

DOI: <https://doi.org/10.31073/mivg202401-383>

Available at (PDF): <https://mivg.iwpim.com.ua/index.php/mivg/article/view/383>

UDC 633.31/37:631.847.211(477.7)

## ACCUMULATION OF SYMBIOTIC NITROGEN BY PERENNIAL LEGUMINOUS CROPS IN THE SOUTHERN STEPPE OF UKRAINE

G.O. Iutynska<sup>1</sup>, Dr. of Biological Sciences, S.P. Holoborodko<sup>2</sup>, Dr. of Agricultural Sciences, L.V. Tytova<sup>3</sup>, Ph.D. in Biological Sciences, O.D. Dubynska<sup>4</sup>, Ph.D., N.V. Shevchuk<sup>5</sup>, Ph.D. student

<sup>1</sup>D.K. Zabolotny Institute of Microbiology and Virology of the NAS of Ukraine, 154, Akademika Zabolotnoho str., 03143, Kyiv, Ukraine; <https://orcid.org/0000-0001-6692-2946>, e-mail: [galyna.iutynska@gmail.com](mailto:galyna.iutynska@gmail.com);

<sup>2</sup>Institute of Climate-Oriented Agriculture of NAAS of Ukraine, 9, Omelyanovycha-Pavlenka Mykhaila str., 01010, Kyiv, Ukraine; <https://orcid.org/0000-0002-6968-985X>, e-mail: [goloborodko1939@gmail.com](mailto:goloborodko1939@gmail.com);

<sup>3</sup>D.K. Zabolotny Institute of Microbiology and Virology of the NAS of Ukraine, 154, Akademika Zabolotnoho str., 03143, Kyiv, Ukraine; <https://orcid.org/0000-0003-3131-4355>, e-mail: [ltytova.07@gmail.com](mailto:ltytova.07@gmail.com);

<sup>4</sup>Institute of Climate-Oriented Agriculture of NAAS of Ukraine, 9, Omelyanovycha-Pavlenka Mykhaila str., 01010, Kyiv, Ukraine; <https://orcid.org/0000-0002-5572-0094>, e-mail: [klenova-dubinskaelena76@ukr.net](mailto:klenova-dubinskaelena76@ukr.net);

<sup>5</sup>D.K. Zabolotny Institute of Microbiology and Virology of the NAS of Ukraine, 154, Akademika Zabolotnoho str., 03143, Kyiv, Ukraine; <https://orcid.org/0000-0001-5574-4961>, e-mail: [nadia.shevchuk.48@ukr.net](mailto:nadia.shevchuk.48@ukr.net)

**Abstract.** *The use of perennial leguminous crops is an energetically and economically beneficial and ecologically friendly measure for improving the nitrogen balance of soils, which implementation requires specification and additional research of certain soil and climatic conditions and farming systems. The work aimed at studying the accumulation of biologically fixed nitrogen by alfalfa and Hungarian sainfoin in single-species crops, as well as in wheatgrass-alfalfa and wheatgrass-sainfoin grass mixtures on the dark-chestnut soil of southern Ukraine. Laboratory, field, and statistical research methods were applied. The obtained results showed that the accumulation of nitrogen in the biomass of leguminous perennial grasses in single-species crops of blue hybrid alfalfa, Hungarian sainfoin, and intermediate wheatgrass and their grass mixtures depended on the hydrothermal conditions of the growing season, changes in species botanical composition by the years of grass stand used. During the first and second years of use, the nitrogen content in the biomass of alfalfa was 3,54–3,75 %, sainfoin – 3,49–3,65 %, and was significantly higher than in single-species crops of intermediate wheatgrass – 2,62–2,77 % to dry matter. The removal of total nitrogen by intermediate wheatgrass during this period did not exceed 90 kg/ha; by alfalfa and sainfoin, it was 125–134 kg/ha, including symbiotic nitrogen removed by the alfalfa crop – 35–39 kg/ha and Hungarian sainfoin – 37–44 kg/ha. In the fractional composition of nitrogen in the soil of the experimental field after three years of using alfalfa when inoculating seeds with the complex microbial preparation Ecovital, the nitrogen content was the highest compared to other types of crop rotation, including total nitrogen – 1006,3–1428,8, mineral nitrogen – 24,9–46,3; alkaline hydrolyzed nitrogen – 113,8–186,0 mg/kg of soil. The obtained results allow us to conclude that the creation of highly productive symbiotic systems when using the latest biological preparations will contribute to improving soil nitrogen balance, eliminate the catastrophic decrease in fertility and soil degradation, improve the fodder base for animal production, and reduce the ecological burden on agricultural lands.*

**Key words:** *biological nitrogen, symbiotic systems, perennial leguminous grasses, nitrogen regime, extreme weather conditions*

**Relevance of research.** The increase in the production of high-quality plant products in Ukraine, given the limited supply of energy products and the constant increase in their prices, is generally solved by further increasing the production of mineral nitrogen fertilizers. The production of nitrogen fertilizers is an energy-intensive process since producing 1 ton of mineral nitrogen fertilizers consumes about 6 million kcal, which is as much as 5 tons of hard coal when burning. Over the past 10 years, the rate of mineral nitrogen fertilizers application

was 97 kg/ha of active substance (a.d.), against 141 kg/ha in 1990 [1]. Since the beginning of the full-scale Russian invasion of Ukraine, only two factories – “Rivneazot” and Cherkasy “Azot” have produced nitrogen fertilizers. Therefore, the volume of mineral fertilizer application will continue to decrease as a result of the challenges of wartime.

