06	527	
	Control 1 (no inoculation)	234.1	936.40	243.10	1.04	693.30	285	
Zoryana	Rhizobophyte	271.8	1,087.20	247.47	0.91	839.73	339	
	СВР	303.6	1,214.40	249.60	0.82	964.80	387	
	CBC	337.3	1,349.20	251.47	0.75	1,097.73	437	
	СВР	361.1	1,444.40	253.67	0.70	1,190.73	469	
	average	301.6	1,206.32	249.06	0.84	957.26	383	
	Assessment of the significance of partial differences							
$LSD_{05}$	А	23.76						

В LSD<sub>05</sub> 18.61

Evaluation of the significance of the main effects LSD<sub>05</sub> A 10.63 LSD<sub>05</sub> B 13.16

Note: The cost of 1 kg of seeds is €4.00/kg Source: Own results.

It should be noted that a strongly pronounced stimulatory effect was noted in associations with cyanobacteria CBC and CBP. A high effect was shown by the cyanorhizobial consortium (CBC) (337.3; 424.6 kg/ha), but the maximum result was obtained in the variant with the use of CBP 361.1 and 456.4

kg/ha in Zoryana and Unitro varieties, respectively.

During the years of research, the cost price of 1 kg of seeds in the second year of life of Unitro and Zoryana alfalfa plants when grown in conditions of natural moisture significantly depended on the weather conditions of the year, and above all, the amount of atmospheric precipitation that fell during the growing season, and amounted to 0.56–0.79 €/kg and 0.70–1.04 €/kg, respectively.

The use of bacterial preparations helped to increase the productivity of the culture and reduce its cost. Thus, when treating seeds with Rhizobophyte, CBP and CBC, the cost price in the Unitro variety was 0.67, 0.63 and 0.59  $\epsilon$ /kg and 0.91, 0.82 and 0.75  $\epsilon$ /kg in the Zoryana variety, while in the control version it was 0.79 and 1.04  $\epsilon$ /kg, respectively. The maximum yield and the lowest cost were obtained for the inoculation of CBP seeds -0.56  $\epsilon$ /kg in the Unitro variety and 0.70  $\epsilon$ /kg in the Zoryana variety.

Conditionally net profit according to the field experiment options depended on the amount of production costs for growing the seeds of the crop and the amount of the obtained harvest. In the control version, the conditional net profit of the Unitro variety was €986.90 and the Zoryana variety was €693.30/ha. Inoculation of seeds with bacterial preparations contributed to obtaining a higher conditional net profit - 1,228.93-1,571.93 €/ha in the Unitro variety and 839.73-1,190.73 €/ha in the Zoryana variety.

Our studies have shown that together with the increase in seed yield, there are also changes in the parameters of accumulation of air-dry root mass and nitrogen fixation. The accumulation of dry mass of the roots according to the experimental variants also has significant fluctuations depending on the use of bacterial preparations. The highest mass was observed in Zoryana and Unitro varieties when using the cyanobacterial preparation (CBP) 5.76 and 5.80 t/ha, respectively, while it was 4.52 and 4.50 t/ha in the control variants (Table 2).

An increase in the activity of nitrogen fixation processes was noted when treated with the same preparations of CBC and CBP, but the highest nitrogen fixation was noted when using a cyanobacterial preparation (CBP), which amounted to 222 and 228 kg/ha in the varieties Unitro and Zoryana, respectively, with low indicators in the control variant of 162 kg/ha (Unitro) and 168 kg/ha (Zoryana). But we not only determined the mass of the system and the accumulation of root symbiotic nitrogen in the soil by alfalfa plants after two years of use, but also tried to translate this nitrogen into monetary units, that is, what can be saved in the form of organic and mineral fertilizers for the next crop. The cost of symbiotic nitrogen was calculated based on the weighted average market price of ammonium nitrate. Using the humification coefficient for the root mass of alfalfa (18%), the amount of humus input after two years of alfalfa use was calculated [7]. The calculations did not take into account root secretions, which make up to 10% of the root mass, as well as annually dead root hairs up to 20% of the root mass [14]. Losses of nitrogen from fertilizers that were not absorbed by the plants of the next crop were also not taken into account, because symbiotic nitrogen is absorbed almost completely. The cost of humus was calculated based on the weighted average market price of manure (50% of dry matter) in terms of humus with a humification factor of 25%.

