

The influence of effective microorganisms on the yield and quality of individual seed components of different soybean genotypes

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A B S T R A C T

Soybean as a member of the legume family has a high economic value, thanks to the high protein and oil content of the seed. In addition, its importance is evidenced by its breeding potential and symbiosis with nodulating bacteria, which facilitates production in increasingly extreme climatic conditions and degraded soil conditions. Considering the need to search for optimal solutions in the production of this legume, in order to achieve high yields and grain quality, research was conducted to determine the effect of applying a microbiological preparation containing effective microorganisms on the yield, and protein and oil content of soybeans. Research was conducted in the period 2016–2018 (factor A), on 6 different soybean genotypes belonging to different ripening groups: 0 (Galina and Valjevka), I (Sava and NS Apolo) and II (Rubin and NS Zita) (factor B), and under fertilization treatments: F0 – control, F1 – NPK fertilizer, and F2 – NPK + microbiological preparation (incorporated into the soil and two foliar treatments) (factor C). It was shown that the application of NPK fertilization treatment in combination with effective microorganisms was the most beneficial for all investigated parameters, with a high statistical significance. This treatment increased yield by 15.67%, protein content by 0.34% and oil content by 0.47% compared to the control. The additional importance of this research is the simultaneous positive impact on protein and oil contents in soybeans, given that they are generally negatively correlated.

Keywords: soybean, effective microorganisms, yield, proteins, oils

ИЗВОД

Соја као члан породице махунарки има високу економску вредност, захваљујући високом садржају протеина и садржају уља у семену. Осим тога, њен значај се огледа у могућности оплемењивања сорти и симбиози са квржичним бактеријама, те је олакшана производња у све екстремнијим климатским условима и деградираним земљишним условима. С обзиром на потребу тражења оптималних решења у производњи ове махунарке, у циљу постизања високих приноса и квалитета зрна, спроведена су истраживања како би се утврдио ефекат примене микробиолошког препарата са ефективним микроорганизмима на принос и садржај протеина и уља у зрну соје. Истраживања су спроведена у периоду 2016–2018 (фактор А), на 6 различитих генотипова соје различитих група зрења: 0 (Галина и Ваљевка), I (Сава и НС Аполо) и II (Рубин и НС Зита) (фактор Б) и третманима ђубрења: ФО – контрола, Ф1 – НПК ђубриво и Ф2 – НПК + микробиолошки препарат (инкорпориран у земљиште и два фолијарна третмана) (фактор Ц). Показало се да је примена третмана НПК ђубрења у комбинацији са ефективним микроорганизмима била најкориснија за све испитиване параметре, са високом статистичком значајношћу. Овај третман је утицао на повећање приноса за 15,67%, садржаја протеина за 0,34% и садржаја уља за 0,47% у односу на контролу. Додатни значај овог истраживања је истовремени позитиван утицај на садржај протеина и уља у зрну соје, с обзиром на то да су они генерално у негативној корелацији.

Кључне речи: соја, ефективни микроорганизми, принос, протеини, уља

1. Introduction

The tendency for ecological, sustainable and economical agricultural production (Santos & Ribeiro 2019) implied the creation of different strategies and the implementation of innovations in production technologies, with the aim of reducing intensive agricultural practices (Faria et al., 2023). Innovations refer to the use and evaluation of various inputs in order to increase production, and are the main factor for the creation and development of technologies and their impact on various production factors (Giovanini et al., 2019).

An additional important parameter on which progress in crop development depends is climatic conditions (He and Matthews, 2023), whose oscillations act differently during the development phases of plant culture (Wu et al., 2023). Đukić et al. (2018) observed abnormal weather changes, which are manifested in increased average daily temperatures and different amounts of precipitation in certain years, which implies the alternation of rainy and extremely dry years. This adversely affects the quantity and quality of crop yields, especially the occurrence of drought (Bajagić et al., 2021). On the other hand, Đukić et al. (2018a) explained that if the amount of precipitation is significantly higher in the first vegetative stages, it leads to the vigorous development of the plant's habitus and the development of the root system in the shallow part of the soil, which causes adverse effects in the later stages of development if precipitation is absent.

The development of modern civilization and the global increase in demand for food have influenced soybean (Glycine max L. Merr.) to become one of the most economically important crops in the world and the main crop for increasing the area and grain production (Faria et al., 2023). Although it is not a basic food in the human diet, it is an essential source of vegetable oil and protein (Bazzo et al., 2021; Silva et al., 2022). The composition of soybeans is 45% protein, 20-22% oil, 20-26% carbohydrates, 5% minerals (phosphorus, potassium, calcium, sulfur, magnesium, etc.) and many vitamins (mainly A and B) (Yalçin, 2018). Soybean proteins contain all twenty types of amino acids, including nine essential amino acids (Hu et al., 2023), whose nutritional value is equivalent to that of animal proteins, and are important in vegetarian diets (Zhang et al., 2017). Soybean oils are beneficial in many ways for a diet lacking in omega-3 and omega-6 fatty acids. Soybean oil is characterized by a good ratio of fatty acids, including unsaturated fatty acids: linoleic, oleic and linolenic acids, and saturated fatty acids: palmitic and stearic acids (Wang et al., 2019).

