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# The effect of various inoculants on the productivity, chemical composition of soybean seeds (*Glycine max* (L. Merrill) Magnolia PZO) and chemical properties of soil in southwest part of Poland – preliminary studies

Karolina Dłutowska<sup>1</sup>, Bernard Gałka<sup>1</sup>\*, Anna Szuba-Trznadel<sup>2</sup>, Rafał Ramut<sup>1</sup>, Marcin Włodarczyk<sup>3</sup>, Anna Jama-Rodzeńska<sup>4</sup>, Wiesław Fialkiewicz<sup>5</sup>

<sup>1</sup>Institute of Soil Science, Plant Nutrition and Environmental Protection, Faculty of Life Sciences and Technology, Wrocław University of Environmental and Life Sciences, 50-363, Wroclaw, POLAND, <a href="mailto:bernard.galka@upwr.edu.pl">bernard.galka@upwr.edu.pl</a>, rafal.ramut@upwr.edu.pl
 <sup>2</sup>Department of Animal Nutrition and Feed Science, Faculty of Biology and Animal Science,
 Wroclaw University of Environmental and Life Sciences, 51-630, Wroclaw, POLAND, <a href="mailto:anna.szuba-trznadel@upwr.edu.pl">anna.szuba-trznadel@upwr.edu.pl</a>
 <sup>3</sup>COBORU, Research Centre for Cultivar Testing, SDOO Zybiszów, POLAND, <a href="mailto:m.wlodarczyk@zybiszow.coboru.gov.pl">m.wlodarczyk@zybiszow.coboru.gov.pl</a>
 <sup>4</sup>Institute of Agroecology and Plant Production, Faculty of Life Sciences and Technology, Wrocław, POLAND University of Environmental and Life Sciences, 50-363, Wroclaw, POLAND, anna.jama@upwr.edu.pl
 <sup>5</sup>Institute of Environmental Engineering, Faculty of Environmental Engineering and Geodesy, University of Environmental and Life Sciences, 50-363 Wrocław, POLAND

\*Correspondence author: e-mail: <u>bernard.galka@upwr.edu.pl</u>

Abstract. The introduction of bacterial inoculations into soybean cultivation is becoming an essential agronomic practice. The procedure of inoculating soybean seeds with microorganisms capable of fixing atmospheric nitrogen is recommended in situations where there is a deficit in soil native strains of symbiotic bacteria or their number is limited. The nodule bacteria enter the soil with the inoculated seeds and develop on the roots of the plants. Through this process, plants can use atmospheric nitrogen directly from the air. In 2023, a one-factor field experiment in a randomized block design in three replications was conducted at the Variety Evaluation Experiment Station (SDOO) in Zybiszow, Lower Silesia that belongs to Research Centre for Cultivar Testing. One soybean cultivar, Magnolia PZO, and four different inoculants - Liquifix Glycine 120, Turbosoy, Rhizobium Soi and Bi Soya - were applicated in the study. The inoculants were applied to the seeds immediately before sowing. The aim of the study was to determine the effect of the applied inoculants on soil abundance of basic elements, selected plant biometric traits, yield and chemical composition of soybean seeds. The soil was analyzed before the establishment of the experiment and after its liquidation for pH values, phosphorus, potassium and magnesium content. The applied inoculations had a positive effect on yield and number and weight of nodules and soybean yield. The greatest yield was observed under using Liquifix Glycine 120 (6.90 t/ha), while the lowest under using Rhizobium Soi (5.19 t/ha) compared to control plots (5.59 t/ha). Similar relation was observed in a case of nodule weight and number with the greatest value under Liquifix Glycine 120 (2.97 g and 27.33 pcs) compared to control plots (0.12 g and 1.33 pcs). However, there was no effect on the chemical properties of the soil and the results of chemical analyses of the seeds. The research presented here is preliminary field experiments using bacterial inoculations in soybean cultivation. Their aim was to evaluate the potential of these inoculants in increasing yields and improving biometric traits, which can contribute to sustainable agricultural development by reducing the need for mineral fertilizers. The results of this study may provide a basis for further work on biological methods of promoting plant growth, which is particularly important in the context of the growing need to reduce the environmental impact of agriculture.

Keywords: bacterial inoculations, soybean, yield, nodules, Liquifix Glycine 120, Turbosoy, Rhizobium Soi, Bi Soya



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# INTRODUCTION

Contemporary challenges facing the agricultural sector, such as climate change, soil degradation and the need for sustainable management of natural resources, posed to scientists and agricultural practitioners, create the need to look for innovative solutions (Gaind, 2011). One important aspect in today's agriculture is the excessive use of nitrogen fertilizers. As a result, there is a strong emphasis on implementing legumes in crop rotations, which makes it possible to break the dominance of cereal crop monocultures and allow for a reduction in the amount of nitrogen fertilizer doses (Abou-Shanab et al., 2017; Alori et al., 2017). Farmers are looking for effective fertilizers, leading to consider introducing legumes crops into their crop rotations due to the potential to use symbiosis and reduce the use of artificial nitrogen fertilizers (Engoke et al., 2022). In the context of this challenge, the development of modern agricultural technologies, including the use of inoculants in crop cultivation, is becoming increasingly important. Fabaceous plants establish a symbiotic relationship with soil bacteria, mainly of the Rhizobiaceae family, which are generally referred to as rhizobia, to fix atmospheric nitrogen. The studies conducted have shown that the amount of biologically fixed nitrogen can reach up to 300 kg N/ha, which provides up to 94% of the plant's requirements (Albareda et al., 2009).

