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MICROBIOLOGICAL FERTILIZERS' IMPACT ON PHYSIOLOGICAL AND BIOCHEMICAL PROPERTIES OF SOYBEAN

D. AMONOVA¹, H. MATNIYAZOVA^{1,2,*}, G. ERGASHEVA³, G. SHAKHMUROVA³, and G. MAVLONOVA²

¹Institute of Genetics and Plants Experimental Biology, Academy of Sciences, Tashkent, Uzbekistan ²Department of Biology, Chirchik State Pedagogical University, Chirchik City, Uzbekistan ³Tashkent State Pedagogical University named after Nizomi, Tashkent, Uzbekistan Corresponding author's emails: abror0727@mail.ru, matniyazova@mail.ru Email addresses of co-authors: egulruxsor@mail.ru, shga2065@yandex.ru, gulnozamavlyanova21@gmail.com

SUMMARY

The effect of microbiological fertilizers on the leaf chlorophyll and carotenoid pigments and the seed protein and oil content in introduced and local cultivars of soybean (*Glycine max* L.) planted as a repeat crop was this research's focus for investigation. Based on the analysis, the microbiological preparation Rhizotorphin enhanced the leaf chlorophyll and carotenoids by 8.3%–16.7% and 1%–23.7%, respectively, as well as depending on the cultivar. The microbiological fertilizer Bioazot increased the leaf chlorophyll and carotenoids by 1%–18.7% and 1.04%–19.7%, respectively. The soybean grain's protein content also significantly rose with the influence of biofertilizers. However, the highest grain protein content was evident in the introduced cultivar Arleta (40.47%) and local cultivars Ehtiyoj (39.38%) and Xotira (38.02%) treated with biofertilizer Bioazot. A negative relationship emerged between the seed protein and oil content in analyzing the seed oil characteristics in soybean cultivars under the influence of microbiological preparations. The soybean cultivars observed with a higher protein content resulted from the influence of microbiological fertilizers but showed a reduced seed oil content.

Keywords: Soybean (*G. max* L.), cultivars, microbiological fertilizers, Rhizotorphin, Bioazot, chlorophyll, carotenoids, oil, protein, oil content

Key findings: Results revealed microbiological fertilizers had a significant positive effect on the physio-biochemical processes of soybean (*G. max* L.). The microbiological fertilizer Bioazot seed treatment had a considerable positive effect on the leaf chlorophyll and grain protein content of the cultivars Ehtiyoj, Xotira, and Arleta.

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INTRODUCTION

Nitrogen, considered the most essential macronutrient mineral in crop plants, had its deficiency as one of the critical limiting factors in the growth, development, and productivity of crop plants (Samudin and Kuswantoro, 2018). Nitrogen is an influential component of chlorophyll, organic bases, nucleic acids, nucleotides, some vitamins, and enzymes. Moreover, the lack of nitrogen considerably affects the physiological properties in leaf cells, the structure of chloroplasts, and the overall activities in crop plants (Beknazarov, 2009). Nitrogen is essential for protein synthesis; therefore, improving nitrogen availability to plants accounts for a higher assimilation rate for the specific element and a higher rate of protein synthesis within plant seed tissues (Sagdiyev and Alimova, 2007; Zarei et al., 2011).

Among the cultivated plants, soybean (Glycine max L.) has the highest demand for nitrogen. Fixation of biological nitrogen and nitrogen fertilizers are the main sources to meet the nitrogen demand of high-yielding soybean genotypes (Peric et al., 2009). The growing demand of modern agriculture with excessive synthesized mineral fertilizers causes several environmental issues related to the greenhouse effect, soil degradation, and air and water pollution. Therefore, as an effective solution for these problems and well-managed practices, biofertilizers agricultural the produced based on microorganisms have become an urgent proposal. The biofertilizers are not only natural, ecological, and economical but also preserve the soil structure and biological diversity of cultivated soils (Thomas and Singh, 2019).