The fact that intensive agricultural production has a negative impact on sustainable systems and safe food production increases the interest in new technologies, such as the application of biofertilizers (Gawęda et al., 2018). Fertilization as a cultural practice provides a basic strategy for sustainable production systems with the aim of obtaining high and stable yields (Bazoo et al., 2021; Faria et al., 2023). Certain technologies related to fertilization aim to bioactivate life in the soil, which accelerates the decomposition of organic matter, increases and balances microbiological activities in the soil, initiates the metabolism of soil biomass and facilitates the interaction between plants and beneficial microorganisms (Cobucci et al., 2015).

Microbial biomass in the soil is defined as a living component, which participates in decomposition processes, nutrient cycling and energy flow. Microorganisms are the main source of food for plants. Their activity results in the release of enzymes and other compounds that affect soil pH and create nutrients available to plants. Any change in their number and activity interferes with biochemical processes in the soil, which indirectly affects the growth and development of plants (Agostinho et al., 2017; Cvijanović et al., 2020; Cvijanović et al., 2022a). A commercially available solution of natural microbes in the soil, i.e., effective microorganisms (EM), is used in order to increase their abundance, activity and impact on the availability and absorption of nutrients needed by plants.

According to many studies, the use of effective microorganisms has exceptional advantages. The benefits include the release of nutrients from organic matter when composted with EM (Olle and Williams, 2013), greater resistance of soil and plants to water stress, higher rates of carbon mineralization, improved soil properties and better root penetration after EM application, the suppression of pests and diseases (Chen et al., 2022), stimulation of plant growth, elimination of the effects of biotic and abiotic stress, yield increases and crop quality improvement (Persson et al., 2003; Cvijanović et al., 2022). Also, they gradually eliminate the consequences of soil degradation and bioremediation, thus serving as a permanent solution for the creation of a "healthy soil" necessary for organic plant production (Cvijanović et al., 2012).

Considering the need to find optimal solutions in sustainable agricultural production, the aim of the research was to determine the effect of different fertilizing treatments with NPK fertilizer and a microbiological preparation containing effective microorganisms on the yield and quality of certain components of soybean seed.

2. Materials and methods

2.1. Design of experimental research

Experimental research was carried out in the period 2016-2018 at the Institute of Crop and Vegetable Farming in Rimski Šančevi. The experiment in dry farming was set up as a three-factorial split-plot design, in four replicates. Plot size was 15 m², with six rows of soybeans with an inter-row spacing of 50 cm and a row length of 5 m. The experiment had a 3-mwide protective ecological belt of sown soybeans. Also, within each plot, the marginal rows additionally served as isolation, while the inner 4 rows were used for the analyses. The first crop was corn. Across all three years of the research, standard cultural practices were used, i.e., autumn tillage and application of NPK fertilizers, closing furrows, seedbed preparation, inoculation of soybean seeds with the NS Nitragin microbiological preparation containing highly active strains of the symbiotic nitrogen fixer Bradyrhizobium japonicum, machine sowing, and crop care.

Factor A: Meteorological conditions (temperature and precipitation), considering that soybean production depends on climatic factors, and that there is no irrigation system.

Factor B: Soybean varieties of different ripening groups: 0 (Galina and Valjevka), I (Sava and NS Apolo) and II (Rubin, NS Zita).

Factor C: Fertilization, which was applied in the following treatment variants:

F0 – control

F1 – NPK mineral fertilizer formulation 8:15:15 + 3% Ca + 9% S in the amount of 400 kg ha⁻¹

F2 – NPK mineral fertilizer formulation 8:15:15 + 3% Ca + 9% S in the amount of 400 kg ha⁻¹ + EM Aktiv microbiological preparation (incorporation before sowing in the amount of 20 l ha⁻¹ in the ratio to water of 1:10 and two foliar treatments in the three to four trefoil stage and budding stage of plant development , in the amount of 6 l ha⁻¹ in the ratio to water of 1.5:100). The EM active preparation is a liquid concentrate, certified for organic production in Serbia and the EU (http://www.minpolj.gov.rs/organska/), in which more than 80 strains of the main aerobic and anaerobic organisms found in nature have been grown in the soil.

Soybeans were harvested at the stage of harvest maturity, with the help of a Wintersteiger elite combine harvester, with a small amount of work involved. Soybean biomass and grain moisture were measured and yield (kg/ha) was calculated at 14% moisture. Grain protein and oil contents (%) were measured by a Perten NIR spectrophotometer at the Soybean Department (Balešević-Tubić et al., 2007).

2.2. Meteorological conditions for plant growth

Mean monthly air temperatures during the vegetative period of soybeans were a uniform 19.2°C to 20.6°C, but higher than the long-term average of 18.1°C. The amount of precipitation was different and deviated from the long-term average. Precipitation was higher in 2016 (450.6 l m⁻²) and 2018 (435.9 l m⁻²) than the long-term average. In 2017, the sum of precipitation was 316.5 l m⁻², which was 58.5 l m⁻² less than the long-term average (375.0 l m⁻²) (Table 1). In order to obtain high yields, temperatures and precipitation in the years of production should be in accordance with crop requirements.

Table 1.