The amount of future savings depended on the amount of accumulated root mass and symbiotic nitrogen by alfalfa plants. In the variants of the experiment, the amount was the greater, the more the alfalfa plants accumulated root mass and symbiotic nitrogen in the soil. The minimum value of symbiotic nitrogen (133.43  $\in$ /ha in the Unitro variety and 138.67  $\in$ /ha in the Zoryana variety) and humified root mass of 108.00 and 108.47  $\in$ /ha, respectively, was obtained on the control variant without inoculation. In total, it was also the smallest in this variant and amounted to 241.43  $\in$ /ha for the Unitro variety and 247.13  $\in$ /ha for Zoryana.

The use of bacterial preparations increased the accumulation of root mass and symbiotic nitrogen, and accordingly, the amount of money saved in the future was greater. Thus, the increase in the value of symbiotic nitrogen when using bacterial preparations compared to the control was  $25.67-49.84 \in$ /ha in the Unitro variety and  $21.03-48.80 \in$ /ha in Zoryana, while the value of the humified root

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mass was 2.40–31.20  $\notin$ /ha and 4.80–29.76  $\notin$ /ha, respectively.

Table 2. The cost of symbiotic nitr	gen and humified	l root mass depending	on the variety	and application of
bacterial preparations (average for 20)	3-2020)			

Variety (factor A)	Application of bacterial preparations (factor B)	Accumulation of air-dry root mass, t/ha	Atmospheric nitrogen fixation, kg/ha	Root mass humification, t/ha	Cost of symbiotic nitrogen, €/ha	Cost of humified root mass, €/ha	Σ, €/ha
	Control 1 (no inoculation)	4.50	162	0.81	133.43	108.00	241.43
	Rhizobophyte	4.60	193	0.83	159.10	110.40	269.50
itro	CBP	5.00	205	0.90	168.87	120.00	288.87
Unitro	CBC	5.62	215	1.01	177.30	134.87	312.17
<i></i>	CBP	5.80	222	1.04	183.27	139.20	322.47
	average	5.10	200	0.92	164.39	122.49	286.89
	Control 1 (no inoculation)	4.52	168	0.81	138.67	108.47	247.13
Zoryana	Rhizobophyte	4.72	194	0.85	159.70	113.27	272.97
	CBP	5.08	213	0.91	175.53	121.93	297.47
	CBC	5.62	220	1.01	181.43	134.87	316.30
	CBP	5.76	228	1.04	187.47	138.23	325.70
	average	5.14	205	0.93	168.56	123.35	291.91
LSD <sub>05</sub>	А	Assessment of t	the significan 4.40	ce of partial diff	ferences		

LSD <sub>05</sub>	А	0.228	4.40	
LSD <sub>05</sub>	В	0.209	10.35	
		Evaluation of	of the significanc	e of the main effects
LSD <sub>05</sub>	А	0.114	1.97	
LSD <sub>05</sub>	В	0.148	7.32	

Note: The cost of 1 ton of ammonium nitrate (34.4% nitrogen) is  $\notin$  283.33/t and the cost of 1 ton of cattle manure (50% dry matter) is  $\notin$  33.33/t

Source: Own results.