In increasing the crop yield, chemical fertilizers' use for nitrogen has proven to reduce the formation of nodules and, eventually, nitrogen absorption of leguminous crop plants (Ohyama et al., 2017). Biofertilizers are extremely safe for humans, less polluting for animals and the environment, and cheaper than the high-costing mineral fertilization. Moreover, their application improves the soil biota and reduces the use of chemical fertilizers (Al-Erwy et al., 2016).

Several studies using bacterial fertilizers in crops revealed their effective influence, including enriching plants with nutrients, increasing plant resistance to biotic and abiotic and enhancing photosynthetic stresses, efficiency and productivity (Kumar and Kumar, 2020). The biofertilizers also improve the root system, vegetative growth, and nitrogen fixation, as well as stimulating the production of growth-promoting substances, such as vitamin B complex, auxin, gibberellins, and cytokinins. Additionally, these fertilizers help maintain soil fertility by secreting growthpromoting substances and vitamins (Baral and Adhikari, 2014).

Biological nitrogen fixation is the chief source of nitrogen required by soybean plants. Nitrogen-fixing bacteria application is most common in the cultivated area, which includes various microorganisms, such as Rhizobium, Bradyrhizobium, and Azotobacter. As it penetrates the biological cycle, atmospheric nitrogen ensures the production of additional protein on the surface of the earth. The plants that assimilate biological nitrogen contain more protein content, and it is ecologically clean, high quality, and provides better results in food animal feeding (Atabayeva and and Xudaykulov, 2018).

Rhizobium is а nitrogen-fixing symbiotic bacterium belonging to the family Rhizobiaceae, fixing around 50-100 kg of nitrogen per hectare. Proper use of *Rhizobium* strains has notably boosted the nodulation, uptake, and crop productivity nitrogen (Kaushal and Kukreja, 2020). Past studies enunciated that the treatment of sovbean with Rhizobium exhibited a significant effect on its vegetative growth, formation of nodules, yield components, and grain yield (Islam et al., 2021). Application of *Rhizobium* strains to common bean cultivars has proven to positively influence the leaf chlorophyll content and seed yield (Safikhani et al., 2013).

Azotobacter is a gram-negative, motile, aerobic, and free-living bacterium belonging to the family Azotobacteraceae. The *azotobacter* application as a biofertilizer showed positive effects on the seed germination, growth, and development of plants and increased the roots and shoots' length in crop plants. The said biofertilizer also enhanced the productivity of different crops under saline conditions isolated from other bacterial biomasses (Dar *et al.*, 2020). Moreover, *azotobacter* showed improved protein, nitrogen, potassium, phosphorus, and chlorophyll content compared with the control plants (Naseri *et al.*, 2013).

Past studies authenticated that the microbiological fertilizer Bioazot favorably affected physiological processes of soybean plants and increased their productivity (Matkarimov *et al.*, 2023). In addition, by applying this preparation to soybean seeds, the seed germination and germination power appeared significantly improved (Amonova *et al.*, 2023). In light of the above discussion, the presented research aimed to determine the effect of microbiological fertilizers on the leaf chlorophyll and carotenoid pigments and the seed protein and oil content in the introduced and local cultivars of soybean (*G. max* L.).

MATERIALS AND METHODS

This research progressed during 2022–2024 at the experimental field of the Faculty of Agronomy and Biotechnology, Bukhara State University, Bukhara, Uzbekistan. The three introduced cultivars, Arleta, Evrika-357, and Nena, and two local cultivars of soybean (*G. max* L.), i.e., Xotira and Ehtiyoj, and microbiological preparations Bioazot and Rhizotorphin were the main items of the research.

Seed inoculation

Before sowing, the two-hour treatment of soybean cultivar seeds with solutions of Rhizotorphin (ratio of 1:1.5) and Bioazot (ratio of 1:10) occurred. During the vegetation, soybean cultivars' nourishment with Bioazot continued twice through the leaves. All the cultivars' seeds reached sowing in wide rows with a row spacing of 60 cm and a depth of 4– 5 cm in the soil, in three replicates. The laboratory analysis proceeded at the Institute of Genetics and Plant Experimental Biology, Uzbekistan.

