

БІОЛОГІЧНІ НАУКИ

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SOYBEAN SYMBIOTIC APPARATUS FORMED BY BRADYRHIZOBIUM JAPONICUM

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The analysis of the root system of soybean plants was carried out using presowing treatment with the inoculant Rizoaktiv R, and those on which nodule formation was observed with the *locally* abundant species of mutualistic nitrogen-fixing *rhizobial* bacteria. Positive effect of both the preparation with inoculation and the effect of the native rhizobia on the symbiotic apparatus of soybean plants was noted. The variant with presowing bacterization counted twice as many nodules as the control variant in the first year of studies and 1.5 times in the next year. High nitrogenase activity of soybean plants were noted. Accordingly to which the increase in the yield during the year 2017 was 24%.

Keywords: nodule, nitrogenase activity, nitrogen-fixing bacteria, legume-rhizobium symbioses, soybean.

Introduction. In our time, inoculation of plants with biological preparations is one of the irreplaceable technologies for growing legumes. The yield of legumes increases due to the presence of effective nodule bacteria on soybean roots and sufficient nitrogen nutrition. This becomes possible because of the ability of plants and bacteria to create legume-rhizobium symbiosis (LRS) – root nodules [1]. An important condition for the symbiotic system formation of legumes is the presence in the soil of free-living nodule bacteria. The formation of nitrogen-fixing nodules is possible due to the interaction of a certain species of the bacteria with the host plant.

Formulation of the problem. It is known that in the rhizosphere of the roots of leguminous plants is a huge number of the microorganisms, but only *Rhizobia* is able to form nitrogen-fixing nodules there. The bacteria *Bradyrhizobium japonicum* is strict soybean symbiont. The development of root system promotes the formation of an active symbiotic soybean apparatus, provides the plant with the main part of nitrogen and the effective reduction of this element from the atmosphere to ammonia. As the result of photosynthesis, in the presence of organic bonds, biological fixation of nitrogen is forming [2]. The effectiveness of LRS is characterized by the properties of nitrogen-fixing bacteria, such as: competitiveness, virulence, nitrogenase activity, complementarity with the host plant. It is important to use biological preparations based on nodule bacteria such as *Bradyrhizobium japonicum* to improve vegetative parameters and yield of soybean plants. Therefore, the aim of the work was to establish the influence of *B. japonicum* on the formation of the symbiotic apparatus of the soybean.

Materials and methods. Field research was conducted during 2016–2017 on the experimental field of the department of agroecology and biosafety of the Institute of Agroecology and Environmental Management of the National Academy of Agricultural Sciences of Ukraine (Khmilnitsky district, Vinnytsia region). Presowing treatment of soybean seeds (Moravia variety) was carried out

directly on the day of planting with the inoculant Rizoaktiv R. The experimental variants: 1. Without bacterization (control); 2. With bacterization (preparation Rizoaktiv R). Samples were selected in the budding phase of plants – when the largest nitrogenase activity is noted. Biometric indicators of plants were determined by conventional methods [3]. The above ground mass of plants, the mass of roots and the mass of the nodules was determined by the weight method. The field studies, observations, records and sampling were carried out in accordance with the methodology of carrying out field experiments on Dospehov [4].

The nodules nitrogenase activity was determined by acetylene-reductase method on Chrom-4 chromatograph [5]. The determination of the protein content was carried out on near infrared (NIR) grain analyzer Perten 8800 (Italy) intended for express analysis of seeds moisture, fat and protein quantity. Statistical processing of research results, calculation of standard deviation and the least significant difference (LSD) were performed using standard computer programs Statistica 10 and Excel 2016.

Results and discussion. The results of the study show that under field conditions with insufficient moisture, the variant with bacterialisation of soybean seeds with Rizoaktiv R preparation was characterized by an increase in practically all the studied vegetative parameters. Root length and root mass of soybean plants grew by 1.29 and 1.4 times compared to the variant without inoculant (Table 1).

The number of beans was 21.3 units / plant, it was in 1.26 times greater than in the control variant, but the weight of the beans decreased. It was argued that it arose because of low amount of precipitation during the vegetation period of the 2016 year. The below ground part of the plant developed better than the above ground part through the compensatory properties that presented in soybean variety. In addition, the data obtained confirm the resistance to drought. Literature reviews indicate that, with insufficient precipitation, the root system of plants continues to develop [6].