Average monthly temperatures (°C) and total precipitation (mm) for the soybean growing season 2016–2018

| Year | Mean monthly air temperatures | | | | | | Average | Long-term average |
|-------------------------------|-------------------------------|------|-------|------|------|------|-----------|-------------------|
| | IV | V | VI | VII | VIII | IX | 2016-2018 | 1964-2015 |
| 2016 | 14.2 | 16.9 | 21.7 | 22.8 | 21.1 | 18.5 | 19.2 | |
| 2017 | 11.4 | 17.6 | 23.2 | 24.3 | 24.8 | 16.9 | 19.7 | 18.1 |
| 2018 | 17.2 | 20.4 | 21.5 | 22.0 | 24.0 | 18.5 | 20.6 | |
| Sums of monthly precipitation | | | | | | | Sum | Long-term sum |
| 2016 | 74.5 | 85.0 | 143.2 | 68.4 | 45.8 | 33.7 | 450.6 | |
| 2017 | 57.0 | 82.9 | 65.7 | 12.0 | 17.4 | 81.5 | 316.5 | 375.0 |
| 2018 | 49.0 | 64.2 | 163.2 | 81.2 | 51.2 | 27.1 | 435.9 | |

Source: http://www.hidmet.gov.rs/ciril/meteorologija/klimatologija_godisnjaci.php

2.3. Statistical analysis

The results of the research were processed using descriptive statistics and the analysis of variance of the three-factorial experiment in the DSAASTAT program (Perugia, Italy). A three-way ANOVA was used to test for the effects of year, different genotypes of soybean and fertilization treatments. Correlations were determined between the traits tested. The significance of the differences was tested with the LSD test at the P < 0.01 and P < 0.05 significance levels. The results are presented in tabular form.

3. Results and discussions

Grain yield, as well as the chemical composition of soybeans, depends on climatic conditions, production technology and the measures used in production. In production years when the absence of precipitation is accompanied by high temperatures, drought occurs, which adversely affects the yield. According to Bajagić et al. (2021), about 300-350 mm of precipitation is needed for the production of high-yielding soybeans, of which about 80% is needed in the reproductive stages, from the end of June to September. Also, a yield reduction of up to 30% can be caused by abiotic factors (Staniak et al., 2023). The impact of drought has increased in recent decades, given the trend of increasing temperatures from year to year, which reduces crop productivity (Cunha et al., 2019; Petrović et al., 2020; Silva and Arima, 2023). For the research period 2016-2021, Đukić et al. (2021) reported an impact unfavorable on the morphological characteristics of several varieties in 2017 and 2021, due to the deficit of precipitation in the reproductive stages of soybean development, accompanied by extremely high temperatures. Also, the obtained results are correlated with MacCarthy et al. (2022), who cited

high variations in climate conditions with increased CO_2 levels, which negatively affected both soybean yields and other crops.

The analysis of variance for all examined parameters based on the LSD test showed that the main sources of variation, as well as their interactions, had a very significant impact on grain yield, and the obtained differences were statistically significant (P < 0.01). The total average soybean yield was 4,046.39 kg ha⁻¹ (Table 2). The average soybean yield was higher in 2016 by 58.86% (4,752.72 kg ha⁻¹), and in 2018 by 50.16% (4,492.34 kg ha⁻¹) compared to 2017 (2,991.70 kg ha⁻¹). The differences in values are attributed to the amount of precipitation, which in 2017, in June, July and August, was extremely lower than soybean requirements, and was accompanied by extremely high temperatures.

Averaged across varieties, the highest yield was achieved with the Rubin variety (4,354.26 kg ha⁻¹), and the lowest with the Galina variety (3,714.47 kg ha⁻¹). When observed by year, on average, higher yields were in mid-late genotypes, such as Rubin and NS Zita. Miladinov et al. (2017) explained that safer production and achieving high and stable yields can be solved by growing varieties of different ripening groups, because they go through critical periods of growth and development differently. Đukić et al. (2018) stated that priority should be given to new varieties, which were created in the already oscillating climatic conditions.

Significant differences in grain yield per unit area were recorded in different fertilization variants, in all three years of the research. In the F2 fertilization variant, where the treatment with NPK fertilizer and the preparation EM Aktiv was applied, the highest average yield was recorded, i.e., an increase of 14.05 % (4,228.70 kg ha⁻¹) in relation to the control (3,707.47 kg ha⁻¹). Also, the F1 treatment increased yield by 11.74% (4,143,01 kg ha⁻¹). Abduladin et al. (2020) stated that the incorporation of effective microorganisms into the soil has a positive effect on the number and activity of the autochthonous microbial population. Cvijanović et al. (2019) introduced different amounts of effective microorganisms, and obtained an increase in yield of 1.69% to 3.35% compared to the control. These results are correlated with the results of Dozet et al. (2015). In the study of Dozet et al. (2022), the use of effective microorganisms increased soybean yield by 7.28%, NPK fertilizer supplemented with microelements improved yield by 11.20%, while the combination of effective microorganisms and NPK fertilizer with microelements increased soybean yield by 14.86%.

Cvijanović et al. (2022) stated that the use of different levels and combinations of effective microorganisms in wheat production can significantly improve the quality and yield of different wheat genotypes by 1.53% to 5.24%. Similar results were obtained by Filho et al. (2017) in an experiment where only wheat was inoculated with *Azospirillum brasilense* species, and they found that higher yield was achieved with less than 139 kg N ha⁻¹ in inoculated wheat. In a two-year study by Moradgholi et al. (2021) an increase in wheat grain yield was obtained in treatments with a combination of Azotobacter and Phosphobacteria biofertilizers compared to the control.

Table 2.

