**Influence of perennial legumes on the productivity of meadow phytocenoses**

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We investigated the effectiveness of soil liming during grass sowing and application of inoculation of perennial legume grass seeds with nitrogen-fixing and phosphate-mobilizing preparations. Methods. Field studies were conducted during 2016-2020 in the National Scientific Center "Institute of Farming of the National Academy of Agrarian Sciences of Ukraine" (northern part of the Forest-steppe of Ukraine, Chabany settlement, Kiev-Svyatoshinsky district of Kyiv region). Area of the sowing plot was 30 m², control plot - 25 m². Repetition of experience four times. Technology of cultivation of perennial grasses and their mixtures was generally accepted. Nitrogen fertilizers in the form of ammonium nitrate containing 34% of active substance, phosphorus as simple superphosphate (18.7%) and potassium as potassium chloride (60%) were applied annually in early spring according to the experimental scheme. Liming was carried out by the lime application under pre-sowing cultivation during perennial grass sowing in a dose of 5 t/ha. Seeds of perennial legume grasses were inoculated with the strains of nodule bacteria, according to the technological instructions, immediately before the sowing. Dry weight content was determined by thermostatic weight method at 105 °C. Green mass yield was accounted by weight method, by weighing with subsequent recalculation of yield from 1 ha of dry weight. Results. The productivity of monogenic sowings of perennial leguminous grasses and their mixtures with awnless brome grass in the variant without fertilization varied within the range of 5.12-8.38 tons, which is 1.7-2.5 times higher compared to the cereal grass stand which was formed from monogenic sowings of awnless brome grass, for the five years on the average and for the yield from 1 ha of dry weight. The highest productivity and the level of accumulation of symbiotic nitrogen (191-266 kg/ha) were obtained in agrocenoses with alfalfa and the lowest one - with meadow clover.  
*Key words*: perennial leguminous grasses, bacterial preparations, organic grassland, productivity, dry mass, liming.

**Introduction**

Due to the deepening ecological crisis, there is a steady trend of increasing demand for food organic production of agricultural products. Organic grassland farming is not only a set of measures aimed at obtaining environmentally safe forage. Organic grassland farming is closely integrated into the context of sustainable development and environmental management. It also includes a set of measures to preserve the environment, where a huge environmental and stabilizing role in agrolandscapes meadows, protecting the soil from erosion and sources from siltation and pollution (Kurgak, 2013). It is the reduction of ploughed land, the reproduction of meadows and a network of sanctuaries, nature reserves, hunting grounds, along with an increase in the production of organic fodder, helps to improve the ecological balance of the environment, the conservation of biodiversity of plants, animals, in particular species included in the Red Book (Kurgak, 2019; Mkhalchuk, 2010; Demyanchik, 2012). Taking into account the huge unused potential of Ukrainian grasslands, (about 8 million hectares with migrations), in the direction of obtaining organic fodder products on them and transfer them from the category of the so-called "abandoned" lands in the fodder-producing, nature conservation, recreational and ecological zones. This will not only turn them into exemplary fodder crops and at the same time conservation elements of agrolandscapes on the basis of adaptive conservation ecological and recreational land use (Kurgak, 2019).  
Natural forage lands in the vast majority as an ecologically clean area and a source of environmentally safe forage is an important component of ecological tourism, in combination with organic production meets the requirements of sustainable development and rational nature management (Parente, 2012; Kurgak, 2013). In line with the development of the tourism industry it is necessary to design and implement a program for the development of ecological tourism, and in particular agritourism, in which considerable attention is paid to the quality of food as an important element of the quality of service.
An integral component of organic production is the development and implementation of a set of measures aimed at the conservation of flora and fauna of the onion grasslands by creating a network of nature reserves, wildlife refuges, hunting and deer farms with keeping and grazing animals in cages (Tyshekevich, 2010). In organic fodder production great attention should be paid to quality control of fodder by certified laboratories. Fodder should meet the requirements of state standards of Ukraine, where control is provided not only on the main indicators of quality, but also on safety indicators (Kurgak, 2013, 2019; Demidass, 2020).