Extraction and determination of pigment concentration

The following experiment had the leaf taken from the third trifoliolate leaf in the upper layer of the soybean cultivar plants. The obtained leaf samples underwent grinding before being placed in 50-mg test tubes. Each leaf sample's homogenization in 5 ml of 95% ethyl alcohol solution continued for 15 minutes to settle down. Chlorophyll a and b and carotenoid contents' determination in the obtained liquid using the light absorption indicator at 664, 649, and 470 nm (Agilent Cary 60 UV-Vis Spectrophotometer of Germany). Based on the obtained results, calculating the pigments in the soybean cultivar leaves used the Lichtenthaler equation below (Lichtenthaler and Wellburn, 1983).

Ch-a =
$$13.36A_{664} - 5.19A_{649}$$

Ch-b = $27.43A_{649} - 8.12A_{664}$
= $(1000A_{470} - 2.13C_a - 97.63C_b)/209$

$$F[mg/g] = \frac{V * C}{P}$$

Seed protein content

C x+c

Total protein content determination employed the Kjeldahl method. Detecting the total protein content in the soybean seed samples relied on total nitrogen content (GOST, 2012).

Seed oil content

In soybean cultivars, the seed oil content analysis used a DW-SXT-02 DRAWELL Lab 500 ml Soxhlet extractor. The degreasing process proceeded in the Soxhlet, first in acetone and then in ethyl ether. After removing the degreased samples from the apparatus, their drying followed, then weighing the masses of flour and filter paper to know the difference between the mass before and after computing the degreasing. Based on this difference in mass, the determined amount of oil in the grain reached conversion as a percentage (Mirxamidova *et al.*, 2002).

Statistical analysis

The obtained data's analysis ran through Microsoft Excel 2016 by using the analysis of variance (ANOVA) in the STAT VIEW software.

RESULTS

Chlorophyll and carotenoid contents

By studying the different pigments in the leaves of soybean (G. max L.) cultivars during the flowering period, the highest chlorophyll a content resulted with the control option in leaves of soybean local cultivar Xotira (3.21 ± 0.07 mg/g), followed by an introduced cultivar, Nena $(3.11 \pm 0.07 \text{ mg/g})$. However, the lowest chlorophyll a content was apparently in the introduced cultivar Evrika-357 (2.84 ± 0.06 mg/g) belonging to the Kazakhstan selection (Table 1). During the flowering period of soybean cultivars, the topmost chlorophyll b content was notable within the control option in the local cultivar Ehtiyoj (1.44 ± 0.06) mg/g), followed by the introduced cultivar Nena $(1.35 \pm 0.02 \text{ mg/g})$. Meanwhile, the lowest chlorophyll b appeared in the introduced cultivar Evrika-357 (1.29 \pm 0.04 mg/g). Regarding carotenoids' content in the soybean plant leaves, the optimum content was prominent with the control option in local cultivar Xotira (0.79 \pm 0.02 mg/g), followed by the soybean introduced cultivar Nena (0.77 \pm 0.01 mg/g). However, the lowest level of carotenoids surfaced in the introduced cultivar Evrika-357 ($0.68 \pm 0.02 \text{ mg/g}$).

The results also revealed that the chlorophylls a and b and carotenoids in the leaves of all the soybean experimental cultivars enhanced with different degrees under the influence of the microbiological preparation Rhizotorphin compared with the control option. During the soybean flowering period, the highest chlorophyll a pigment was evident in the local cultivar Ehtiyoj $(3.62 \pm 0.07 \text{ mg/g})$, followed by the introduced cultivar Nena (3.34 \pm 0.03 mg/g) treated with the microbiological preparation Rhizotorphin. However, as influenced by this preparation, the lowest content of chlorophyll a resulted in the introduced cultivar Arleta $(3.25 \pm 0.07 \text{ mg/g})$. One should note that the microbiological preparation Rhizotorphin had a considerable effect on the chlorophyll a content in the leaves of soybean local versus the introduced cultivars.