Effect of fertilization treatment on grain yield (kg ha-1)

| Year (A) | Soybean variety | Fert | ilization treatmo | | Ξ.4 | |
|------------|-----------------|-----------------|-------------------|----------|----------|----------|
| | (B) | F0 | F1 | F2 | X AXB | ХA |
| 2016 | Galina | 4,098.75 | 4,485.95 | 4,597.14 | 4,393.95 | |
| | Valjevka | 4,694.83 | 5,160.67 | 5,307.57 | 5,054.36 | |
| | Sava | 4,223.51 | 4,658.83 | 4,792.95 | 4,558.43 | 4 724 00 |
| | NS Apolo | 4,416.86 | 4,847.78 | 4,997.56 | 4,754.07 | 4,724.90 |
| | Rubin | 4,651.84 | 5,163.52 | 5,332.10 | 5,049.15 | |
| | NS Zita | 4,149.30 | 4,623.19 | 4,845.91 | 4,539.47 | |
| | x AxC | 4,372.52 | 4,823.32 | 4,978.87 | | |
| | Galina | 2,442.34 | 2,796.86 | 2,902.38 | 2,713.86 | |
| | Valjevka | 2,533.45 | 2,842.01 | 2,948.26 | 2,774.57 | |
| | Sava | 2,604.15 | 2,941.24 | 3,067.14 | 2,870.84 | 2,964.76 |
| 2017 | NS Apolo | 2,463.76 | 2,732.22 | 2,850.58 | 2,682.19 | |
| | Rubin | 2,724.15 | 3,462.63 | 3,654.63 | 3,280.47 | |
| | NS Zita | 2,917.28 | 3,550.76 | 3,931.87 | 3,466.64 | |
| | x AxC | 2,614.19 | 3,054.29 | 3,225.81 | | |
| | Galina | 3,756.12 | 4,122.59 | 4,228.10 | 4,035.60 | |
| | Valjevka | 4,008.95 | 4,407.14 | 4,495.88 | 4,303.99 | |
| | Sava | 4,060.04 | 4,433.50 | 4,565.09 | 4,352.88 | |
| 2018 | NS Apolo | 4,212.79 | 4,644.38 | 4,795.24 | 4,550.80 | 4,460.63 |
| | Rubin | 4,352.21 | 4,823.96 | 5,023.33 | 4,733.17 | |
| | NS Zita | 4,424.10 | 4,877.01 | 5,060.82 | 4,787.31 | |
| | x AxC | 4,135.70 | 4,551.43 | 4,694.74 | ĀВ | |
| x̄ BxC | Galina | 3,432.40 | 3,801.80 | 3,909.21 | 3,714.47 | |
| | Valjevka | 3,745.74 | 4,136.60 | 4,250.57 | 4,044.30 | |
| | Sava | 3,629.23 | 4,011.19 | 4,141.73 | 3,927.38 | |
| | NS Apolo | 3,697.80 | 4,074.79 | 4,214.46 | 3,995.68 | |
| | Rubin | 3,909.40 | 4,483.37 | 4,670.02 | 4,354.26 | |
| | NS Zita | 3,830.22 | 4,350.32 | 4,546.20 | 4,242.25 | |
| | хc | 3,707.47 | 4,143.01 | 4,288.70 | | |
| | А | verage 2016–201 | erage 2016–2018 | | 4,04 | 6.39 |
| LSD | A** B** | C** | AxB** | AxC** | BxC** | AxBxC** |
| P < 0.01** | 536.42 397.48 | 552.14 | 612.23 | 572.61 | 607.08 | 682.01 |
| P < 0.05* | 380.20 288.69 | 398.47 | 446.82 | 408.16 | 440.22 | 524.62 |

The average value of protein content (40.52%) was statistically very significantly different (P < 0.01) at all factor levels (Table 3), except for the interaction of year x fertilization treatments and the interaction of all three factors, which were at the level of 5%, and the factor years, where there was no statistical significance. The highest protein content was determined in 2018 at 40.69%, compared to 2016 (40.41%) and 2017 (40.48%). The average values of the protein content in soybean varieties show that the highest value was recorded for Valjevka (41.30%), and it was statistically very significantly higher than in NS Zita (39.12%). Many reports have pointed out that the protein content

is determined by genetic potential. Soybean varieties with a shorter growing season contain a higher percentage of protein, while varieties with a longer growing season accumulate more oil in the grain (Đukić et al., 2017). On the other hand, the percentage of protein in the grain depends on climatic conditions, the fertility of the grassland, the applied fertilizer and the cultural operations used (Abuatwarat, 2018).

The effect of different variants of fertilization treatment on protein content was at the level of statistical significance P < 0.01, with the highest values being recorded in treatments with NPK fertilizer (40.57%) and NPK fertilizer and the preparation EM

increase in total protein and dry matter content

compared to the control. According to Abduladim

(2020), in three-year research on the application of

different amounts of NPK fertilizer and microbiological

preparation, the increase in protein content ranged

from 0.09% to 0.62%, which is in correlation with the

research of Gawęd et al. (2018).

Aktiv (40.57%), compared to the control (40.43%). The results are correlated with Kocira et al. (2018), who concluded that the application of biostimulants increased the content of fat and protein in seeds, compared to control samples. Radkowski et al. (2021) reported that foliar treatments of meadow grasses for silage production with effective microorganisms and in combination with amino acids resulted in a significant

Table 3.

