

Soil agrochemical and microbiological changes during leguminous forage grass cultivation in the southern steppe of Ukraine

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Abstract. Increased use of arable land during the last century has led to a total decrease in soil fertility on the planet. Dehumification processes are activated in the absence of the supply of organic matter and unbalanced application of mineral fertilizers, ignoring crop rotations, minimizing the area of growing leguminous crops, burning straw in the soil. Therefore, the question of increasing the efficiency of agriculture and reproduction of soil fertility becomes urgent. The goal is to investigate the influence of leguminous forage grasses on changes in the agrochemical and microbiological composition of the soil under different methods of soil cultivation in irrigated and non-irrigated conditions. The research was carried out in the conditions of the Odesa region (Ukraine) during 2022-2024 by the method of a three-factor experiment (factor A - the presence of irrigation; Factor B - the method of cultivation of hryvnia, Factor C - fodder leguminous grasses). It was established that the NO₃ content decreased according to the experiment options during the vegetation period from 12.17-12.31 mg/100 g of soil to 9.97-10.19 mg/100 g of soil. The content of P₂O₅ decreased from 6.27-6.40 mg/100 g of soil to 5.02-5.23 mg/100 g of soil, which represents an average to high level of soil provision. Significant differences were not observed in the use of available potassium. The total number of microorganisms in 1 g of completely dry soil during the growing season increased from 24.89-25.09 million to 29.87-31.20 million. In the experiment variants where irrigation was used, the values of the indicator were slightly higher than in non-irrigated areas, which indicates the positive effect of irrigation on improving the microbiological composition of the soil. The introduction of ecologically safe leguminous fodder crops into crop rotations contributes to the improvement of agrocenoses in the crop rotation of organic farming and ensures the improvement of the ecological and phytosanitary condition of the soil.

Keywords: fodder leguminous grasses, *Melilotus albus* Medik, *Medicago sativa* L, *Onobrychis arenaria* (Kit.) DC, soil microflora.

1 Introduction

Topicality. Soil cover is one of the main components of the environment that performs vital biospheric functions. Soils participate in the process of regulating the quality of surface and underground waters, the composition of atmospheric air, are the habitat of most living organisms on the land surface, provide a favorable environment for humans and the production of agricultural products [1-4].

Soil is a unique organo-mineral, heterogeneous, multi-component natural formation that performs irreplaceable economic-production and biosphere-ecological functions [4, 5].

Possessing the property of fertility, the soil acts as the main means of production in agriculture. Using it as a means of production, a person significantly changes soil formation, directly influencing both soil properties, its modes and fertility, and natural factors that determine soil formation [6, 7].

It is difficult to overestimate the importance of soils not only in the management of agriculture, but also in many other areas of management. Quality soil is a basic component of a country's sustainable development and its food security. At the same time, soils are classified as non-renewable and exhaustible resources, which means that preserving soils in their original state is a guarantee of the well-being of future generations.

Literature review. Increased use of arable land during the last century has led to a total decrease in soil fertility on the planet. This problem is also extremely relevant for Ukraine. Since plants and microorganisms are the mandatory and most active participants in the processes of soil formation, the question of the formation of soil fertility should quite legitimately be considered as a biological one. However, the biological state of many soils in the country today should be recognized as degrading. Dehumification processes are activated in the absence of the supply of organic matter and unbalanced application of mineral fertilizers, ignoring crop rotations, minimizing the area of growing leguminous crops and burning straw in the soil. The composition of soil biocenoses is significantly depleted, there is a reduction to a minimum and even the loss of certain types of useful organisms from them. Many agrocenoses have turned into reservoirs of pathogens. The amplitude of such phenomena causes serious concern and the need to urgently take measures at the state level to optimize the state of agrocenoses in general and soil-forming processes in particular [4, 8].

In the arid conditions of the south of the country, the use of irrigation helps to grow high yields. However, the use of water with increased mineralization > 1.5 g/l for irrigation contributes to salinization of cultivated areas. This is especially true for agricultural lands located in the Ingulets irrigated area, where the mineralization of the Ingulets River water used for irrigation can reach a level of > 4.0 g/l, which makes it impossible to use it for irrigation. Therefore, the issue of increasing the efficiency of agriculture and restoration of soil fertility becomes relevant. [9].

Currently, the presence of flooded land, which occupies 20% of arable land, leads to a significant decrease in the efficiency of using irrigated arable land in the southern moderately dry zone and dry-steppe soil-ecological subzones. They have no drainage, which leads to various types of salinization (chloride, sulfate, carbonate, sodium, and other salinities) and a decrease in soil fertility [4, 9].