In recent years, due to the increasing processes of climate aridization and xerophytization of vegetation to ensure sustainable development there is a need to search and introduce drought-resistant species from the group of mesoxerophytes and xeromorphophytes in the Forest-steppe and even Polesie for stable organic fodder production should be introduced drought-resistant species of fodder plants or irrigation (Petrychenko, 2012). Organic feed production combined with the corresponding direction of livestock, is an inseparable link between crop and livestock production and an important element of organic production in general. For Ukraine, the development of organic feed production should go hand in hand with the development of cattle breeding and livestock and increase the number of cattle and not only the dairy, but the meat direction. Livestock provides an opportunity to obtain organic fertilizers, which are the source of receipt of nutrients in the soil for organic production. Optimal load per 1 hectare of forage land to bring up to 1.5 cattle or 15 sheep, which will produce and introduce 10 tons of organic matter into the soil, which will ensure high and sufficient yields not only crops of forage group, but also the other crops (Kurgak, 2013).

Natural forage lands and hayfields and pastures placed on them are a complex integral system, in which all components are functionally closely connected with each other by exchange processes of substance and energy flows. Laws, which are characteristic for natural meadows, are in the basis of existence and sowing meadows (Mirkin, 1986; Kurgak, 2010, 2017; Petrichenko, 2013). When forming sowing herbage, as a rule, perennial grasses and legumes should be included before herbage mixtures, taking into account the basic principles of component selection. Creation of sown grasses with an increased content of legumes is one of the most promising directions of organic grassland farming in the world (Hamacher, 2016; Weiß, 2016; Wallenhammar, 2018; Karbivska, 2020). Inclusion of legume grasses in legume-grass censoses without mineral nitrogen application increases the productivity of grasslands by 1.5-2.5 and by 2-3 times in terms of protein collection compared to cereal grasses (Nilsdotter-Linde, 2016; Karbivska et al., 2020a, 2020b; Kovtun, 2020). It is known that legume grasses are not durable. Therefore, the main provisions and practical measures to increase their productive longevity in sown meadow legume-grass agrocnoses are formulated (Kurgak, 2010, 2018, 2020; Petrichenko, 2013; Damborg, 2016; Hynes, 2018). The productive longevity of legumes is influenced not only by the correct selection of components, but also by the ways of placement of cereal and legume components (the best way with alternate placement of cereal and legume components in separate rows from non-widespread rows or narrow strips), and effective interannual combination of mineral and symbiotic nitrogen.

Despite the significant amount of research issues of substantiation of conceptual and technological foundations of organic grasslanding in Ukraine and identifying the role of perennial legumes in increasing the productivity of meadow phytocenoses until recently remain insufficiently studied, which was the subject of our research, are covered in the article.

Materials and methods

Field studies were conducted during 2016-2020. In the National Scientific Center "Institute of Farming of the National Academy of Agrarian Sciences of Ukraine" (northern part of the Forest-steppe of Ukraine, Chabany settlement, Kiev-Svyatoshinsky district of Kiev region). Soil of the research site is the dark gray podzolized coarse-pilvurate light loam on loess-like loam. The depth of humus horizon is 35-40 cm. Humus content in 0-20 cm layer was 2.4%, pH5.2, hydrolytic acidity 4.2 mg-eq/100 g, lunohydrolyzed nitrogen 13.1, labile phosphorus 17.1, and exchangeable potassium 12.9 mg per 100 g of soil. Depth of groundwater was about 3 m. Relief was flat with a barely noticeable slope towards the Dnieper River. The climate of the region where the study was conducted is moderately continental. Average annual air temperature is 6-7 °C. Average annual precipitation is 480-620 mm. Weather conditions in Chabany settlement, during the years of research (2016-2020) were favorable for growth and development of perennial grasses, despite the fact that during the year and the growing season air temperature exceeded by 1.5-2.0 °C, and the amount of precipitation was less than the average perennial rate. Scheme of experiments is given in the following tables. Area of the sowing plot is 30 m, recording area is 25 m². Repetition of the experiment is four times. Cultivation technology of perennial grasses and their mixtures, excluding the studied factors was generally accepted. Nitrogen fertilizers in the form of ammonium nitrate with the active substance content of 34%, phosphorus as simple superphosphate (18.7%) and potassium as potassium chloride (60%), according to the experiment scheme, were applied annually in early spring. Liming was carried out by lime application under pre-sowing cultivation during perennial grass sowing in a dose of 5 t/ha. Seeds of perennial legume grasses were inoculated with strains of nodule bacteria, according to the technological instructions, immediately before sowing. Dry weight content was determined by thermostatic weight method at 105 °C. The yield of green mass was accounted by weight method, by weighing with subsequent recalculation of yield from 1 ha of dry weight (Dospekhov, 1985).