When cultivating most crops in recent years, mostly mineral fertilizers were used, while applying organic fertilizers significantly decreased [2]. The latter is associated with a

significant reduction in cattle population in all regions of Ukraine. In 1990, with a cattle population of 25,2 million heads (including 8,5 million cows) in Ukraine, 257,1 million tons of organic fertilizers were produced and 8,6 tons of organic matter were applied per hectare of sown area. In general, during the period 1990–2020, the number of dairy herds in Ukraine decreased by 6,4 million heads, that is 74,6 %. The production of organic fertilizers decreased to 9,8 million tons in recent years, due to which only 0,5 tons of organic and 41 kg/ha (dry matter) of mineral fertilizers were applied per hectare of sown area [3–5]. A decrease in nitrogen application rates disrupts the ratio of carbon and nitrogen in the soil; it leads to the situation when plants compensate for the lack of nitrogen by consuming it from humus compounds. Such processes cause dehumification and degradation of arable land [4].

Currently, there are two ways to improve the nitrogen balance in the soil in Ukraine: a) expansion of the production of nitrogen mineral fertilizers on an industrial basis, b) creation of agrotechnological conditions for increasing the sown area of leguminous crops and perennial leguminous grasses, characterized by the maximum nitrogen-fixing activity of symbiotic and free-living diazotrophs [6–8]. Expanding the sown areas of perennial leguminous grasses, which provide high nitrogen-fixing activity of symbiotic and free-living bacteria, is an extremely urgent issue in modern farming conditions [9]. In recent years, there has been an increase in the aridity of the climate and xerophytization of vegetation. Therefore, the development of measures to ensure the stable production of fodder products in the conditions of global climate change is becoming increasingly urgent [10]. Despite the relevance of these issues in the conditions of Southern Ukraine, they remain little studied.

**Analysis of recent research and publications.** Nitrogen in the earth's crust is in the form of various compounds, which make up 0,005 % of the earth's crust mass [11]. The budget, distribution, and evolution of the earth's nitrogen are regulated by biological and geological cycles. The biological cycle provides the geological cycle with nitrogen, which, in turn, supplies part of the nitrogen to the Earth's interior [12]. It is known, that the main mass of nitrogen is in the air of the atmosphere, where molecular nitrogen is up to 78.08 %, the mass of which is  $4 \times 10^{15}$  tons. However, plants consume only nitrogen from mineral compounds and, due to the inability

to assimilate molecular nitrogen from the air, they often experience its deficiency.

Leguminous crops play an important role in soil nitrogen balance. During symbiotic nitrogen fixation from the air by nodule bacteria, leguminous crops, and nodule bacteria function in a close interaction in a biological system, and a connecting electron and transport chain between them is leghemoglobin. Leguminous crops are a source of carbon-containing compounds – products of photosynthesis, which are energy material, necessary for activating and reduction  $N_2$  to  $NH_4^+$ .

Among the leguminous grasses, clover, birdsfoot deer vetch, and Hungarian sainfoin are characterized by high productivity, but in this regard, alfalfa is considered the most effective [13–16]. Perennial legumes in symbiosis with nodule bacteria, which fix atmospheric nitrogen, accumulate it in phytomass. In the experiments with short crop rotations, it was found that biological nitrogen compensates the total removed amount of nitrogen with the yield in crop rotations with peas and soybeans by 25–62 % and with alfalfa by 89 % [17]. In the post-harvest and root residues of the specified types of leguminous perennial grasses, up to 20–30 t/ha of organic matter is accumulated, which is equivalent to 70–80 t/ha of high-quality humus, due to which winter wheat after alfalfa, without the use of mineral nitrogen fertilizers, forms a grain yield of up to 3,5–4,5 t/ha [18]. The amount of nitrogen accumulated by annual and perennial leguminous plants was determined in a long-term field experiment on typical black soil: for 3 years, alfalfa in a seven-field crop rotation accumulated 39,7–43,5 kg of nitrogen per hectare for a year in the form of above-ground biomass; during that period, the following crops took 14,9 kg of nitrogen from the soil [19].

The temperature regime and the thermotolerance of various strains of nodule bacteria, which have their specific temperature optimums for development and active nitrogen fixation, play an important role in the interaction of nodule bacteria and leguminous plants. The maximum nitrogen fixation of leguminous plants is observed at a temperature of 20–25 °C, and 30 °C and above, there is a decrease in nitrogen accumulation.

Among leguminous perennial grasses, alfalfa and Hungarian sainfoin are characterized by the highest potential of symbiotic nitrogen fixation [13]. Inoculation of leguminous perennial grass seeds with microbial preparations based on effective strains of nodule bacteria before sowing is economically and ecologically appropriate

since the indicated crops during the second and third years of cultivation form an effective symbiotic apparatus and fully provide themselves with nitrogen, forming high yields of both above-ground mass and seeds. The expansion of the sown areas of leguminous grasses (alfalfa) ensures an increase in the yield of the least energy-intensive plant protein, an increase in soil fertility and, in general, a decrease in the anthropogenic load on agricultural land. In the EU countries with highly developed agriculture, up to 20–25 % of arable and natural fodder lands are under leguminous perennial grasses [20].