Table 1

Vegetative parameters of soybean plants

Variant	Root length, sm	Weight of root, g	Number of beans units/ plant	Weight of beans, g
2016				
Control (whithout bacterization)	16.5±0.6	4.53±0.7	16.9±1.2	3.45±0.3
Rizoaktiv R	20.3±1.1	6.35±0.8	21.3±0.6	8.84±0.4
2017				
Control (whithout bacterization)	14.25±0.8	5.7±0.7	18.2±0,6	8.21±0.4
Rizoaktiv R	18.45±1.2	7.3±1.1	25.1±1,5	9.16±0.7

Джерело: розроблено авторами

With a satisfactory amount of precipitation and humidification during 2017 year, the variant with presowing treatment of the inoculant Rizoaktiv R contained 1.37 times more beans, and their mass remained as in the control.

Therefore, in the both conditions, of insufficient amount of precipitation of the first year and with sufficient amount of precipitation in the following year, presowing treatment of soybean seeds with nodule bacteria *B. japonicum* contributed to a better development of the below ground part of plants.

The results of root system investigation in variants with inoculation of soybean plants with Rizoaktiv R preparation and without its application showed the formation of the active symbiotic apparatus of this culture. It is suggested that such structures were created by native nodule bacteria contained in the soil. It was considered that the investigated soybean variety was complementary to some aboriginal rhizobia strains, which contributed to the formation of an effective symbiotic system [7].

Note that the search and update not only soybean varieties, but also nitrogen-fixing bacteria – symbionts of the legumes, which can react to the root extract of the plants by the formation of functional nodules is an extremely important practice in our time. These studies reflect that the seed bacterization positively influenced the formation and development of the nodulation apparatus of soybean plants and promotes its active functioning regardless of the weather conditions.

Comparison of the parameters of the symbiotic apparatus of plants treated with a preparation based on nitrogen-fixing bacteria *B. japonicum* with plants without treatment, showed its active functioning in both variants of the experiment. The variant with presowing bacterization was characterized by twice as many nodules in the first year and 1.5 times during the second year of studies compared to the control variant (Fig. 1).

At the same time, we noted an increase of the number of nodules during year 2017 under conditions of a satisfactory amount of precipitation formed by aboriginal soybean bacteria, which is 2.5 times more than in the previous year 2016 with insufficient precipitation. This may indicate their rapid activation in the soil during the period of research, as in the literature reviews [8].

Observing the development of the nodulation apparatus of soybean under conditions of insufficient moisture, noted that in the variant with the using of the inoculum, its formation occurred on the basal roots, and in the variant with the local soil rhizobia, the nodules were located on the lateral roots, it was possible that the soil composition and moisture content did not give the opportunity to get on the central root [9].

Studies have shown that there was an increase in nitrogenase activity when using an inoculant during the growing season during the year with a low amount of precipitation by 24% compared to the control. Under conditions of satisfactory moistening the following year, a similar situation was observed with the functioning of the symbiotic apparatus of soybean by the established aboriginal soybean nodule bacteria – their activation was taking place (Fig. 2) [10].

In the variant with bacterization with Rizoaktiv R, which contains active strains of nodule bacteria, during the year with a satisfactory amount of precipitation nitrogenase activity of soybean nodules was 2.2 μmol of C_2H_4 per plant per hour, and in the variant without bacterization, 2.1 μmol of C_2H_4 per plant per hour, accordingly.

So nitrogenase activity of soybean symbiotic apparatus, formed by active strains of nitrogen-fixing soybean bacteria, only slightly exceeded that of the variant with ab-