In agricultural production, the chemical method of melioration (introduction of phosphogypsum) is mainly used. The complex of special agrotechnical measures includes plantation plowing and a number of other methods that provide an opportunity to activate the carbonate and gypsum layers of the soil. However, the above-mentioned melioration methods have significant drawbacks. Thus, the use of a chemical melioration method leads to environmental pollution due to the content of heavy metals in phosphogypsum. Deep plowing on medium-saline and other types of saline soils can lead to a decrease in soil fertility due to further salinization of the upper layer [4, 5, 10-14].

In the process of transition from traditional to organic farming, the production of agricultural products without the use of chemical fertilizers and pesticides is becoming increasingly important. In this regard, the Institute of Climate Smart Agriculture of the National Academy of Agrarian Sciences of Ukraine has been conducting research for a long time on the development of individual elements and components of biologization of agricultural crop cultivation technology.

Goal. Research conducted at the Institute of Climate-Oriented Agriculture of the National Academy of Sciences of Ukraine aimed at solving an urgent issue - improving the agrocenoses of grain, leguminous and industrial crops in crop rotation of organic farming through the use of ecologically safe agricultural crops, which will ensure the improvement of the ecological and phytosanitary condition of the soil and contribute to increasing the yield of agricultural crops in crop rotation. The goal was to investigate the influence of leguminous forage grasses on changes in the agrochemical and microbiological composition of the soil under different methods of soil cultivation in irrigated and non-irrigated conditions.

The task is to determine the influence of the main tillage methods on the growth and development of leguminous crops in irrigated and non-irrigated conditions and to investigate changes in the agrochemical and microbiological composition of the soil.

2 Methods

The study was conducted in the conditions of Odesa region (Ukraine) in 2022-2024, by the method of a three-factor experiment: factor A - presence of irrigation (A1 - no irrigation, A2 - irrigation); Factor B – soil cultivation (B1 – plowing to a depth of 27-30 cm, B2 – chiselling to a depth of 22-24 cm, B3 – disking to a depth of 15-17 cm); Factor C - fodder leguminous grasses (C1 – Nadiezhda variety of *Medicago sativa* L., C2 – Arsei variety of *Onobrychis arenaria* (Kit.) DC., C3 – Pivdennyi variety of *Melilotus albus* Medik). The following methods were used during the research: field method (to study the interaction of the research object with biotic and abiotic factors of the southern steppe zone); laboratory method (to determine the qualitative and quantitative composition of the microflora and the agrochemical composition of the soil); mathematical and statistical method (for conducting dispersion analysis and statistical data processing in order to assess the reliability of the obtained research results) [15, 16].

3 Results and Discussions

Improvement of ecological and meliorative indicators and phytosanitary condition of dark chestnut soils can be achieved by using forage legumes in crop rotation, which significantly increase soil fertility due to biological fixation of nitrogen from the air and improve its structure. Due to the release of carbon dioxide by the roots of these plants, a chemical process of salinization of the soils of the irrigated area occurs.

The obtained experimental data show that comparing the indicators obtained during the agrochemical analysis of the soil at the beginning of the growing season and before harvesting forage legumes, it can be concluded that the amount of nitrates in the soil had a tendency to decrease at the end of the growing season (Table 1). This is the result of the consumption of nitrate nitrogen for crop formation and leaching with irrigation water.

Thus, at the beginning of the growing season, the NO₃ content varied within the range of 12.17-12.31 mg/100 g of soil, and before harvesting it was equal to 9.97-10.19 mg/100 g of soil (Table 1). At the same time, with the use of irrigation, the amount of NO₃ was slightly lower than in non-irrigated conditions.

Table 1. Results of agrochemical soil analysis at the beginning of the growing season and before harvesting leguminous fodder crops, average for 2022-2024.