Results and discussion

Analysis of the research results showed that in all research variants the best preserved grasses in five years were sown alfalfa and hornwort (Table 1). The average share of these plants when liming and simultaneous application of nitrogen-fixing and phosphate-mobilizing bacterial preparations in single-species crops was 80-84% and in mixtures with awnless bromegrass - 65-78%. The average proportion of eastern bromegrass was at 59% and 45%, respectively, which, unlike other legumes, was greatest in years 3-5 of use. The lowest share of the seeding crop was in agrocnoses with meadow clover (48 and 35%).
respectively), which remained at a high level only in the first three years of vegetation, later it almost completely fell out of the grass stand. In our studies, the accumulated symbiotic nitrogen ranging from 95 to 266 kg/ha against the background of liming and joint application of nitrogen-fixing and phosphate-mobilizing bacterial preparations, different species of perennial legume grasses in single and joint crops on average for 2016-2020 (see Table 1). Legume grass species both in single-species crops and in mixtures with awnless bromegrass accumulated more symbiotic nitrogen. Alfalfa in single-species crops accumulated the most nitrogen, while meadow clover in mixtures with awnless brome accumulated the least. All species of perennial leguminous grasses accumulated more of it in single-species crops than in mixtures with cereals. The productivity of single-species crops of perennial legumes and their mixtures with awnless bromegrass with yield of dry mass per 1 ha in the variant without liming and without application of nitrogen fixing preparations, due to the action of symbiotic nitrogen, was high and ranged from 5.12-8.38 tons on average within 2012-2020 (Table 1). This productivity was 1.7-2.5 times higher in comparison with the grass herbage formed from single-species awnless bromegrass crops.

**Table 1.** Productivity of perennial leguminous grasses and their mixtures with awnless bromegrass depending on soil liming and application of bacterial preparations, t/ha (average for 2016-2020rr).

| Grass species and sowing rate, kg/ha | Liming | Legume share, %* | Nc**, kg/ha | Bacterial preparation productivity | nitrogen-fixing | phosphate-mobilizing | nitrogen-fixing and phosphate-mobilizing |
|-------------------------------------|--------|------------------|-------------|-----------------------------------|----------------|---------------------|---------------------------------------|
| Meadow clover – 20                  | -      | 48               | 132         | 5.12                              | 5.24           | 5.22                | 5.30                                  |
|                                     | +      |                  |             | 5.23                              | 5.39           | 5.21                | 5.55                                  |
| Alfalfa – 20                        | -      | 84               | 266         | 8.38                              | 8.71           | 8.61                | 8.92                                  |
|                                     | +      |                  |             | 9.27                              | 9.73           | 9.36                | 9.91                                  |
| birdfoot deer vetch – 14            | -      | 89               | 187         | 6.95                              | 7.16           | 7.15                | 7.18                                  |
|                                     | +      |                  |             | 7.10                              | 7.27           | 7.24                | 7.43                                  |
| Eastern galega – 30                 | -      | 59               | 177         | 6.27                              | 6.56           | 6.33                | 6.70                                  |
|                                     | +      |                  |             | 6.92                              | 7.08           | 6.89                | 7.37                                  |
| Meadow clover – 10 +                | -      | 35               | 95          | 5.37                              | 5.53           | 5.40                | 5.57                                  |
| Anwless bromegrass – 15             | +      |                  |             | 5.42                              | 5.67           | 5.47                | 5.84                                  |
| Alfalfa – 10 + Anwless bromegrass – 15 | -      | 65              | 191         | 7.73                              | 8.13           | 7.98                | 8.18                                  |
|                                     | +      |                  |             | 8.61                              | 8.82           | 8.69                | 8.95                                  |
| birdfoot deer vetch – 7 +           | -      | 78               | 140         | 6.69                              | 7.02           | 6.85                | 7.02                                  |
| Anwless bromegrass – 15             | +      |                  |             | 7.05                              | 7.14           | 7.01                | 7.33                                  |
| Eastern galega – 15 +               | -      | 45               | 135         | 6.03                              | 6.16           | 6.06                | 6.32                                  |
| Anwless bromegrass – 15             | +      |                  |             | 6.54                              | 6.68           | 6.57                | 6.89                                  |
| Anwless bromegrass – 30 + N120      | -      |                  |             | 5.60                              | -              | -                   | -                                     |
| Anwless bromegrass – 30 (control)   | +      |                  |             | 2.81                              | -              | -                   | -                                     |