Effect of fertilization treatment on protein content in grain (%)

| | Sovhean variety | - Fe | | | | | |
|------------|-----------------|----------------|-------|-------|---------|--------|--|
| Year (A) | (B) | F0 | F0 F1 | | — Ā AxB | ĀΑ | |
| 2016 | Galina | 40.48 | 40.65 | 40.55 | 40.56 | | |
| | Valjevka | 40.78 | 40.98 | 41.13 | 40.96 | | |
| | Sava | 40.38 | 40.63 | 40.58 | 40.53 | | |
| | NS Apolo | 39.75 | 39.95 | 40.13 | 39.94 | 40.41 | |
| | Rubin | 40.88 | 40.98 | 41.20 | 41.02 | | |
| | NS Zita | 39.33 | 39.58 | 39.50 | 39.47 | | |
| | x AxC | 40.26 | 40.45 | 40.51 | | | |
| | Galina | 41.13 | 41.53 | 41.53 | 41.40 | | |
| | Valjevka | 41.58 | 41.65 | 41.55 | 41.59 | | |
| | Sava | 40.48 | 40.53 | 40.53 | 40.51 | 40.40 | |
| 2017 | NS Apolo | 40.40 | 40.45 | 40.58 | 40.48 | 40.48 | |
| | Rubin | 39.58 | 39.58 | 39.70 | 39.62 | | |
| | NS Zita | 39.20 | 39.30 | 39.33 | 39.28 | | |
| | x AxC | 40.39 | 40.50 | 40.53 | | | |
| | Galina | 41.03 | 41.25 | 41.25 | 41.18 | | |
| | Valjevka | 41.23 | 41.43 | 41.38 | 41.35 | | |
| | Sava | 41.63 | 41.70 | 41.73 | 41.69 | | |
| 2018 | NS Apolo | 40.58 | 40.68 | 40.45 | 40.57 | 40.69 | |
| | Rubin | 40.85 | 40.80 | 40.60 | 40.75 | | |
| | NS Zita | 38.55 | 38.65 | 38.68 | 38.63 | | |
| | x AxC | 40.64 | 40.75 | 40.67 | | хB | |
| х ВхС | Galina | 40.87 | 41.14 | 41.10 | 41.04 | | |
| | Valjevka | 41.19 | 41.35 | 41.35 | 41.30 | | |
| | Sava | 40.82 | 40.95 | 40.94 | 40.90 | | |
| | NS Apolo | 40.24 | 40.35 | 40.38 | 40.32 | | |
| | Rubin | 40.43 | 40.45 | 40.50 | 40.46 | | |
| | NS Zita | 39.02 | 39.17 | 39.16 | 39.12 | | |
| | хc | 40.43 | 40.57 | 40.57 | | | |
| | | Average 2016–2 | 018 | | | 40.52 | |
| LSD | A B** | * C** | AxB** | AxC* | BxC** | AxBxC* | |
| P < 0.01** | 0.521 1.95 | 0.124 U.124 | 2.210 | 0.168 | 0.139 | 1.850 | |
| r < 0.05 | 0.303 1.45 | 0.079 | 1.033 | 0.124 | 0.092 | 1.301 | |

It is known that protein and oil content are negatively correlated (Gawęda et al., 2018; Sobko et al. 2019). The results for soybean seed oil content were opposite to protein content. In Table 4, the average value of the oil content (21.00%) differed statistically very significantly (P < 0.01) for all investigated factors. The highest oil content was determined in 2016 and 2018, 20.91%, while the lowest yield was recorded in 2017 (20.20%), which was correlated with the yield of soybeans. As regards the influence of the variety, on average, at all levels, the oil content increased according to ripening groups, from the shortest to the longest, that is, from Galina (20.13%) to NS Zita

(22.15%). The highest oil content, when it comes to the influence of fertilization treatment, was with NPK + EM Aktiv (21.06%). The negative correlation between protein and oil content hinders soybean breeding from improving the quality of soybean seeds; therefore, breeding should focus on developing varieties that have increased protein and oil content (Wang et al., 2019). Gawęda et al. (2018) found that the application of mineral fertilization with the addition of EM spraying had a favorable effect on the yield and protein content of soybean seeds of the Merlin variety compared to the control. Also, the same authors determined an opposite ratio in the case of percentage oil content.

Table 4.

| Year (A) | Soybean vari | iety | Fertilization treatments (C) | | | | ×۸ |
|--------------|-------------------|-------|------------------------------|--------|--------|-------|---------|
| | (B) | | F0 | F1 | F2 | X AXD | XA |
| | Galina | 2 | 20.73 | 20.58 | 20.85 | 20.72 | |
| | Valjevka | 2 | 21.63 | 21.50 | 21.70 | 21.61 | |
| | Sava | 2 | 22.28 | 22.20 | 22.33 | 22.27 | |
| 2016 | NS Apolo | 2 | 21.85 | 21.78 | 21.85 | 21.83 | 21.91 |
| | Rubin | 2 | 22.35 | 22.30 | 22.40 | 22.35 | |
| | NS Zita | 2 | 22.68 | 22.60 | 22.73 | 22.67 | |
| | x AxC | 2 | 21.91 | 21.825 | 21.975 | | |
| | Galina | 1 | 19.35 | 19.50 | 19.48 | 19.44 | |
| | Valjevka | 1 | 19.20 | 19.35 | 19.43 | 19.33 | |
| | Sava | 1 | 19.50 | 19.58 | 19.55 | 19.54 | 20.20 |
| 2017 | NS Apolo | 2 | 20.23 | 20.35 | 20.43 | 20.34 | 20.20 |
| | Rubin | 2 | 20.95 | 21.00 | 21.18 | 21.04 | |
| | NS Zita | | 21.45 | 21.48 | 21.58 | 21.50 | |
| | x AxC | 2 | 20.11 | 20.208 | 20.271 | | |
| | Galina | 2 | 20.20 | 20.25 | 20.30 | 20.25 | |
| | Valjevka | 2 | 21.33 | 21.38 | 21.38 | 21.36 | |
| | Sava | 1 | 19.85 | 19.95 | 19.98 | 19.93 | 20.91 |
| 2018 | NS Apolo | 2 | 20.80 | 20.83 | 20.95 | 20.86 | |
| | Rubin | 2 | 20.68 | 20.78 | 20.83 | 20.76 | |
| | NS Zita | 2 | 22.25 | 22.30 | 22.35 | 22.30 | |
| | x AxC | 2 | 20.85 | 20.913 | 20.96 | | хB |
| | Galina | 2 | 20.09 | 20.10 | 20.21 | 20.13 | |
| | Valjevka | 2 | 20.71 | 20.74 | 20.83 | 20.76 | |
| | Sava | 2 | 20.54 | 20.57 | 20.62 | 20.58 | |
| x BxC | NS Apolo | | 20.95 | 20.98 | 21.07 | 21.00 | |
| | Rubin | | 21.32 | 21.36 | 21.47 | 21.38 | |
| | NS Zita | | 22.12 | 22.12 | 22.22 | 22.15 | |
| | хc | 2 | 20.96 | 20.98 | 21.06 | | |
| | Average 2016–2018 | | | | | | 21.00 |
| LSD | A** | B** | C** | AxB** | AxC** | BxC** | AxBxC** |
| P < 0.01** | 1.802 | 2.210 | 0.142 | 2.247 | 0.199 | 0.145 | 0.250 |
| P < 0.05* | 1.330 | 1.633 | 0.099 | 1.665 | 0.157 | 0.099 | 0.200 |