Comparative studies of the nitrogen balance and productivity of leguminous and non-leguminous crop cultivation systems in European agriculture showed that the highest contribution of biologically fixed nitrogen is achieved when the share of legumes in a crop rotation is about 50 % [21]. The ecologists emphasize that along with the increase in the production of organic feed, the cultivation of leguminous crops contributes to the improvement of the environmental ecological balance, and the preservation of the biodiversity of plants and animals [22–24]. Studies carried out in different climatic zones with different types of leguminous grasses revealed that the inclusion of leguminous crops in grass mixtures increases the productivity of meadows by 1,5–2,5 times and the yield of crude protein by 2–3 times compared to cereals grass stands [25–27].

The role of perennial leguminous grasses alfalfa and safflower in the nitrogen regime on non-irrigated lands of the southern Steppe of Ukraine has not been sufficiently studied.

**The purpose of the research** is to study the accumulation of biologically bound nitrogen by alfalfa and Hungarian sainfoin in single-species crops, as well as in wheatgrass-alfalfa and wheatgrass – sainfoin mixtures on the dark chestnut soil of Southern Ukraine.

**Research materials and methods.** Field and laboratory studies on the productivity of blue hybrid alfalfa, sand Hungarian sainfoin, and intermediate wheatgrass in single-species crops and wheatgrass-alfalfa and wheatgrass – sainfoin mixtures were conducted on the dark chestnut soil of the experimental farm “Kopani” of the Institute of Irrigated Agriculture of the National Academy of Agrarian Sciences of Ukraine

The area of the experimental farm “Kopani” of Bilozersky District, the Kherson Region is located in the Black Sea Lowland. The soil-forming process in the greater part of the farm area was going in the conditions of a flat topography on carbonate-forest underlying bedrocks. Soils

of different fertility, including dark chestnut, were formed in the farm territory, influenced by climate, vegetation, bedrock, groundwater depth, and human economic activity.

The sowing of perennial grasses in a two-factor field experiment was carried out in the early spring of 2020; the experiments were performed in the growing seasons of 2021–2022. The seed sowing rates in crops were (kg/ha): intermediate wheatgrass (variety Vitas) – 32,0, blue hybrid alfalfa (Unitro variety) – 24,0, Hungarian sainfoin (Ingulsky variety) – 90; in grass mixtures: wheatgrass + blue hybrid alfalfa – 16,0 and 12,0, respectively; intermediate wheatgrass + Hungarian sainfoin – 16,0 and 45,0; intermediate wheatgrass + blue hybrid alfalfa + Hungarian sainfoin – 12,0, 8,0 and 30,0. The experiment was laid out by the method of split plots, where the main plots (plots of the first order) are single-species crops of perennial grasses, and sub-plots (plots of the second order) are grass mixtures. The area of the sowing plot was 60 m<sup>2</sup>, and the accounting area was 20 m<sup>2</sup>. Inoculation of seeds of perennial legumes was carried out with the complex preparation Ecovital, made based on complementary strains of nodule bacteria and phosphate-mobilizing bacilli from the collection of the D. K. Zabolotny Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine.

The content of dry matter in plants before mowing was determined by the weight method in two non-adjacent repetitions by the variants of the field experiment. Forage evaluation of the nutritional value of monospecies crops of blue hybrid alfalfa and Hungarian sainfoin and their grass mixtures with wheatgrass was performed when determining the chemical composition of perennial grasses and grass mixtures. Chemical analysis of fodder was made by infrared spectroscopy on a NIP Systems 4500 analyzer. The content of crude and digestible protein, crude fat, crude fiber, crude ash, as well as fodder units, non-nitrogenous extractive compounds, gross and exchangeable energy, and fodder mineral composition were determined in the dry matter. After determining the total moisture, when hygroscopic moisture and air-dry matter in the samples were taken into account, recalculation of nutrient content in fodder as a percentage of absolutely dry matter was performed. Fodder units and gross and exchangeable energy were calculated based on the chemical composition of plants using the coefficients of nutrient productive action and digestibility [28].

The amount of symbiotic nitrogen fixed from the atmosphere by blue hybrid alfalfa of the

Unitro variety and Hungarian sainfoin of the Ingulsky variety was determined using a modified comparative method [29]. The difference between the nitrogen content (removal) in the above-ground mass and roots of alfalfa or Hungarian sainfoin and intermediate wheatgrass of the Vitas variety was determined with correction for seed nitrogen. Since under the same conditions of their cultivation, the specified crops consumed the same amount of nitrogen from the soil, the difference in nitrogen uptake by leguminous grasses and wheatgrass was attributed to symbiotic nitrogen fixed from the air. The coefficient of nitrogen fixation was calculated based on the ratio of the growth of nitrogen removal by leguminous perennial grasses to the total crop nitrogen removal. Statistical processing of the obtained data was performed according to [30].

**Research results and their discussion.** It was established that along with a high yield of fodder units and digestible protein, the sowing of leguminous perennial grasses contributed to the accumulation of symbiotic nitrogen, which during all the years of field experiments significantly depended on the yield of absolutely dry matter, the nitrogen content of plants, the lack of moisture supply and the duration of the use of grass stands.