HIP05, t/hadry weight; standing grass = 0.36, bacterial preparation = 0.38, liming = 0.32.

* Share of legume component and accumulation of symbiotic nitrogen by leguminous grasses (Nc **) are given on the background of liming and joint application of nitrogen-fixing and phosphate-mobilizing bacterial preparations.

The most productive was the agrophytocenosis that was formed on the basis of single-species alfalfa crops, and the least productive was the agrophytocenosis that was formed on the basis of a mixture of meadow clover and awnless bromegrass. On average for five years legume-grass herbage in relation to equal-species legume crops, formed a total for three mowing by 3-9% less productivity from 1 hectare of dry weight. The grass stand formed from awnless bromegrass formed the lowest productivity of dry mass (2.81 t/ha) without fertilizers. After application of N120 the productivity of awnless bromegrass increased 2.2 times and was 5.60 t/ha of dry mass.

The research revealed a positive effect on the productivity of perennial legumes and their mixtures with awnless bromegrass by inoculating the seeds with nitrogen-fixing and phosphorus-mobilizing bacterial strains during sowing. Although the effect of bacterial preparations on the development of legume species weakened somewhat over the years, the productivity of perennial legume grasses and legume-straw grasses increased by an average of 2-6% from their action. Alfalfa responded better to the application of bacterial preparations. Its productivity on joint application of nitrogen-fixing and phosphate-mobilizing bacterial preparations was the highest of all the studied variants and reached 9.91 t/ha of dry weight. Somewhat greater productivity gains from the use of bacterial preparations were on the background of liming.
Studies also revealed a positive effect of liming the soil on the productivity of perennial legumes and their mixtures with awnless brome. Against the background of liming all legumes and legume-grasses grasses provided 3-11% higher productivity than the cenoses of these grasses without liming. Both in single-species and joint crops with awnless bromegrass responded better to lime treatment. Legume-grass grasses over the years after reducing the legume component in the grasses reacted worse to lime treatment. Legume-grass herbage grasses responded to lime treatment somewhat weaker than pure legume crops.

Productivity of legume and legume-grass stands changed significantly depending on the time of existence and use of grasses (Table 2). Liming had a positive effect on the productivity of agrocenoses in all years. In all years, single-species crops of perennial leguminous grasses were more productive than their joint crops with awnless bromegrass. The exceptions were agrocenoses that were formed with meadow clover, whose joint crops with awnless brome starting from the third year were more productive than grasses that were formed on the basis of single-species clover crops.