Factor A, irrigation	Factor B, methods of basic soil cultivation	Factor C, fodder legumes	Content in soil, mg/100 g					
			at the beginning of the growing season			before harvesting		
			NO ₃	P ₂ O ₅	K ₂ O	NO ₃	P ₂ O ₅	K ₂ O
without irrigation	plowing (27-30 cm)	<i>Medicago sativa L.</i>	12.25	6.33	28.68	10.16	5.16	22.04
		<i>Onobrychis arenaria (Kit.) DC.</i>	12.22	6.31	28.71	10.13	5.14	22.09
		<i>Melilotus albus Medik.</i>	12.28	6.36	28.70	10.14	5.17	22.03
	chiseling (22-24 cm)	<i>Medicago sativa L.</i>	12.24	6.33	28.67	10.18	5.18	22.06
		<i>Onobrychis arenaria (Kit.) DC.</i>	12.21	6.29	28.72	10.17	5.16	22.07
		<i>Medicago sativa L.</i>	12.26	6.34	28.69	10.19	5.14	22.04
	disking (15-17 cm)	<i>Onobrychis arenaria (Kit.) DC.</i>	12.21	6.30	28.67	10.17	5.16	22.08
		<i>Medicago sativa L.</i>	12.17	6.27	28.70	10.15	5.15	22.09
		<i>Onobrychis arenaria (Kit.) DC.</i>	12.20	6.32	28.69	10.16	5.16	22.07
irrigation	plowing (27-30 cm)	<i>Medicago sativa L.</i>	12.28	6.37	28.65	9.98	5.21	22.05
		<i>Onobrychis arenaria (Kit.) DC.</i>	12.26	6.35	28.64	10.01	5.23	22.04
		<i>Medicago sativa L.</i>	12.31	6.40	28.69	9.97	5.19	22.07
	chiseling (22-24 cm)	<i>Onobrychis arenaria (Kit.) DC.</i>	12.27	6.36	28.63	10.03	5.17	22.05
		<i>Medicago sativa L.</i>	12.24	6.33	28.61	10.05	5.18	22.03
		<i>Onobrychis arenaria (Kit.) DC.</i>	12.29	6.38	28.67	10.01	5.15	22.04
	disking (15-17 cm)	<i>Medicago sativa L.</i>	12.25	6.33	28.60	10.04	5.07	22.06
		<i>Onobrychis arenaria (Kit.) DC.</i>	12.21	6.30	28.59	10.06	5.11	22.03
		<i>Medicago sativa L.</i>	12.24	6.35	28.66	10.02	5.02	22.01
Assessment of the significance of partial differences								
LSD ₀₅ , mg/100 g		A = 0.01; B = 0.02; C = 0.01						

When determining the content of mobile phosphorus compounds before harvesting, it was found that there is a tendency for its decrease in the soil, regardless of the investigated factors. Thus, at the beginning of the growing season, the content of P_2O_5 varied between 6.27-6.40 mg/100 g of soil, and before harvesting - 5.02-5.23 mg/100 g of soil, which is an average and high level of soil supply. Removal of mobile compounds P_2O_5 is the result of formation of crop yield.

As for the content of mobile compounds of potassium, it can be noted that at the beginning of the growing season of fodder crops, their content in the soil was within the average supply level. No significant differences were found in the use of available potassium during the growing season by plants of leguminous crops, depending on the investigated factors.

In general, it can be stated that the content of the main nutrients before harvesting was lower than before sowing, which is related to the formation of the crop yield.

Any microbiocenosis consists of microorganisms of different functional and taxonomic groups, which differ in their requirements for environmental conditions, nutrition and energy sources. The quantitative ratio between these groups completely depends on the environmental conditions (abiotic and biotic factors) in which the microbial coenosis is formed.

The ecological state of the soil of the agroecosystem is characterized by various indicators of the level of its biological activity, which depend on the type and fertility of the soil itself, as well as the applied agricultural measures.

It is well known that as a result of the activity of soil microorganisms, the compounds containing nutrients that are not readily available for plant nutrition are gradually transformed into digestible forms. The number of microorganisms in the soil largely depends on the water, heat, and air regimes and on crops grown in crop rotation. Of greatest interest is the microflora that participates in providing plants with nitrogen nutrition from the soil. In the study, we took soil samples at the beginning of the growing season and before harvesting leguminous fodder crops to determine its microbiological composition (Table 2).

According to the obtained data, it was established that the total number of microorganisms in 1 g of completely dry soil at the beginning of the growing season of leguminous crops was 24.89-25.09 million, slightly increased during the growing season for all variants of the experiment, and for the period of harvesting was within 29.87 -31.20 million (Table 2).

It is worth noting that in the variants of the experiment, where irrigation was used, the values of the indicator were slightly higher than in non-irrigated areas, which indicates the positive effect of irrigation on improving the microbiological composition of the soil.

Ammonifiers are a group of microorganisms with different systematic positions that carry out the process of ammonification (decomposition of organic nitrogenous substances with the release of ammonia). They participate in the nitrogen cycle in nature and plant nutrition.