The symbiotic fixation of nitrogen by single-species seed crops of blue hybrid alfalfa and Hungarian sainfoin of the first and second years of use was studied in years with different precipitation availability. By hydrothermal conditions, 2021 was classified as wet (50 %) in terms of precipitation availability, and 2022 – as moderately dry (75 %). The symbiotic fixation of nitrogen by single-species crops of blue hybrid alfalfa, Hungarian sainfoin, and wheatgrass + alfalfa and wheatgrass + sainfoin grass mixtures depended significantly on the change in the species botanical composition of the grass stands over the years of use and the nitrogen content in perennial grasses. Nitrogen content in single-species crops of blue hybrid alfalfa, Hungarian sainfoin, and intermediate wheatgrass and their grass mixtures depended significantly on the hydrothermal conditions of the growing season of each year of the field experiment. Compared to single-species wheatgrass crops, the nitrogen content in leguminous perennial grasses during all years of the field experiment was significantly higher.

The average nitrogen content in single-species wheatgrass crops in the first year of use was 2,73 and 2,77 %, and in the second year – 2,62 and 2,66 %. In single-species alfalfa crops nitrogen content increased to 3,54 and 3,75 % in the first year and 3,60 and 3,64 in the second

year. In single-species Hungarian sainfoin crops nitrogen content also increased up to 3,62 and 3,65 % in the first year and up to 3,49 and 3,63 % of absolutely dry matter in the second year.

During the first and second years of use, the removal of total nitrogen by the intermediate wheatgrass did not exceed 90 kg/ha, by the blue hybrid alfalfa and Hungarian sainfoin – 125 and 129 kg/ha, and 127 and 134 kg/ha, respectively; including the removal of symbiotic nitrogen by blue hybrid alfalfa – 35 and 39 kg/ha and Hungarian sainfoin – 37 and 44 kg/ha. The removal of symbiotic nitrogen over the years of the study by the intermediate wheatgrass + blue hybrid alfalfa grass mixture was 38 and 41 kg/ha, by the intermediate wheatgrass + Hungarian sainfoin grass mixture – 33 and 41 kg/ha, and by the intermediate wheatgrass + blue hybrid alfalfa + Hungarian sainfoin three-component grass mixture – 37 and 44 kg/ha (Table 1).

The coefficient of nitrogen fixation, as the ratio of the growth of nitrogen removal by leguminous perennial grasses to the total crop removal, under conditions of natural moistening (without irrigation) in the southern part of the Steppe zone, significantly depended on presence of alfalfa and Hungarian sainfoin in the species botanical composition, precipitation availability, and the duration of the use of grass stands over the years. For blue-hybrid alfalfa of the Unitro variety in the first and second year of use, it was 28,0 and 30,2 % respectively, and for Hungarian sainfoin of the Ingulsky variety – 29,1 and 32,8 % respectively (Table 1).

The cultivation of single-species crops of blue hybrid alfalfa, Hungarian sainfoin, and their grass mixtures with intermediate wheatgrass made it possible to obtain green fodder, balanced with digestible protein, without applying mineral nutrients and to provide cereal grain crops with the best forecrops during all the years of scientific research.

The determined fractional composition of nitrogen in the 0–20 cm and 20–40 cm layers of the dark chestnut soil of the experimental field of EE “EF “Kopani” after three years of using alfalfa when inoculating seeds with Ecovital shows that, compared to other types of the crop rotation, it was the highest and, depending on the soil layer, consisted of: total – 1006,3 and 1428,8 mg/kg, mineral – 24,9 and 46,3, alkaline hydrolyzed – 113,8 and 186,0, heavily hydrolyzed – 155,5 and 214,4 and non-hydrolyzed nitrogen – 712,1 and 982,1 mg/kg of soil (Table 2).

Sufficient accumulation of all forms of nitrogen in the soil is due to inoculating alfalfa seeds with the complex biological preparation Ecovital.



## 1. Accumulation of symbiotic nitrogen by perennial legumes in the first and second years of use

Types of grass and grass mixtures	Removal of nitrogen by yield and roots			Nitrogen fixation coefficient, %	Equivalent to mineral nitrogen in	
	Total removal		Including symbiotic nitrogen, kg/ha		kg/ha	GJ/ha
	kg/ha	%				
First year of use (2021)						
Intermediate wheatgrass	90	100	–	–	–	–
Alfalfa	125	139	35	28,0	105	9,1
Intermediate wheatgrass + Alfalfa	128	142	38	29,7	114	9,9
Hungarian sainfoin	127	141	37	29,1	111	9,6
Intermediate wheatgrass + Hungarian sainfoin	123	137	33	26,8	99	8,6
Intermediate wheatgrass + Alfalfa + Hungarian sainfoin	127	141	37	29,1	111	9,6
LSD <sub>05</sub>	13,8	15,4	2,0	1,3	6,1	0,3
Second year of use (2022)						
Intermediate wheatgrass	90	100	–	–	–	–
Alfalfa	129	143	39	30,2	117	10,1
Intermediate wheatgrass + Alfalfa	131	145	41	31,3	123	10,7
Hungarian sainfoin	134	149	44	32,8	132	11,4
Intermediate wheatgrass + Hungarian sainfoin	131	145	41	31,3	123	10,7
Intermediate wheatgrass + Alfalfa + Hungarian sainfoin	134	149	44	32,8	132	11,4
LSD <sub>05</sub>	19,5	20,9	2,2	1,1	6,7	0,6
On average for 2021–2022						
Intermediate wheatgrass	90	100	–	–	–	–
Alfalfa	127	141	37	29,1	111	9,6
Intermediate wheatgrass + Alfalfa	130	144	40	30,5	119	10,3
Hungarian sainfoin	131	145	41	31,0	122	10,5
Intermediate wheatgrass + Hungarian sainfoin	127	141	37	29,1	111	9,7
Intermediate wheatgrass + Alfalfa + Hungarian sainfoin	131	145	41	31,0	122	10,5
LSD <sub>05</sub>	16,7	18,2	2,1	1,2	6,4	0,5