| Grass specie and sowing rate, kg/ha | Liming | Year |
|------------------------------------|--------|------|
| Meadow clover – 20                 | -      | 2016 | 2.32 |
|                                   | +      | 2016 | 2.47 |
| Alfalfa – 20                       | -      | 2016 | 2.14 |
|                                   | +      | 2016 | 2.25 |
| birdsfoot deer vetch – 14          | -      | 2016 | 2.54 |
|                                   | +      | 2016 | 2.66 |
| Eastern galega – 30                | -      | 2016 | 1.02 |
|                                   | +      | 2016 | 1.09 |
| Meadow clover – 10 + Anwless bromegrass – 15 | - | 2016 | 1.51 |
|                                   | +      | 2016 | 1.52 |
| Alfalfa – 10 + Anwless bromegrass – 15 | - | 2016 | 1.46 |
|                                   | +      | 2016 | 1.89 |
| birdsfoot deer vetch – 7 + Anwless bromegrass – 15 | - | 2016 | 1.66 |
|                                   | +      | 2016 | 1.92 |
| Eastern galega – 15 + Anwless bromegrass – 15 | - | 2016 | 0.91 |
|                                   | +      | 2016 | 1.21 |

HIP0.5, t/ha

| Year | 2016 | 2017 | 2018 | 2019 | 2020 |
|------|------|------|------|------|------|
| 0.21 | 0.39 | 0.43 | 0.41 | 0.37 | 0.33 |

* Productivity indicators are given against the background of joint application of nitrogen-fixing and phosphate-mobilizing bacterial preparations.

The lowest productivity was in 2016. In this year, grass crops for spring barley ranged from 0.9-2.66 t / ha of dry weight. The least responsive to the presence of the cover crop in the year of sowing were hornbeam and meadow clover, which provided the highest fodder productivity after mowing the cover crop, which was at 2.32-2.66 t / ha dry weight. In the year of sowing, the cover crop negatively affected the productivity of agrocenoses when adding awnless bromegrass to perennial legumes, which decreased in comparison with single-seeded crops. In 2017, the second year of crop life, the productivity of single-species crops of perennial legume grasses and their mixtures with cereals ranged from 6.41-10.30 t/ha of dry weight. This year, agrocenoses with meadow clover, hornwort, and alfalfa were the most productive, both in single-species crops and in mixtures with awnless bromegrass. The least productive were the agrocenoses which were formed with eastern goatweed. The highest level of productivity was provided by legumes and legume-grass agrocenoses in 2018. In the third year of their use, in the range of 7.92-13.43 t/ha of dry weight. This year in terms of productivity, the difference between different herbage stands with the participation of different types of legume grasses was the smallest. This is due to the fact that along with the good preservation in the herbage of alfalfa and hornwort, meadow clover was well preserved in the agrocenoses, and also powerfully grew and increased its share in the yield of goatweed. The best response to liming, as well as to the application of bacterial preparations this year was lucerne sowing, when its productivity under the influence of these factors increased by 1.1-1.2 times and reached a maximum value of 13.43 t/ha of dry weight at the combined application of lime, nitrogen-fixing and phosphate-mobilizing preparations.

In 2019, the fourth year of vegetation, the productivity of legume and legume-grass agrocenoses ranged from 4.00-12.63 t/ha dry weight. Both between single-species crops and joint crops of legumes and awnless bromegrass, the highest productivity was provided by agrocenoses with alfalfa. Productivity of agrocenoses formed on the basis of meadow clover, due to its loss from herbage was low and approached the productivity of cereal stoloniferous grass. Productivity of Oriental goatweed
approached to that of hornwort. Alfalfa responded best to lime treatment both in single-species crops and joint crops with awnless brome. Legume-grass herbage grasses in comparison with single-species legume crops formed by 5-7% lower productivity. The exception was meadow clover crops whose productivity, after its exclusion from the herbage was 1.8-2 times higher than in single-species crops.

A similar pattern was observed in 2020. This year compared with 2019, there was a tendency to decrease the productivity of these agroecosystems, excluding goat-grass and goat-grass-grass stands, the productivity of which did not change much. The greatest decrease in productivity occurred in phytocenoses, which were formed on the basis of straw-calf mixture. The productivity of swaths in the studied agroecosystems changed significantly. In all swaths high productivity was noted in agrophytocenoses which were formed on single-species alfalfa sowings. In all studied variants it was the highest in the first swath with parameters of 3.20-5.55 t/ha of dry weight, and the lowest in the third swath. In all swaths, the highest productivity was provided by joint application of nitrogen-fixing and phosphate-mobilizing bacterial preparations. During entire research period, we found that legume components in the legume-grass censuses, depending on soil and climatic factors in the Polissya and the northern part of the steppe accumulate aboveground mass of the symbiotic fixed nitrogen in the range from 50 to 200 kg/ha. Average weighted values of its accumulation by leguminous grasses are shown in Table 3. Alfalfa and sandy sainfoin accumulate it most in joint crops with cereal species. Single-species crops of perennial legume grasses accumulate 20-30% more symbiotic nitrogen than legume-grass agroecosystems.