By treating soybean cultivars with the microbiological preparation Rhizotorphin during the flowering period, the recorded premier chlorophyll b pigment was in the local cultivar Ehtiyoj $(1.62 \pm 0.07 \text{ mg/g})$, followed by the exotic cultivar Nena $(1.49 \pm 0.04 \text{ mg/g})$. Although the lowest chlorophyll b pigment appeared in the introduced cultivar Evrika-357 $(1.42 \pm 0.01 \text{ mg/g})$. Under the influence of the biopreparation Rhizotorphin, the highest carotenoid content prevailed in the cultivars Ehtiyoj, Xotira, and Evrika-357 (0.94 ± 0.02 , 0.93 ± 0.02 , and $0.81 \pm 0.03 \text{ mg/g}$, respectively). According to the results, the carotenoids with the lowest content were existent in the soybean cultivar Arleta (0.76 \pm 0.03 mg/g) (Table 2).

In the study of the effect of the microbiological preparation Bioazot on the chlorophyll a content in the leaves of soybean cultivars during the flowering period, the highest chlorophyll a content was evident in the local cultivar Ehtiyoj $(3.66 \pm 0.10 \text{ mg/g})$. Second to it was the soybean introduced cultivar Arleta $(3.46 \pm 0.04 \text{ mg/g})$. However, the lowest indicator under the influence of the preparation Bioazot was noticeable in the introduced cultivar Evrika-357 (2.84 \pm 0.06 mg/g) belonging to the Kazakhstan selection. With preparation Bioazot, the uppermost chlorophyll b pigment was in the leaves of soybean local cultivar Ehtiyoj (1.67 ± 0.05 mq/q), followed by the soybean foreign cultivar Arleta (1.55 \pm 0.03 mg/g). The lowest level of chlorophyll b in leaves manifested in the exotic cultivar Evrika-357 $(1.33 \pm 0.01 \text{ mg/g})$ with the preparation Bioazot. Moreover, with Bioazot, the maximum carotenoid content resulted in the local cultivar Xotira (0.93 \pm 0.01 mg/g), followed by the exotic cultivar Nena $(0.91 \pm 0.03 \text{ mg/g})$. The minimum carotenoid index was notably in the foreign cultivar Evrika-357 ($0.71 \pm 0.01 \text{ mg/g}$).

No.	Soybean cultivars	Control		Rhizotorphin		Bioazot	
		chlorophyll	chlorophyll	chlorophyll a	chlorophyll b	chlorophyll a	chlorophyll
		a (mg/g)	b (mg/g)	(mg/g)	(mg/g)	(mg/g)	b (mg/g)
Intro	Introduced cultivars						
1.	Arleta	2.90±0.07	1.34±0.02	3.25±0.07	1.47±0.03	3.46±0.04	1.55±0.03
2.	Evrika-357	2.84±0.06	1.29 ± 0.04	3.29±0.09	1.42 ± 0.01	2.84±0.06	1.33 ± 0.01
3.	Nena	3.11±0.07	1.35 ± 0.02	3.34±0.03	1.49±0.04	3.32±0.09	1.46 ± 0.05
Local cultivars							
4.	Ehtiyoj	3.05±0.02	1.44±0.06	3.62±0.07	1.62±0.07	3.66±0.10	1.67±0.05
5.	Xotira	3.21±0.07	1.40 ± 0.06	3.52±0.02	1.53 ± 0.05	3.54 ± 0.04	1.53 ± 0.02

Table 1. Influence of microbiological preparations on the leaf's chlorophyll a and b in the soybean at the flowering stage.

Table 2. Influence of microbiological preparations on the leaf's total chlorophyll and carotenoid contents in the soybean at the flowering stage.