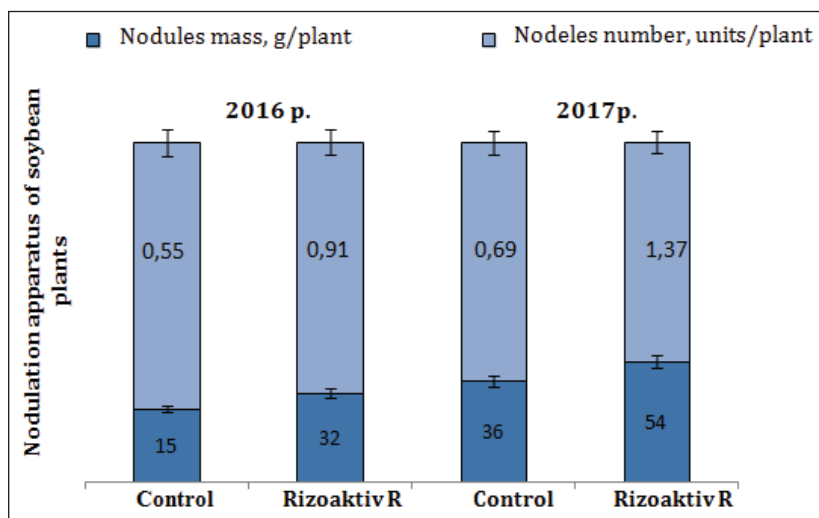


Fig. 1. Nodulation apparatus of soybean plants

Джерело: розроблено авторами

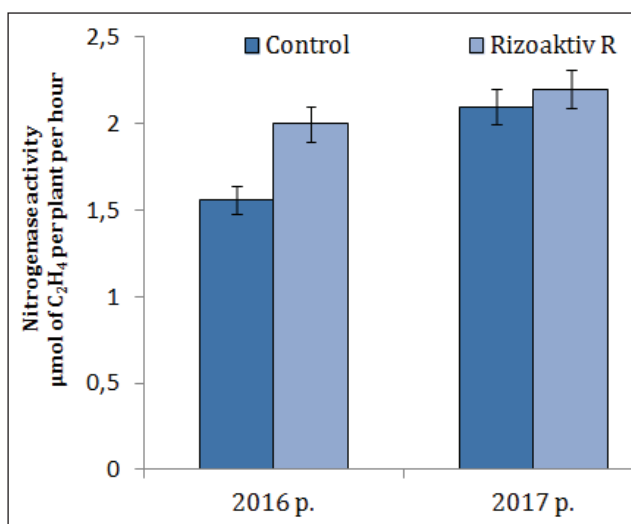


Fig. 2. Nitrogenase activity of soybean nodules

Джерело: розроблено авторами

original bacteria, it was assumed that in the soil there are competitive populations of rhizobia.

Thus, preparations of nitrogen-fixing bacteria not only form an active symbiotic apparatus and promote nitrogen fixation, but also positive influence on the yield and seeds quality (Table 2).

In the variant with presowing Rizoaktiv R bacterization the yield was 2.61 t/ha in the insufficient precipitation conditions. In the year with

Table 2

Yield and seeds quality		
Variant	Yield, t /ha	Protein content,%
2016		
Control (without bacterization)	1.86	36.9
Rizoaktiv R	2.61	38.9
LSD ₀₅	0.16	0.41
2017		
Control (without bacterization)	2.31	38.1
Rizoaktiv R	2.87	41.4
LSD ₀₅	0.11	0.56

Джерело: розроблено авторами

satisfactory moistening, the harvest of soybean in this experimental variant was at the level of 2.87 t/ha. Therefore, the yield increment was 24%.

Under conditions of insufficient moisture, protein content in soybean seeds increased by 5.4-8.7% in this two years.

Conclusion. Presowing inoculation positively influences the development of plants under different weather conditions. Consequently, the use of a highly active soybean inoculant Rizoactive R with *B. japonicum*, help to form the active nitrogen fixing apparatus of soybean *Glycine max* (L.) Merril. During the second year of the study, in the variant with usage of the microbiological preparation the yield increase from 0,56 to 0,75 tons per hectare.

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Анотація

Проведено аналіз кореневої системи рослин сої, при застосуванні передпосівної обробки інокулянтом Ризоактив Р, та тих, на яких спостерігали формування бульбочок спонтанними расами симбіотичних ґрунтових бактерій. Спостерігали позитивний вплив препарату та місцевих рас бульбочкових бактерій на симбіотичний апарат рослин сої. Варіант із передпосівною бактеризацією налічував у два рази більшу кількість бульбочок на відміну від контрольного варіанту першого року досліджень та в 1,5 рази – протягом наступного року. Відзначено високу нітрогеназну активність рослин сої. Завдяки цьому приривок врожаю протягом 2017 року досліджень становила 24%.

Ключові слова: бульбочки, нітрогеназна активність, азотфіксувальні бактерії, бобово-ризобіальний симбіоз, соя.

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Аннотация

Проведен анализ корневой системы растений сои, при использовании предпосевной обработки инокулянтом Ризоактив Р, и тех, на которых наблюдалось образование клубеньков спонтанными расами симбиотических почвенных бактерий. Отмечено положительное влияние как препарата так и местных рас клубеньковых бактерий на симбиотический аппарат растений сои. Вариант с предпосевной бактеризацией содержал в два раза большее количество клубеньков в отличие от контрольного варианта в первый год исследований, и в 1,5 раза – в течение следующего года. Отмечено высокие показатели нитрогеназной активности растений сои. Благодаря чему приривок урожая 2017 года исследований составляла 24%.

Ключевые слова: клубеньки, нитрогеназная активность, азотфиксирующие бактерии, бобово-ризобіальний симбіоз, соя.