Table 3. Approximate amount of symbiotic fixed nitrogen accumulated by legumes in meadow legume-grass phytocenoses, kg/ha

| Grass                                | Sowing stand grass | Natural stand grass |
|--------------------------------------|--------------------|---------------------|
| Alfalfa-grass                        | 150-170            | 70-100              |
| Meadow clover-grass                  | 130-150            |                     |
| White clover-grass                   | 110-130            |                     |
| Crowfoot-grass                       | 100-120            |                     |
| Holy clover-grass                    | 150-170            |                     |
| Legume grass with various legumes    |                    |                     |

Data are given for legume content of (50-60%). At legume content of 30-50% the symbiotic nitrogen is accumulated 1.2-1.5 times less, and at 60-80% - 1.2-1.5 times more. Data are given for optimal soil and climatic conditions for a particular type of legume component. Sown alfalfa, hornwort, and sandy sainfoin were used in the experiments.

To enhance the ecological role of grasslands, surface improvement measures of herbaceous ecosystems are applied in combination with protection measures, which are to preserve a wide range of biodiversity of natural and sown grass vegetation, to protect soils from erosion, and water sources from pollution and siltation. It should be borne in mind that as a conservation measure on natural forage lands, it is desirable to apply phytoreclamation, when instead of chemical ameliorants (lime, dolomite flour, and gypsum) which pollute the environment, perennial grasses resistant to increased acidity or soil salinity are used. Phytomeliorative ability on saline soils is characterized by melilot, which, in addition to high yields, removes toxic salts.

In organic production on forage lands it is advisable to apply organic fertilizers (manure, green manure, etc.) in doses of 10-30 t/ha in the amount of manure in light texture soils. The dose of application of litterless manure (semi-liquid, liquid and manure runoff) is calculated by the content of nitrogen or nutrient element (nitrogen, phosphorus or potassium), which is at its maximum (Kurgak, 2010).

Conclusions

The conceptual and technological basis of organic grassland management provides a set of measures for the production of environmentally safe forage for animals with the maximum use of biological factors of intensification and aimed at the conservation of the environment, in particular soils, water sources and biodiversity. Creation of sown grasses with an increased content of perennial leguminous grasses is one of the most promising areas of organic grassland farming. We have established that the productivity of monoeccious leguminous grasses and their mixtures with awnless brome was high and varied within the range of 5.12-8.38 tons during five years on the average per 1 ha of dry weight in the variant without fertilization, which is 1.7-2.5 times higher than the productivity of grass stand formed from monoeccious grasses with awnless brome grass. The highest productivity and the level of accumulation of symbiotic nitrogen (191-266 kg/ha) are provided by agroecosystems with lucerne sowing and the lowest one with meadow clover. Inoculation of perennial legume grass seeds with nitrogen-fixing and phosphate-mobilizing preparations increases the productivity of perennial legume grasses and their mixtures with cereals by 2-6%. The greatest effect is provided by joint application of these preparations. Soil liming before sowing perennial leguminous grasses and their mixtures with cereals increases their productivity by 3-11%. Alfalfa responds best to liming and the use of bacterial preparations.

References
Effect of wilting and silage additives on silage quality of legume and legume-ryegrass mixtures. Proceedings of the 26th General Meeting of the European Grassland Federation. Trondheim, Norway, 4-8 September, 2020, 121–125.

Parente, G., Stoltz, E., Kurhak, V.G., Kudrya, S.O. (2020). Influence of perennial legumes on the productivity of meadow phytocenosises. Ukrainian Journal of Ecology, 10(6), 310-315.

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