	Soybean cultivars	Control		Rhizotorphin		Bioazot	
No.		Total chlorophyll (mg/g)	Carotenoids (mg/g)	Total chlorophyll (mg/g)	Carotenoids (mg/g)	Total chlorophyll (mg/g)	Carotenoids (mg/g)
Introduced cultivars							
1.	Arleta	4.24±0.08	0.70±0.02	4.72±0.09	0.76±0.03	5.00±0.07	0.80±0.03
2.	Evrika-357	4.13±0.10	0.68±0.02	4.71±0.16	0.81±0.03	4.17±0.06	0.71±0.01
3.	Nena	4.46±0.09	0.77±0.01	4.83±0.02	0.77±0.01	4.79±0.14	0.91±0.03
Local cultivars							
4.	Ehtiyoj	4.49±0.17	0.76±0.03	5.24±0.14	0.94±0.02	5.33±0.14	0.91±0.01
5.	Xotira	4.65±0.16	0.79±0.02	5.05±0.07	0.93±0.02	5.07±0.04	0.93 ± 0.01

Seed protein content

Studying the soybean grain's protein content also succeeded in this experiment. The highest protein content (32.73%) was prominent in the local cultivar Ehtiyoj, followed by the introduced cultivar Evrika-357 (34.52%) with control options (Figure 1). However, the lowest grain protein content resulted in the introduced cultivar from the Russian selection, Arleta (30.43%).

By treating the soybean cultivars with microbiological fertilizer Rhizotorphin, it was apparent that the said preparation enhanced the grain protein content in all the soybean cultivars, except Evrika-357, depending on the cultivars with different degrees compared with the control options. With the influence of the preparation Rhizotorphin, the recorded highest seed protein content materialized in the local cultivar Ehtiyoj local (37.03%), followed by the introduced cultivar from the Russian selection, Arleta (33.78%). Meanwhile, the lowest protein content appeared in the exotic cultivar Evrika-357 (32.76%).

On the effect of microbiological fertilizer Bioazot on grain protein content in soybean cultivars, the said biofertilizer significantly increased the grain protein in the soybean cultivars compared with the control. Under the influence of the preparation Bioazot, the utmost value of grain protein content occurred in the introduced cultivar Arleta and the local cultivar Ehtiyoj (40.47% and 39.38%, respectively). However, in the Evrika-357 cultivar, the seed protein content showed a slight reduction under the influence of this biofertilizer compared with the control option.

According to the outcomes, it was evident that the microbiological preparation Bioazot had a significant positive effect on the grain protein content versus the Rhizotorphin microbiological preparation in soybean cultivars.

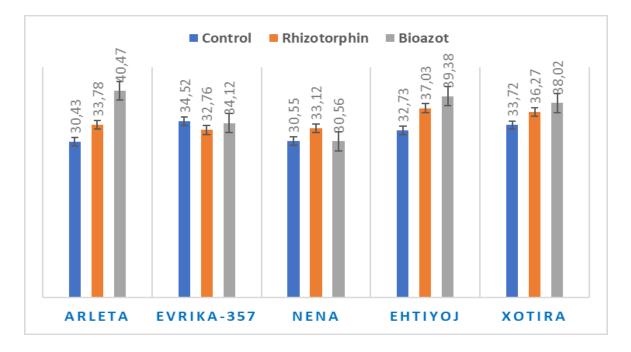


Figure 1. Influence of microbiological preparations on the seed protein content in soybeans.

Seed oil content

The scrutiny of soybean's seed oil content under the influence of microbiological fertilizers continued in different soybean cultivars. The findings revealed that biofertilizers and soybean cultivars have varied effects on the seed oil content. By comparing the soybean cultivars, the highest seed oil resulted in the local cultivar Xotira (22.46%), while the lowest value for the said trait was apparent in the exotic cultivar Evrika-357 (18.41%) (Figure 2).

Regarding the effect of the microbiological preparation Rhizotorphin on soybean's seed oil content, the results revealed varied effects on the oil content, depending on the soybean cultivars. By treating with the microbiological preparation Rhizotorphin, the introduced cultivar Arleta did not differ significantly from the control option for seed oil content. Noticeably, the exotic cultivar Evrika-357 treated with the preparation Rhizotorphin showed an increased grain oil content by 17.7% compared with the control option. However, in other cultivars, the seed oil content was slightly lower under the influence

of the preparation Rhizotorphin than the control options.