## 2. Fractional composition of nitrogen in different types of crop rotation (EE “EF “Kopani” of the Institute of Irrigated Agriculture of NAS)

Type of crop rotation	Soil layer depth, cm	Fractional composition of nitrogen, mg/kg				
		total	mineral*	alkaline hydrolyzed	heavily hydrolyzed	non-hydrolyzed
Alfalfa	0–20	1428,8	46,3	186,0	214,4	982,1
	20–40	1006,3	24,9	113,8	155,5	712,1
Winter wheat	0–20	1176,0	19,2	121,2	179,7	855,9
	20–40	892,0	21,1	95,1	132,6	643,2
Sunflower	0–20	1123,0	22,3	110,7	168,4	821,6
	20–40	834,0	12,6	81,6	127,1	612,7
Black fallow	0–20	1231,0	39,4	146,4	170,8	874,4
	20–40	917,0	25,1	99,5	134,3	658,1
LSD <sub>05</sub> (0–20 cm), mg/kg		152,90	15,03	38,34	24,36	79,32
LSD <sub>05</sub> (20–40 cm), mg/kg		82,04	6,69	15,19	14,28	47,62

\*Note: mineral nitrogen – (N –NO<sub>3</sub>+N–NH<sub>4</sub>).

The high content of mineral and alkaline hydrolyzed nitrogen in the dark chestnut soil during the cultivation of alfalfa for fodder purposes and seeds makes it possible to obtain sufficiently high yields of grain crops, winter rape, and sunflower in conditions of natural moisture supply without applying mineral nitrogen fertilizers. Therefore, the expansion of alfalfa sown areas in combination with complex inoculation in modern farming conditions is one of the most effective solutions to overcome the difficult situation that small-scale and private farms of the Southern Steppe of Ukraine have faced in recent years.

Restoration of degraded dark-chestnut soil fertility was achieved by the long-term use of drought-resistant perennial leguminous grasses, namely blue hybrid alfalfa and Hungarian sainfoin, which are the most adapted to the natural and climatic conditions of the Southern Steppe subzone. At the same time, high productivity of single-species crops of blue hybrid alfalfa in the range of 1,33–2,67 t/ha of fodder units and 0,30–0,62 t/ha of digestible protein, in the southern part of the Steppe zone was obtained under conditions of natural water supply (without irrigation).

The obtained results emphasize the important role of perennial legumes in increasing the production of fodder protein, which was confirmed by the experiment results in different natural and climatic zones of Ukraine [31, 32]. An increase in the productivity of perennial legumes is provided by the pre-sowing treatment of seeds with complementary strains of nodule bacteria [33]. As shown by the results of our research,

the accumulation of nitrogen by perennial leguminous grasses is significantly activated when using nodule bacteria in a complex with phosphate-mobilizing bacteria. In the arid climate of southern Ukraine, the complex application of nodule and phosphate-mobilizing bacteria contributed not only to effective symbiotic nitrogen fixation but also increased the resistance of plants to adverse weather conditions. It is an important factor that ensures the sustainable development of agriculture in the conditions of climate aridization, which has been observed in recent years in the Steppe zone.

**Conclusions.** Cultivation of single-species crops of blue-hybrid alfalfa, Hungarian sainfoin, and their grass mixtures with wheatgrass when using complex bacterial preparations allows us to accumulate 37–41 kg/ha of symbiotic nitrogen, obtain nitrogen-enriched green fodder containing 141–145 kg/ha of nitrogen in the phytomass. The creation of highly productive symbiotic systems of single-species agrophytocenoses of perennial leguminous grasses and leguminous-cereal grass mixtures, resistant to adverse climatic factors, in combination with the introduction of the latest biological preparations, contributes to increasing the content of mineral (nitrate and ammonium) and alkaline hydrolyzed nitrogen in the soil. Increasing the sown area with alfalfa, alfalfa-cereal, and sainfoin-cereal grass mixtures will avoid a decrease in soil fertility and soil degradation, improve the fodder base for animal production, reduce the use of nitrogen mineral fertilizers, decrease the ecological burden on agricultural land, and provide grain, technical and vegetable crops with the best forecrops.