| Effect of fertilization treatment on o | oil content in grain (| %) |
|--|------------------------|----|
|--|------------------------|----|

4. Conclusions

Based on the obtained results, it can be concluded that in unfavorable climatic conditions, soybean yield, protein and oil content decrease. Soybean varieties with a longer vegetation period showed better results at all test levels, compared to earlier soybean varieties. The use of NPK fertilizers with EM Aktiv had a statistically significant and positive effect on the yield and quality of soybeans. Also, the use of effective microorganisms increased the protein and oil content, and there was no negative correlation. The introduction of new technologies such as the application of effective microorganisms can result in high and stable yields, and their use along with regular NPK fertilizers in permanent production based on soil analysis is recommended.

Declaration of competing interests

No potential conflict of interest was reported by the authors.

References

- Abduladim, E. (2020), Interakcija genotipa soje (Glycine Max, L) i dubrenja na morfološke osobine biljaka, hemijski sastav i prinos zrna, Doktorska disertacija, Megatrend univerzitet, fakultet za biofarming, Bačka Topola, p. 211.
- Abduladin, E., Cvijanović, G., Đukić, V., Đurić, N., Miladinov, Z., Cvijanović, M., Abduladin, A. M. (2020). Uticaj efektivnih mikroorganizama na parametre biogenosti zemljišta u rizosferi različitih genotipova soje i prinosa soje. *Ratarstvo i povrtarstvo*, 57(3), 72-79. https://doi.org/10.5937/ratnov57-27962
- https://doi.org/10.5937/ratpov57-27962 Abuatwarat S. (2018). Uticaj sorte, organskog dubriva i mikrobioloskih preparata na prinos i kvalitet pasulja (Phaseolus vulgaris L.) gajenom po ekološkim principima. Doktorska disertacija. Megatrend Univerzitet, Fakultet za biofarming, Bačka Topola.

- Agostinho, P. R., Gomes, S. S., Gallo, A. S., Guimarães, N. F., Gomes, M. S., Silva, R. F. (2017). Biomassa microbiana em solo adubado com vinhaça e cultivado com milho safrinha em sucessão a leguminosas, *Acta Iguazu*, 6 (3), 31-43.
- Bajagić, M., Đukić, V., Miladinov, Z., Dozet, G., Cvijanović, G., Miladinović, J., Cvijanović, V. (2021). Effects of fall and spring primary tillage on soybean yield and 1000-grain weight in agroecological conditions of Serbia, *Book of abstracts of X International symposium on agricultural sciences AgroReS 2021*, 27-29, May, Trebinje, Bosnia and Herzegovina, 59.
- Bazzo, J. H. B., Santos, A. C., Marinho, J. L. (2021). Doses de potássio e boro em cobertura no desempenho produtivo da soja, *Agrarian*, 14 (54), 404-411.
- Chen, W., Chen, Y., Siddique, H. M. K., Li, S. (2022). Root penetration ability and plant growth in agroecosystems, *Plant Physiology and Biochemistry*, 183, 160-168. https://doi.org/10.1016/j.plaphy.2022.04.024
- Cobucci, T., NascentE, A. S., Lima, D. P. (2015). Adubação fosfatada e aplicação de Penergetic na produtividade do feijoeiro comum, *Revista Agrarian*, 8 (30), 358-368.
- Cunha, A. P. M. A., Zeri, M., Leal, K. D., Costa, L., Cuartas, L. A., Marengo, J. A., Tomasella, J., Vieira, R. M., Barbosa, A. A., Cunningham, C., Cal Garcia, J. V., Broedel, E., Alvalá, R., & Ribeiro-Neto, G. (2019). Extreme drought events over Brazil from 2011 to 2019. *Atmosphere*, 10 (11), 642-662. https://doi.org/10.3390/atmos10110642
- Cvijanović, G., Dozet, G., Đukić, V., Đorđević, S., Puzić, G. (2012). Microbial activity of soil during the inoculation of soya bean with symbiotic and freeliving nitrogen-fixing bacteria, *African Journal of Biotechnology* 11 (3), 590-597.
- Cvijanović, G., Simin, L., Stepić, V., Marinković, J., Đukić, V., Đurić, N., Cvijanović, V. (2019). Uticaj efektivnih mikroorganizama na visinu prinosa zrna kukuruza i biogenost zemljišta, Selo i poljoprivreda, Zbornik radova, Naučni skup sa međunarodnim učešćem, Bijeljina: Univerzitet Bijeljina, 124-131.
- Cvijanović, G., Čolić, N., Đurić, N., Dozet, G., Eltreki, A., Cvijanović, M., Žuža, M. (2020). Effect of biostimulants on soybean seedlings, *Acta Agriculturae Serbica*, 25 (50), 99-104.