Soybeans (Glycine max L. Merrill), an integral part of the world's animal as well as human nutrition system due to their valuable seeds, which are a source of protein and vegetable oil, are one of the key species undergoing these practices. The primary goal of using inoculants in the context of plants is to improve overall crop productivity. Soybeans, being a leguminous crop, benefit from symbiosis with nodule bacteria (Bradyrhizobium japonicum), which join nitrogen (N) from the air and supply it to the plant (da Silva et al., 2018). However, the natural supply of nodule bacteria in soils is usually insufficient, highlighting the importance of seed inoculation (Hungria et al., 2015). The inoculations used in soybean cultivation potentially affect soil chemistry. A full understanding of this issue is a key element for a comprehensive understanding of the interactions occurring between the plant, inoculant and soil.

The working hypothesis was that the tested inoculants would positively alter the soil chemical composition and biometric traits of the soybean. The purpose of this study was to conduct a comprehensive analysis of the effects of applied inoculation on soil chemical properties and biometric traits, yield and chemical composition of the Magnolia cultivar of soybean.

# MATERIALS AND METHODS

# **Research conditions**

In 2023, at the Experimental Station for Variety Evaluation (SDOO) in Zybiszów, a field study was conducted on the reaction of soybean to inoculants. The station is located about 11 kilometers east of Kąty Wrocławskie and 12 kilometers southwest of Wroclaw (geographical coordinates:  $\varphi = 51^{\circ}04'$ ,  $\lambda = 16^{\circ}55'$ ). The site is located at an altitude of 130 m above sea level. The unit directly reports to the Central Plant Variety Research Center (COBORU). This was a single-factor experiment using randomized block method where four inoculants were tested. In the experiment, the control object was plots without the application of bacterial inoculation but with full agronomic treatments as in the others (N, P, K fertilization).

# Characteristics of the inoculants used

1. Liquifix Glycine 120 (liquid form) – This product contains two bacterial strains of *Bradyrhizobium japonicum* reaching a minimum concentration of  $3 \times 10^9$  cfu (colony-forming units) per gram of product. The experiment used the manufacturer's recommended application rate – 4 liters of LiquiFix Glycine 120 combined with 1 liter of bacterial medium per ton of seed. The dressing process was carried out one day before sowing.

2. **Turbosoy** (liquid form) contains a minimum of 20 billion active bacteria per milliliter. The experiment used the manufacturer's recommended application rate -200 ml of Turbosoy combined with 50 ml of glue per 100 kg of seed.

3. **Rhizobium Soi** – the formulation includes carefully selected bacterial strains, with a concentration of not less than  $1 \times 10^9$  cfu/g. In the experiment, the mixture was treated in mixers and dissolving the preparation in a small amount of water (about 0.5 liters) and applying it as a finedrop spray on the seed before sowing.

4. **Bi Soya**  $-2 \times 10^9$  cfu/g of the preparation, deposited on sterilized shredded peat intended for soybeans. In the experiment, the dry method was applied according to the manufacturer's recommendations -3 kg of preparation per ton of seed.

The size of a single plot was about 16 m<sup>2</sup>. The experiment was conducted in 3 replications. Soybeans were sown on 5<sup>th</sup> of May at a density of 90 plants per m<sup>2</sup> at the depth 3–4 cm and the row spacing was 15 cm. On 10<sup>th</sup> of October, soybean seed harvesting was performed. After harvesting, the seed yield of each plot was determined. Soybean seed yields were brought to a constant moisture content of 13%.

#### Seed characteristics

In the experiment soybean variety Magnolia PZO was sown (https://igp-polska.pl/produkty/soja-i-groch/magnolia-pzo/). It was registered by the Central Plant Variety Testing Center in 2021 in the group of early and very early varieties at the request of IGP Poland Ltd. Plants of this variety are characterized by small height, with the lowest pods set at medium height. Pre-harvest lodging resistance is quite high. The uniformity of yield maturation is below average. However, resistance to pod cracking is high. The weight of 1,000 seeds is low - 148.4 g against a standard of 169.3 g. The total protein content of the seeds is 25.6% d.m.