### References

1. Derzhavna sluzhba statystyky Ukrainy. (2016). Osnovni ekonomichni pokaznyky vyrobnytstva produktsii silskoho hospodarstva v silhospiddpryiemstvakh za 1990–2016 – Statystychnyi biuleten [Main economic indicators of agricultural production in agricultural enterprises for 1990–2016. – Statistical bulletin]. Kyiv. Retrieved from: [www.ukrstat.gov.ua](http://www.ukrstat.gov.ua). [in Ukrainian].
2. Tarariko, O.G. (1999). Teoriia i praktyka udoskonalennia struktury zemlekorystuvannia v konteksti konservatsii erodovanykh ornykh zemel i zbilshennia ploshchi kormovykh uhid [Theory and practice of improving the land use structure in the context of conservation of eroded arable land and increasing the area of fodder lands]. *Kormy i kormovyrobnytstvo*, 46, 72–78. [in Ukrainian].
3. Baliuk, S.A., Tymchenko, D.O., & Hychka, M.M. (2009). Kontseptsiiia okhorony gruntiv vid erozii v Ukraini [The concept of soil erosion protection in Ukraine]. *Visnyk ahrarnoi nauky*, 2, 5–10 [in Ukrainian].
4. Baliuk, S.A., Medvediev, V.V., & Tarariko, O.G. (2011). Posibnyk ukrainskoho khliboroba. Pro stan rodiuchosti gruntiv Ukrainy [On the state of soil fertility in Ukraine]. Kyiv, 41–69 [in Ukrainian].
5. Goloborodko, S.P. (1999). Chomu problemne nasinnytstvo liutserny v Ukraini [Why lucerne seed production is problematic in Ukraine]. *Nasinnytstvo kormovykh kultur v suchasnykh umovakh hospodariuvannia. Materialy Vseukr. nauk.- prakt. seminaru*. Kyiv: Nora Print, 23–24. [in Ukrainian].
6. Adamen, F.F. (1999). Azotfiksatsiia ta osnovni napriamky polipshennia azotnoho balansu gruntiv [Nitrogen fixation and key areas for improving soil nitrogen balance]. *Visnyk ahrarnoi nauky*. K.: Ahrarna nauka, 2, 9–16 [in Ukrainian].

7. Antypova, L.K. (1999). Efektyvnist mineralnykh dobryv ta mikrodobryv pry vyroshchuvanni nasinnievoi liutserny v umovakh pivdennoho Stepu Ukrainy [Efficiency of mineral fertilisers and micronutrient fertilisers in the cultivation of seed alfalfa in the southern steppe of Ukraine]. *Zbirnyk naukovykh prats Mykolaivskoi derzhavnoi silskohospodarskoi stantsii*. Kyiv: BMT, 213–221 [in Ukrainian].
8. Antypova, L.K. (1999). Udoskonalennia systemy zhyvlennia nasinnievoi liutserny na chornozemakh pivdennykh [Improvement of the seed lucerne nutrition system on southern black soils]. *Visnyk ahrarnoi nauky*, 6, 33–35 [in Ukrainian].
9. Medvediev, V.V. (2001). Stan rodiuchosti gruntiv Ukrainy ta prohnoz yoho zmin za umov suchasnoho zemlerobstva [The state of soil fertility in Ukraine and forecast of its changes in the conditions of modern agriculture] Kharkiv: Shtrykh. [in Ukrainian].
10. Petrychenko, V., Bohovin, A., & Kurhak, V. (2012). More efficient use of grassland under climate warming. Grassland – a European Resource? *Proceed. XXIV General Meeting of the European Grassland Federation*. Lublin, Poland. 3–7 June, 151–153 [in Ukrainian].
11. Boocock, T. J., Mikhail, S., Boyce, A. J., Prytulak, J., Savage, P. S., & Stüeken, E. E. (2023). A primary magmatic source of nitrogen to Earth's crust. *Nature Geoscience*, 16 (6), 521–526. DOI: <https://doi.org/10.1038/s41561-023-01194-3>
12. Mysen, B. (2019). Nitrogen in the Earth: abundance and transport. *Progress in Earth and Planetary Science*, 6 (1), 1–15. DOI: <https://doi.org/10.1186/s40645-019-0286-x>
13. Goloborodko, S.P., & Goloborodko Y.I. (1995). Efektyvnist zastosuvannia kompleksnykh suspenziiovanykh dobryv (laktofolu) pry pozakorenevomu pidzhyvlenni nasinnievoi liutserny [Efficiency of application of complex suspended fertilizers (lactofol) in foliar feeding of lucerne seed] / *Ukraina v svitovykh zemelnykh, prodovolchyykh i kormovykh resursakh i ekonomichnykh vidnosynakh*. Materialy mizhnar. konf. Vinnytsia: Ahrarna nauka, 76 [in Ukrainian].
14. Buhaiiov, V.D., Shcherbyna, L.P., & Bortnovskyyi, V.M. (2003). Osoblyvosti selektsiinoi roboty na pidvyshchennia rivnia zymostiikosti ta posukhostiikosti koniushyny luchnoi [Peculiarities of selection work to increase the level of winter hardiness and drought resistance of meadow clover]. *Kormy i kormovyrobnytstvo*, 51, 7–9 [in Ukrainian].
15. Olifirovych, V.O. (2003). Pidvyshchennia urozhainosti ta zboru proteinu zi starosiianykh travostoiv [Increasing productivity and protein yield from old-seeded grass stands]. *Kormy i kormovyrobnytstvo*, 51, 201–205 [in Ukrainian].
16. Rudnytskyi, B.O. (2003). Udoskonalennia elementiv tekhnolohii vyroshchuvannia bobovykh trav na korm ta nasinnia [Improvement of elements of technologies for growing legumes for feeds and seeds]. *Kormy i kormovyrobnytstvo*, 51, 43–51 [in Ukrainian].
17. Tanchyk, S., Litvinov, D., Butenko, A., Litvinova, O., Pavlov, O., Babenko, A., Shpyrka, N., Onychko, V., Masyk, I., & Onychko, T. (2021). Fixed nitrogen in agriculture and its role in agrocenoses. *Agronomy Research*, 19 (2), 601–611. DOI: <https://doi.org/10.15159/AR.21.086> [in Ukrainian].
18. Andrusenko, I. I. (1987). Struktura posevnyih ploschadey i sevooboroty. [Oroshaemoe zemledelie Structure of sown areas and crop rotations. Irrigated farming]. Ostapov V. I. (Ed.). Kyiv: Urozhai, 45–54. [in Ukrainian].
19. Boincean, B.P., Rusnac, G.T., Boaghii, I.V., Pasat, D.I., & Gavrilas, S. (2013). Legumes as an Alternative Source of Nitrogen for Modern Agriculture. *Soil as World Heritage*, 343–351. DOI: [https://doi.org/10.1007/978-94-007-6187-2\\_33](https://doi.org/10.1007/978-94-007-6187-2_33)
20. Babich, A.O., Pidpalyi, I.F., & Shelest, V.K. (1994). Hospodarska ta bioenerhetychna otsinka tekhnolohii vyroshchuvannia liutserny v umovakh zroshennia [Economic and bioenergy assessment of alfalfa cultivation technologies under irrigation conditions]. *Visnyk ahrarnoi nauky*, 5, 95–102 [in Ukrainian].
21. Iannetta, P.P.M., Young, M., Bachinger, J., Bergkvist, G., Doltra, J., Lopez-Bellido, R.J., Monti, M., Pappa, V.A., Reckling, M., Topp, C.F.E., Walker, R.L., Rees, R.M., Watson, C.A., James, E.K., Squire, G.R., & Begg, G.S. (2016). A Comparative Nitrogen Balance and Productivity Analysis of Legume and Non-legume Supported Cropping Systems: The Potential Role of Biological Nitrogen Fixation. *Frontiers in Plant Science*, 7, 1–13. DOI: <https://doi.org/10.3389/fpls.2016.01700>
22. Demyanchik, V.T. (2012). Dynamics of ecological and faunal potential of reclamation systems in the mode of flooding. Natural environment of Polesie: features and prospects of development. *Coll. N. Polesie Agrarian and Ecological Institute of the National Academy of Sciences of Belarus*, 5, 74–78 [in Ukrainian]
23. Kurhak, V., & Karbivska, U. (2020). Features of formation of bean-cereal agrophytocenoses on sod-podzolic soils of the Carpathian foothills of Ukraine. *Feeds and Feed Production*, 89, 121–133. <https://doi.org/10.31073/kormovyrobnytstvo202089-12>