https://doi.org/10.5937/AASer2050099C

- Cvijanović, G., Dozet, G., Đukić, V., Mamlić, Z., Bajagić, M., Đurić, N., Stepić, V. (2022). Uticaj primene različitih mikrobioloških preparata na masu 1000 zrna i prinos pasulja, Zbornik radova, Nacionalni naučno-stručni skup sa međunarodnim učešćem, Biotehnologija i savremeni pristup u gajenju i oplemenjivanju bilja, 3. novembar 2022., Smedervska Palanka, Srbija, 118-128.
- Cvijanović V., Cvijanović G., Rajičić V., Marinković J., Đukić V., Bajagić M., Đurić N. (2022a). Influence of different methods of application of effective microorganisms in nutrition of wheat on weight by 1000 grains, yield, and content of crude wheat proteins (*Triticum sp*), *Cereal Research Communications* 50, 1259–1268. https://doi.org/10.1007/s42976-021-00226-1
- Dozet, G., Cvijanovic, G., Vasic, M., Djuric, N., Jaksic, S., Djukic, V. (2015). Effect of microbial fertilizer application on yield of bean (*Phaseolus vulgaris* L,) in organic production system, *Proceedings of XXIII International Conference* "Ecological Truth", 17-20 June 2015, Kopaonik, Serbia, 105-107.
- Dozet, G., Đukić, V., Miladinović, J., Mamlić, Z., Cvijanović, G., Jakšić, S., Kandelinskaja, O. (2022). Uticaj folijarne primene NPK dubriva sa mikroelementima i efektivnih mikroorganizama na prinos soje, *Savetovanje o biotehnologiji sa međunarodnim učešćem*, Čačak, 27, 111-116.

https://doi.org/10.46793/SBT27.111D

- Đukić, V., Stojanović, D., Miladinov, Z., Vidić, M., Tatić, M., Dozet, G., Cvijanović, G. (2017). Kvantitativna i kvalitativna analiza NS sorti soje različitih grupa zrenja. 58. Savetovanje industrije ulja: Proizvodnja i prerada uljarica. Zbornik radova, Herceg Novi, Crna Gora, 67-73.
- Đukić, V., Miladinov, Z., Balešević-Tubić, S., Miladinović, J., Đorđević, V., Valan, D., Petrović, K. (2018). Kritični momenti u proizvodnji soje, Zbornik referata 52,

Savetovanja agronoma i poljoprivrednika Srbije (SAPS) i 1, Savetovanje agronoma Republike Srbije i Republike Srpske, Zlatibor, 21-27. Januar 2018., 34-44.

- Đukić, V., Miladinov, Z., Dozet, G., Cvijanović, M., Marinković, J., Cvijanović, G., Tatić, M. (2018a). Uticaj vremena osnovne obrade zemljišta na masu 1,000 zrna soje, *Zbornik naučnih radova Institut PKB Agroekonomik*, Beograd, 24 (1-2), 93-99.
- Đukić, V., Miladinović, J., Mamlić, Z., Vasiljević, S., Nikolić, Z., Bajagić, M., Kandeliskaja, O. (2021). Uticaj godine i sorte na pojedine morfološke osobine soje, Zbornik radova naučno-stručnog skupa sa međunarodnim učešćem "Biotehnologija i savremeni pristup u gajenju i oplemenjivanju bilja", 15, Decembar, Smederevska Palanka, Srbija, 293-300.
- Faria, L. O., Souza, A. G. V., Mello, M. C. de, Berti, M. P. da S. (2023). Potassium fertilization and bioactivators on the soybean yield and soil microbiota, *Pesquisa Agropecuária Tropical*, 53, e74945.

https://doi.org/10.1590/1983-40632023v5374945

- Filho, M. C. M. T., Galindo, F. S., Buzetti, S., Santini, J. M. K. (2017). Inoculation with *Azospirillum brasilense* improves nutrition and increases wheat yield in association with nitrogen fertilization, *InTech*. https://doi.org/10.5772/67638
- Gawęda, D., Haliniarz, M., Woźniak, A., Harasim, E. (2018). Yield, seed quality and nodule formation of soybean under application of effective microorganisms, *Acta Agrophysica*, 25 (1), 35-43. <u>https://doi.org/10.31545/aagr0003</u>
- Giovanini, A., Saath, K. C., Almeida, H. J. F. (2019). Ineficiências alocativas e sua relação com o crescimento da produtividade agropecuária internacional. *Revista de Economia e Agronegócio*, 17 (3), 420-441.
- He, Y., Matthews L. M. (2023). Seasonal climate conditions impact the effectiveness of improving photosynthesis to increase soybean yield, *Field Crops Research*, 296, 108907.