At the beginning of the crop growing season, significant fluctuations in the number of this group of microorganisms were not detected. Their number was in the range of 25.48-25.68 million/g of completely dry soil (Table 2). During the growing season of fodder leguminous grasses, the number of this group of microorganisms increased significantly to 28.68-30.65 million/g of completely dry soil. According to the results of the microbiological analysis of the soil, it was established that the number of ammonifying bacteria was higher under irrigation conditions and amounted to 30.17-30.65 million/g of completely dry soil, which indicates the positive role of irrigation for the reproduction of this group of microorganisms and the growth of leguminous plants.

Oligonitrophils. Microorganisms capable of growing under the conditions of a small amount of bound nitrogen in the environment. Many of these organisms are diazotrophs, that is, they are able to fix atmospheric nitrogen, as well as fodder leguminous grasses.

Table 2. Results of microbiological analysis of soil during the cultivation of leguminous crops, average for 2022-2024.

Factor B, methods of basic soil cultivation	Factor C, fodder legumes	The number of microorganisms in 1 g of completely dry soil							
		at the beginning of the growing season				before harvesting			
		total number, million	ammonifying, million	oligotrophili c, million	nitrifying, thousand	total number, million	ammonifying, million	oligotrophili c, million	nitrifying, thousand
without irrigation									
plowing (27-30 cm)	<i>Medicago sativa L.</i>	24.99	25.65	20.43	8.67	29.99	28.82	26.64	11.14
	<i>Onobrychis arenaria (Kit.) DC.</i>	24.96	25.63	20.41	9.65	29.96	28.80	26.62	11.09
	<i>Melilotus albus Medik.</i>	25.02	25.67	20.45	9.72	30.01	28.85	26.67	11.16
chiseling (22-24 cm)	<i>Medicago sativa L.</i>	24.95	25.54	20.39	8.61	29.96	28.74	26.57	10.93
	<i>Onobrychis arenaria (Kit.) DC.</i>	24.94	25.51	20.40	9.61	29.92	28.71	26.52	10.87
	<i>Melilotus albus Medik.</i>	24.94	25.51	20.40	9.61	29.98	28.77	26.59	10.98
disking (15-17 cm)	<i>Medicago sativa L.</i>	24.91	25.50	20.37	8.60	29.92	28.81	26.37	10.69
	<i>Onobrychis arenaria (Kit.) DC.</i>	24.89	25.48	20.38	9.59	29.87	28.68	26.35	10.62
	<i>Melilotus albus Medik.</i>	24.94	25.53	20.40	9.63	29.95	28.70	26.39	10.70
irrigation									
plowing (27-30 cm)	<i>Medicago sativa L.</i>	25.02	25.68	20.48	8.76	31.19	30.63	29.00	11.62
	<i>Onobrychis arenaria (Kit.) DC.</i>	25.01	25.65	20.47	9.74	31.08	30.59	28.97	11.58
	<i>Melilotus albus Medik.</i>	25.09	25.69	20.50	9.78	31.20	30.65	29.03	11.63
chiseling (22-24 cm)	<i>Medicago sativa L.</i>	24.99	25.63	20.45	8.71	31.16	30.30	28.94	11.54
	<i>Onobrychis arenaria (Kit.) DC.</i>	24.98	25.59	20.44	9.68	31.03	30.25	28.89	11.46
	<i>Melilotus albus Medik.</i>	25.07	25.64	20.49	9.73	31.17	30.31	28.98	11.57

Continuation of table 2

Factor B, methods of basic soil cultivation	Factor C, fodder legumes	The number of microorganisms in 1 g of completely dry soil							
		at the beginning of the growing season				before harvesting			
		total number, million	ammonifying, million	oligonitrophili c, million	nitrifying, thousand	total number, million	ammonifying, million	oligonitrophili c, million	nitrifying, thousand
without irrigation									
disking (15-17 cm)	<i>Medicago sativa L.</i>	24.98	25.61	20.44	8.69	31.14	30.19	28.86	11.35
	<i>Onobrychi s arenaria (Kit.) DC.</i>	24.96	25.57	20.42	9.65	31.01	30.17	28.81	11.32
	<i>Melilotus albus Medik.</i>	25.06	25.63	20.48	9.71	31.15	30.22	28.89	11.35
Assessment of the significance of partial differences									
LSD ₀₅		A = 0.01; B = 0.02; C = 0.01							

At the beginning of the growing season of the crop, the number of oligonitrophils was in the range of 20.37-20.50 million/g of completely dry soil. During the period of growth and development of plants, before harvesting leguminous fodder crops, the number of this group of microorganisms increased in the variants of the experiment where irrigation was used to 28.81-29.03 million/g of completely dry soil, and in non-irrigated conditions to 26.35- 26.67 million/g of completely dry soil, which undoubtedly indicates the positive role of the use of leguminous plants.