Concerning the study of the soybean's seed oil content under the microbiological fertilizer Bioazot's influence, the outcomes displayed different effects of the said preparation, depending on the soybean cultivars. It was also remarkable that the seed oil does not differ significantly in the cultivar Arleta by treating it with the fertilizer Bioazot and the control option. However, in the soybean's introduced cultivar Evrika-357's treatment with the microbiological fertilizer Bioazot, the grain oil content expressed a considerable enhancement compared with the control options. Although in soybean local cultivars Xotira and Ehtiyoj, the seed oil content was slightly lower in the fertilizer than the Bioazot in control options. Furthermore, results revealed the soybean cultivars with the high-protein content showed a low seed oil content. The increased seed protein content could be due to the sufficient nitrogen supply to the soybean plants under the influence of nitrogen-fixing preparations.

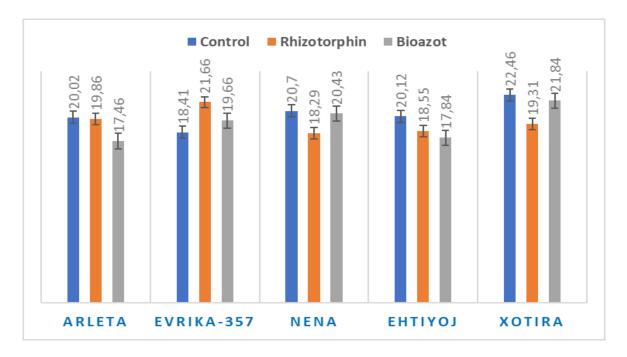


Figure 2. Influence of microbiological preparations on the seed oil content in soybeans.

DISCUSSION

The promising research showed the influence of microbiological fertilizers on leaf pigments and the grain's protein and oil contents in the local and foreign cultivars of soybean at the flowering phase. Soybean cultivars treated with microbiological fertilizers exhibited improved growth and acceleration in physiological processes by better absorption of nutrients. Photosynthesis plays a vital role in the high productivity of crop plants. The high and low photosynthesis indicated associations with the main components of the chloroplast, which directly determine the photosynthetic potential (Kulmamatova et al., 2022). One of the chief components of the chloroplast is the chlorophyll pigment, considered as the most essential green pigment in the photosynthesis process, and its concentration is an indicator of plant health (Porra, 2002). The results revealed the elevated consumption of biological nitrogen by soybean plants leads to boosting the chlorophyll content in leaves, which, in turn, causes the flow of photosynthesis products to the nodes and the activation of atmospheric nitrogen fixation (Tsvetkova et al., 2020).

In this research, the leaf pigments during the flowering phase of soybean cultivars the influence of under microbiological preparations underwent scrutiny. According to results, all the biopreparations showed the highest content of chlorophyll a and b and carotenoids in the leaves of soybean cultivars compared with the control options. It was evident that the preparation Rhizotorphin increased the leaf chlorophyll content by 8.3%-16.7%, and the carotenoids by 1%-23.7%, depending on the soybean cultivars. The results further detailed that under the influence of biofertilizer Bioazot, the chlorophyll content rose by 1%-18.7% and the carotenoid pigments augmented by 1.04%-19.7%. Biofertilizers can also reduce the chlorophyll loss and improve the chlorophyll synthesis by stimulating production of pyridoxal the enzymes. These enzymes play an important role in the production of a-aminolevulinic acid, the primary compound required by chlorophyll synthesis (Yaghoubian et al., 2021).

Better vegetative growth of plants inoculated with *Rhizobium* strains can stimulate the total chlorophyll content in leaves due to greater absorption of nitrogen by crop plants (Chaudhary *et al.*, 2022). Several studies revealed that Rhizobium strains positively affected the leaf chlorophyll content and the seed protein percentage in Phaseolus vulgaris L. (Safikhani et al., 2013). Past studies stated the highest rate of nitrogen absorption appeared between the phases of flowering and pod forming in soybean plants (Tsvetkova et al., 2020). Al-Erwy et al. (2016), as established by their studies, reported that the rise in contents of chlorophyll a and b and carotenoid in wheat was up to 30.5%, 18.5%, and 44%, respectively, when treated with Rhizobium compared with the control option. However, by treating with Azotobacter, the chlorophyll a, chlorophyll b, and carotenoid contents enhanced by 38.3%, 32.4%, and 51.5%, respectively, in wheat (Al-Erwy, 2016).