24. Kurhak, V. H., Panasyuk, S. M., & Asanishvili, N. M. (2020). Influence of perennial legumes on the productivity of meadow phytocenoses. *Ukrainian Journal of Ecology*, 10 (6), 310–315. DOI: [https://doi.org/10.15421/2020\\_298](https://doi.org/10.15421/2020_298)
25. Bohovin, A.V., Kurhak, V.H., & Kletsnyi, O.M. (1997). Rezerv zbilshennia vyrobnytstva travianykh kormiv [Reserve for increasing grass feed production]. *Agroincom*, 8-9, 22–24 [in Ukrainian].
26. Babych, A.O. (1996). Svitovi zemelni, prodovolchi i kormovi resursy [World land, food and feed resources]. Kyiv: Ahrarna nauka [in Ukrainian]
27. Goloborodko, S.P. (1996). Nevykorystani rezervy vyrobnytstva kormiv i kormovoho proteinu v Ukraini [Unused reserves of feed and feed protein production in Ukraine]. *Visnyk ahrarnoi nauky*, 11, 15–19. [in Ukrainian].
28. Medvedovskyi, O.K., Ivanenko, P.I. (1988). Enerhetychnyi analiz intensyvnykh tekhnolohii v silskohospodarskomu vyrobnytstvi [Energy analysis of intensive technologies in agricultural production]. Kyiv: Urozhai. [in Ukrainian].
29. Vozzhehova, R.A., Goloborodko, S.P., & Dymov, O.M. (2021). Ahrobiolohichni osnovy vidtvorennia rodiuchosti dehradovanykh zemel u pivdennomu Stepu Ukrainy [Agrobiological bases of fertility reproduction of degraded lands in the southern Steppe of Ukraine]. Odessa: Oldi Plus [in Ukrainian].
30. Ushkarenko, V.O. Vozzhehova, R.A., Goloborodko, S.P., & Kokovikhin, S.V. (2013). Statystychnyi analiz rezultativ polovykh doslidiv u zemlerobstvi: Monohrafiia [Statistical analysis of the results of field experiments in agriculture: A monograph] Kherson: Ailant [in Ukrainian].
31. Kvitko, H.P. (1999). Vplyv ahroekolohichnykh umov i tekhnolohichnykh pryiomiv na produktyvnist liutserny posivnoi v Lisostepu [The influence of agroecological conditions and technological methods on the productivity of alfalfa in the Forest Steppe]. *Kormy i kormovyrobnytstvo*, 46, 55–65 [in Ukrainian].
32. Petrychenko, V.F. (2007). Teoretychni osnovy intensyfikatsii kormovyrobnytstva v Ukraini [Theoretical foundations of intensification of fodder production in Ukraine]. *Visnyk ahrarnoi nauky*, 10, 19–22 [in Ukrainian].
33. Zapruta, O., Antoniv, S., & Kolisnyk, S. (2023). Uspishne vedennia nasynnytstva bahatorichnykh bobovykh trav – zaporuka rozshyrennia ploshch posivu polovykh ta luchnykh ahrofitotsenoziv [Successful seed management of perennial legumes as the key to expanding the sowing areas of field and meadow agrophytocoenoses]. *Kormy i kormovyrobnytstvo*, 95, 40–52. DOI: <https://doi.org/10.31073/kormovyrobnytstvo202395-03> [in Ukrainian].