Nitrogen plays a decisive role in crop yield formation on all types of soil. The main source of nitrogen nutrition for plants is the nitrate form, which was studied in the experiment under different options for providing moisture and tilling the soil. The root system of plants assimilates nitrogen in nitrate and ammonium forms. The content of these mobile compounds is an indicator of soil fertility [14, 17]. In the arid conditions of the southern steppe zone of Ukraine, the ammonium form of nitrogen is transformed into nitrate under the action of microorganisms. This form of nitrogen does not create poorly soluble salts in the soil, is not absorbed by bulk colloids, which is why it is very mobile and easily leaches into the lower layers of the soil. It is absorbed by plants, which leads to a constant change in the NO₃ content during the growing season of plants.

It is known that perennial leguminous grasses have a comprehensive effect on the improvement of soil fertility indicators: from the accumulation of plant residues in the soil to the symbiotic action of nodule bacteria, which allow to increase the content of nitrogen available to plants. [18, 19].

In our study, there was a tendency to decrease the content of NO₃, P₂O₅, which is explained by the fact that these compounds were consumed by plants for the formation of the crop.

During the growing season of leguminous plants, changes in the microbiological composition of the soil occur. In addition to plants, the soil microbiota is also affected by weather and climate conditions, the use of various elements of plant cultivation technology, including the method of soil cultivation, the presence of irrigation [20-22].

Regarding changes in the microbiological composition of the soil in our study, it should be noted that at the end of the growing season, the number of all groups of microorganisms

that were taken to study the microbiological coenosis of the soil increased. In the irrigation conditions, their number was higher than in the variants of the experiment without irrigation. An increase in the number of all groups of microorganisms was also observed on the experimental variants where plowing was carried out. Thus, the cultivation of fodder leguminous grasses with plowing and the use of irrigation contributed to an increase in the number of all groups of soil microorganisms at the end of the growing season of the crop, which is undoubtedly a positive result of microbiological processes in the development of soil agrocenosis microorganisms.

4 Conclusions

1. The study resolved the issue of improving agrocenoses in crop rotation of organic farming by using ecologically safe fodder leguminous grasses, which will ensure improvement of the ecological and phytosanitary condition of the soil.

2. The amount of nitrates in the soil tended to decrease at the end of the growing season. This is the result of spending nitrate nitrogen on crop formation. At the beginning of the growing season, the NO_3 content varied between 12.17-12.31 mg/100 g of soil, and before harvesting it was equal to 9.97-10.19 mg/100 g of soil.

3. It was established that there is a tendency to decrease the phosphorus content in the soil, regardless of the investigated factors. At the beginning of the growing season, the content of P_2O_5 was 6.27-6.40 mg/100 g of soil, and before harvesting it was 5.02-5.23 mg/100 g of soil, which is a medium and high level of soil supply.

4. No significant differences were found in the use of available potassium during the growing season by forage leguminous plants, depending on the studied factors.

5. It was determined that the total number of microorganisms in 1 g of completely dry soil at the beginning of the growing season of plants was 24.89-25.09 million and slightly increased during the growing season for all variants of the experiment and for the period of harvesting was within 29.87-31.20 million. In the variants of the experiment, where irrigation was used, the values of the indicator were slightly higher than in non-irrigated areas, which indicates the positive effect of irrigation on improving the microbiological composition of the soil.

6. The number of ammonifying bacteria was higher under irrigation conditions and amounted to 30.17-30.65 million/g of completely dry soil, which indicates the positive role of irrigation for the reproduction of this group of microorganisms and the growth of leguminous plants.

7. Over the period of growth and development of plants, the number of oligonitrophils increased in the variants of the experiment where irrigation was applied to 28.81-29.03 million/g of completely dry soil, and in non-irrigated conditions to 26.35-26.67 million/g completely dry soil, which undoubtedly indicates the positive role of the use of fodder legumes.

8. The number of nitrifying bacteria under irrigation conditions was slightly higher than without irrigation. At the beginning of the growing season of plants, it ranged from 8.58 to 9.76 thousand per gram of completely dry soil, while in non-irrigated conditions it ranged from 8.69 to 9.78 thousand/g of completely dry soil. Before harvesting crops, the number of this group of microorganisms increased significantly in irrigated conditions and amounted to 11.32-11.63 thousand/g of completely dry soil, while in non-irrigated conditions the corresponding values of the indicator were 10.62-11.16 thousand/g of completely dry soil, which indicates the positive microbiological role of fodder legumes and irrigation.

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