Nitrogen is the most crucial macroelement in protein synthesis, and its increase in the optimal conditions can also enhance the protein content (Safikhani et al., 2013). The inoculation of chickpea seeds with Rhizobium strains has proven to have a positive influence on the yield characteristics, grain yield, and grain protein content (Ahmed et al., 2007). In the presented study, it was a discovery that microbiological fertilizers had a significant positive influence on the protein content in soybean grains. This could refer to the increased flow of nitrogen to plants due to the acceleration of the nitrogen-fixation process under the control of microbiological preparations in soybean plants. Notably, under the effect of the Rhizotorphin preparation, the grain protein content increased from 32.39% to 34.59%, and up to 2.2%, while under the stimulus of the preparation Bioazot, the grain protein increased from 32.39% to 36.51%, and up to 4.12%.

Mechanisms of plant growth-promoting rhizobacteria, such producing as phytohormones, support plant growth, promote nutrient uptake, nitrogen fixation, and improve the availability of key nutrients to the crop plants, as well as produce enzymes like riboflavin and thiamine (Wu et al., 2005). Notably also that by using rhizobacteria, the seed protein increased while working as nitrogen fixers, with the nitrogen directly used in the protein synthesis. The rhizobacteria increase the production of phytohormones,

such as auxin, gibberellins, and cytokinins, as closely related to nitrogen signaling, expressing the connection of nitrogen and phytohormone signals to adapt the crop plants' morphology and physiology. Phytohormones are the core components of protein modifications and improve crop productivity and quality of seed oil in crop plants (Nosheen *et al.*, 2016).

Along with the protein content, another nutritionally important quantitative characteristic of soybean seeds is the grain oil content (Boczar, 2016; Szostak et al., 2019). Soybeans account for 59% of the world's oil plants and 29% of the total consumed vegetable oil. Nutritional values of soybean oil and fatty acids are responsible for the stability and taste of this product. Soybean oil contains an average of 13% palmitic acid, 4% stearic acid, 20% oleic acid, 55% linoleic acid, and 8% linolenic acid. Soybean oil has a lower oxidation stability than other vegetable oils (Suciu et al., 2022).

By studying the grain oil content in soybean cultivars under the influence of microbiological preparations, it was evident biofertilizers had varied effects on the oil content. An observation revealed that by having the high-protein content in seeds, the soybean cultivars Arleta and Ehtiyoj showed the lowest oil content under the influence of biofertilizers. The soybean cultivar Evrika-357 displayed an increased grain oil content under effects biofertilizer preparations the of compared with the control options. A negative relationship between the grain protein and oil content occurred, and the grain protein content demonstrated the greater variability than oil content in soybeans (Peric et al., 2009; Matniyazova et al., 2024).

On using nitrogen-fixing biofertilizers, the amount of nitrogen increased in crop plants, boosting the grain protein content, leading to a decreased seed oil content. With increased nitrogen consumption in plants, the nitrogen-containing formation of protein and protein precursors rises formation accelerates in the provision of photosynthetic materials, and as a result, the materials required for conversion to seed oil decrease

(Ahmad and Ali, 2018; Taherkhani and Golchin, 2006).

CONCLUSIONS

The soybean (*G. max* L.) seeds' sowing with microbiological fertilizers had a positive effect on the leaf pigments and the seed protein due to better absorption of nutrients by the plants. The increase in the leaf chlorophyll pigments appeared inherently connected with the rise in the seed protein. The negative relationship existed between the seed protein and oil content, and the soybean cultivars with the highest grain protein had the lowest seed oil.

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