https://doi.org/10.1007/s12011-023-04031-1

- Hu, S., Liu, C., Liu, X. (2023). The beneficial effects of soybean proteins and peptides on chronic diseases, *Nutrients*, 15 (8), 1811. https://doi.org/10.3390/nu15081811
- Kocira, S., Szparaga, A., Kocira, A., Czerwińska, E., Wojtowicz, A., Bronowicka-Mielniczuk, U., Koszel, M., Findura, P. (2018). Modeling biometric traits, yield and nutritional and antioxidant properties of seeds of three soybean cultivars through the application of biostimulant containing seaweed and amino acids, *Frontiers in Plant Science*, 9, 388.
- MacCarthy, D. S., Traore, P. S., Freduah, B. S., Adiku, S. G. K., Dodor, D. E., Kumahor, S. K. (2022). Productivity of soybean under projected climate change in a semi-arid region of West Africa: Sensitivity of current production system, Agronomy, 12, 2614.

https://doi.org/10.3390/agronomy12112614

- Miladinov, Z., Stojanović, D., Đukić, V., Balešević-Tubić, S., Miladinović, J., Cvijanović, M., Dozet, G. (2017). Prinos i kvalitet novopriznatih NS sorti soje, Zbornik radova 58, Savetovanje industrije ulja: "Proizvodnja i prerada uljarica", 18-23, Jun 2017, Herceg Novi, Crna Gora, 75-82.
- Moradgholi, A., Mobasser, H., Ganjali, H., et al. (2021). WUE, protein and grain yield of wheat under the interaction of biological and chemical fertilizers and different moisture regimes, *Cereal Research Communications*. https://doi.org/10.1007/s42976-021-00145-1
- Olle, M., Williams, I. H. (2013). Effective microorganisms and their influence on vegetable production – a review, *Journal of Horticultural Science & Biotechnology*, 88 (4), 380-386.
- Persson, J., Hogberg, P., Ekblad, A., Hogberg, M. N., Nordgren, A., Nasholm, T. (2003). Nitrogen acquisition from inorganic and organic sources by boreal forest plants in the field, *Oecologia*, 137, 252-257.
- Petrović, G., Karabašević, D., Vukotić, S., Mirčetić, V., Radosavac, A. (2020). The impact of climate change on

the corn yield in Serbia, *Acta Agriculturae Serbica*, 25 (50), 133-140.

https://doi.org/10.5937/AASer2050133P

Radkowski, A., Radkowska, I., Bocianowski, J., Cyplik, A., Wolski, K., Bujak, H. (2021). Effect of amino acids and effective microorganisms on meadow silage chemical composition, *Agronomy*, 11, 1198.

https://doi.org/10.3390/agronomy11061198

- Santos, C. A.; Ribeiro, J. C. (2019). Os desafios para a agronomia no século XXI. Ponta Grossa: Atena, 2019.
- Silva, A. L., Cordeiro, R. S., Rocha, H. C. R. (2022). Aplicability of Efficient Microorganisms (EM) in agriculture: literature review, Research, *Society and Development*, 11 (1), e32311125054.

https://doi.org/10.33448/rsd-v11i1.25054

- Silva, S. D., Arima, Y. E. (2023). Sustainability consequences of making land change decisions based on current climatology in the brazilian cerrados, *Land* 12 (4), 914.
- Sobko, O., Hartung, J., Zikeli, S., Claupein, W., Gruber, S. (2019). Effect of sowing density on grain yield, protein and oil content and plant morphology of soybean (*Glycine max* L, Merrill), *Plant Soil Environment*, 65, 594-601. <u>https://doi.org/10.17221/346/2019-PSE</u>
- Staniak, M.; Szpunar-Krok, E.; Kocira, A. (2023). Responses of soybean to selected abiotic stphystosperiod, temperature and water. Agriculture, 13, 146. <u>https://doi.org/10.3390/agriculture13010146</u>
- Wang, J., Zhou, P., Shi, X., Yang, N., Yan, L., Zhao, Q., Yang, C., Guan, Y. (2019), Primary metabolite contents are correlated with seed protein and oil traits in nearisogenic lines of soybean, *The Crop Journal*, 7(5), 651-659.

https://doi.org/10.1016/j.ecoenv.2020.111330

Wu, A., Brider, J., Busch, F. A., Chen, M., Chenu, K., Clarke, V. C., Collins, B., Ermakova, M., Evans, G. R., Farquhar, G. D., Forster, B., Furbank, R. T., Groszmann, M., Hernandez-Prieto, M. A., Long, B. M., Mclean, G., Potgieter, A., Price, G. D., Sharwood, R. E., Stower, M., van Oosterom, E., von Caemmerer, S., Whitney, S. M., Hammer, G. L. (2023). A cross-scale analysis to understand and quantify the effects of photosynthetic enhancement on crop growth and yield across environments, *Plant, Cell & Environment*, 46, 23-44.

https://doi.org/10.1111/pce.14453

Yalçin, S. (2018). The effect of magnetic field on three different varieties of soybean seed. *Doğu Fen Bilimleri Dergisi*, 1(1), 1-8.

https://dergipark.org.tr/en/download/articlefile/542555

Zhang, Y., Zhou, F., Zhao, M., Ning, Z., Sun-Waterhouse, D., Sun, B. (2017). Soy peptide aggregates formed during hydrolysis reduced protein extraction without decreasing their nutritional value, *Food & Function*, 8, 4384-4395. https://doi.org/10.1039/C7F000812K