УДК 633.31/37:631.847.211(477.7)

## НАКОПИЧЕННЯ СИМБІОТИЧНОГО АЗОТУ БАГАТОРІЧНИМИ БОБОВИМИ ТРАВАМИ В УМОВАХ ПІВДЕННОГО СТЕПУ УКРАЇНИ

Г.О. Іутинська<sup>1</sup>, д. біол. наук; С.П. Голобородько<sup>2</sup>, д. с.-г. наук; Л.В. Титова<sup>3</sup>, канд. біол. наук; О.Д. Дубинська<sup>4</sup>, д. філософії; Н.В. Шевчук<sup>5</sup>, аспірант

<sup>1</sup> Інститут мікробіології і вірусології ім. Д. К. Заболотного НАН України, вул. Академіка Заболотного, 154, 03143, м. Київ, Україна; <https://orcid.org/0000-0001-6692-2946>; e-mail: [galyna.iutynska@gmail.com](mailto:galyna.iutynska@gmail.com);

<sup>2</sup> Інститут кліматично орієнтованого сільського господарства НААН, вул. Омеляновича-Павленка Михайла, 9, 01010, м. Київ, Україна; <https://orcid.org/0000-0002-6968-985X>; e-mail: [goloborodko1939@gmail.com](mailto:goloborodko1939@gmail.com);

<sup>3</sup> Інститут мікробіології і вірусології ім. Д. К. Заболотного НАН України, вул. Академіка Заболотного, 154, 03143, м. Київ, Україна; <https://orcid.org/0000-0003-3131-4355>; e-mail: [ltytova.07@gmail.com](mailto:ltytova.07@gmail.com);

<sup>4</sup> Інститут кліматично орієнтованого сільського господарства НААН, вул. Омеляновича-Павленка Михайла, 9, 01010, м. Київ, Україна; <https://orcid.org/0000-0002-5572-0094>; e-mail: [klenova-dubinskaelena76@ukr.net](mailto:klenova-dubinskaelena76@ukr.net);

<sup>5</sup> Інститут мікробіології і вірусології ім. Д. К. Заболотного НАН України, вул. Академіка Заболотного, 154, 03143, м. Київ, Україна; <https://orcid.org/0000-0001-5574-4961>; e-mail: [nadia.shevchuk.48@ukr.net](mailto:nadia.shevchuk.48@ukr.net)

**Анотація.** Використання посівів багаторічних бобових культур є енергетично і економічно вигідним та екологічно безпечним заходом поліпшення азотного балансу ґрунтів, упровадження якого потребує конкретизації і додаткових досліджень стосовно певних ґрунтово-кліматичних умов і систем землеробства. Метою роботи було вивчення накопичення біологічно зв'язаного азоту люцерною й еспарцетом піщаним в одновидових посівах, а також в пирійно-люцернових



*і пирійно-еспарцетових травосумішках на темно-каштановому ґрунті півдня України. Застосовано лабораторні, польові та статистичні методи дослідження. Отримані результати засвідчили, що накопичення азоту в біомасі бобових багаторічних трав в одновидових посівах люцерни синьогібридної, еспарцету піщаного й пирію середнього та їх травосумішок залежало від гідротермічних умов вегетаційного періоду, зміни видового ботанічного складу за роками використання травостої. У продовж першого і другого років використання вміст азоту в біомасі люцерни становив 3,54–3,75 %, еспарцету піщаного – 3,49–3,65 % і був істотно вищим, ніж в одновидових посівах пирію середнього – 2,62–2,77 % до абсолютно сухої речовини. Винос загального азоту врожаєм пирію середнього за цей період не перевищував 90 кг/га, люцерною та еспарцетом – становив 125–134 кг/га, зокрема симбіотичного азоту врожаєм люцерни – 35–39 кг/га і еспарцету піщаного – 37–44 кг/га. У фракційному складі азоту в ґрунті дослідного поля після трирічного використання люцерни за інокуляції насіння комплексним мікробним препаратом Ековітал вміст азоту був найвищим порівняно з іншими ланками сівозміни, зокрема загального – 1006,3–1428,8, мінерального – 24,9–46,3; лужногідролізованого – 113,8–186,0 мг/кг ґрунту. Отримані результати дають змогу дійти висновку, що створення високопродуктивних симбіотичних систем за умов застосування новітніх біопрепаратів сприятиме поліпшенню азотного балансу ґрунту, допоможуть усунути катастрофічне зниження родючості й деградацію ґрунтів, поліпшить кормову базу для тваринництва, знизить екологічне навантаження на сільськогосподарські угіддя.*

**Ключові слова:** біологічний азот, симбіотичні системи, багаторічні бобові трави, азотний режим, екстремальні погодні умови