The maximum fixation of symbiotic nitrogen and the accumulation of root mass was obtained in the variant with the use of a cyanobacterial preparation. The price of symbiotic nitrogen was 183.27  $\epsilon$ /ha in the Unitro variety and 187.47  $\epsilon$ /ha in the Zoryana variety, and the humified root mass was 139.20 and 138.23  $\epsilon$ /ha, respectively

When growing the Unitro variety, the amount of money saved for the following crops in the crop rotation (humicized root mass and symbiotic nitrogen) amounted to 286.89 €/ha, with the costs of cultivation - 249.06 €/ha, and with the addition of the cost of the obtained seeds, the total benefit from the cultivation of alfalfa was 1565.12 €/ha for seeds in crop rotation was 1602.95 €/ha with a profitability of 642%. In the Zoryana variety, the amount of funds saved for the following crops in the crop rotation was greater by  $5.02 \notin$ /ha, with the same costs, but the cost of the obtained seeds was lower by  $358.80 \notin$ /ha, and therefore the profit from growing alfalfa for seeds in the crop rotation was less by  $353.78 \notin$ /ha with a profitability of 500% (Table 3).

The lowest profit from growing alfalfa for seeds in crop rotation was obtained in the control variant in the Unitro variety 1,228.33  $\epsilon$ /ha with a profitability of 505%, while in the Zoryana variety 940.43  $\epsilon$ /ha and 387%.

Seed inoculation with Rhizobophyte, a complex of biological preparations (KBP) and a cyanobacterial consortium (CBC) increased the profit from growing alfalfa for seeds in crop rotation in the Unitro variety by 270.10  $\notin$ /ha, 406.14 and 530.77  $\notin$ /ha, respectively, and in the Zoryana variety by 172.27  $\notin$ /ha ha, 321.84 and 473.60  $\notin$ /ha, respectively.

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The maximum benefit of 1,894.40 €/ha from

growing alfalfa for seeds in crop rotation was

obtained in the Unitro variety with CBP inoculations with a profitability of 747%.

Variety (factor A)	Application of bacterial preparations (factor B)	Cost of seeds, €/ha	Amount of aved funds s for the following crops, $\varepsilon/ha$	Costs per 1 ha, $\epsilon$	Benefit received from growing alfalfa for seeds in crop rotation, €/ha	Profitability level of growing alfalfa for seeds in crop rotation, %
	Control 1 (no inoculation)	1,230.00	241.43	243.10	1,228.33	505
	Rhizobophyte	1,476.40	269.50	247.47	1,498.43	606
itro	CBP	1,595.20	288.87	249.60	1,634.47	655
Unitro	CBC	1,698.40	312.17	251.47	1,759.10	700
_	CBP	1,825.60	322.47	253.67	1,894.40	747
	average	1,565.12	286.89	249.06	1,602.95	642
	Control 1 (no inoculation)	936.40	247.13	243.10	940.43	387
a	Rhizobophyte	1,087.20	272.97	247.47	1,112.70	450
/an	CBP	1,214.40	297.47	249.60	1,262.27	506
Zoryana	CBC	1,349.20	316.30	251.47	1,414.03	562
Z	CBP	1,444.40	325.70	253.67	1,516.43	598
	average	1,206.32	291.91	249.06	1,249.17	500

Table 3. Economic efficiency of growing alfalfa for seeds of the second year of life in crop rotation

Source: Own results.

#### CONCLUSIONS

The use of a cyanobacterial preparation made it possible to obtain the highest seed yield of 456.4 kg/ha in the Unitro variety at a cost of 0.56 €/kg and conditional net profit of 1571.93 €/ha and 1190.73 €/ha. The largest root weight of 5.80 t/ha was accumulated by the Unitro variety when using the cyanobacterial preparation, and the highest nitrogen fixation of 228 kg/ha was noted in the Zoryana variety. In conversion, the value of symbiotic nitrogen was 187.47 €/ha in the Zoryana variety and the value of the humified root mass was 139.20 €/ha in the Unitro variety. The greatest profit from growing alfalfa for seeds in crop rotation was obtained inoculation by with a cvanobacterial preparation in the Unitro variety 1,894.40 €/ha and in the Zoryana variety 1,516.43 €/ha.

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