#### Agrotechnical conditions

Soybeans were grown in the stand after winter wheat. Before the establishment of the experiment, winter plowing was carried out, and after pre-sowing fertilization, cultivation with a cultivating unit at a depth of 10 cm (18.03.2023) and then harrowing at a depth of 10 cm (18.03.2023) were done. In the experiment, pre-sowing fertilization was applied – Polifoska 6:20:30 fertilizer. Fertilization was aimed at providing 15 kg/ha of nitrogen, 50 kg/ha of phosphorus and 75 kg/ha of potassium (pure component dose). Crop protection treatments included herbicides and insecticide application (Table 1).

# Weather conditions

In 2023, the pattern of moisture and thermal conditions was not optimal for soybean development. The average air temperature during the growing season was lower than in the multi-year period. At the time of soybean sowing, an optimal temperature of 10 °C was recorded. However, during flowering, which took place at the end of June and the beginning of July, temperatures oscillating between 15 °C and 17 °C were recorded. In contrast, temperatures remained within the optimal range during the maturation

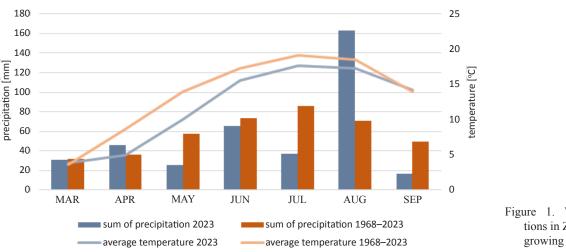


Figure 1. Weather conditions in Zybiszów during growing season in 2023.

Table 1. Crop protection treatments in the experiment.

Product	Active substance	Timing of application	Dose [l/ha]
Herbicides			
Corum 502.4 SL	bentazone 480 g/l imazamox 22.4 g/l	before emergence	1.25
Dash HC (adiuvant)			1
Stomp Aqua 455 CS	pendimethalin 455 g/l	12 BBCH	2
Select Super 120 EC	clethodim 120 g/l	14 BBCH	0.8
Amigo 95 EC (adiuvant)			1
Insecticide			
Cyperkill Max 500 EC	cypermethrin (a compound of the pyrethroid group) 500 g/l	67 BBCH	0.05

period. In 2023, most months had lower precipitation than the multi-year average, with the exception of April and August. April saw an increase in precipitation of 9.3 mm compared to the multi-year average, while August saw an increase of as much as 92.5 mm. Low precipitation in September had a positive effect on soybean yields (Fig. 1).

### **Soil conditions**

The experiment with soybeans and inoculants was established on a soil of a good wheat agricultural complex and of class IIIa (IUNG, 1990). Prior to the establishment of the experiment, the pH of the soil was determined, the value of which was 6.3. Both the soil complex and the pH corresponded to the plant requirements for the cultivation site for soybeans (Kotecki et al., 2020). Tests were also carried out to determine the content of phosphorus, potassium and magnesium in the soil. According to agrotechnical recommendations published by IUNG Pulawy (1990) magnesium levels were within the range, but potassium and phosphorus contents exceeded the recommended values (Table 2).

Table 2. Soil macronutrient content and soil pH in 2023 – SDOO in Zybiszów.

pН	P <sub>2</sub> O <sub>5</sub>	K <sub>2</sub> O	Mg
6.3		mg/100 g	
0.5	21.4	34.6	5

#### Plant material (sampling and chemical analysis)

During the growing season, five plants from each replication (each plot) were taken and the number and weight of nodules (during the flowering) were determined. After the end of the growing season, 10 shoots from each plot were taken for biometric analyses. Plants were taken from the middle row (0.5 meters from the edge of the plot) to avoid edge effects. In the collected material, the following were determined: plant height (cm), height of setting of the first pod (cm), number of first-row branches (pcs), number of pods per plant (pcs), root weight (g), pod weight (without seeds) (g), seed weight (g), stem weight (g), number of seeds per plant (pcs), thousand seed weight (TSW) (g). During the growing season, observations were made for the presence of pests, diseases, and weeds as well as for the determination of developmental stages.

Samples for chemical analysis were taken after harvesting from four replication and mixed into one sample. The concentration of nutrients was determined in a laboratory at the Institute of Agroecology and Plant Production, Wroclaw University of Environmental and Life Sciences, Poland. The seeds were ground using a laboratory grinder before analysis. Chemical analyses included the following:

- a) Determination of dry matter by the dryer method (DM, AOAC: 934.01) for laboratory samples was examined by the gravimetric method at 105 °C for 4 h according to Polish standards. The chemical composition of the seeds was assessed according to the Official Methods of Analysis of AOAC
- International (AOAC, 2012).
  b) Crude protein (CP, Kjeldahl method, AOAC: 984.13)

  crude protein content was assessed by multiplying the nitrogen percentage (N%) determined in the sample using a Kjeltec 2300 Foss Tecator apparatus (AOAC: 984.13) with a factor 6.25.
- c) Crude ash (CA, AOAC: 942.05) crude ash was investigated by combustion of the sample in a muffle furnace (Czylok Company, Jastrzębie-Zdrój, Poland) at 550 °C for 24 h.
- d) Ether extract (EE, Soxhlet method comprising the extraction with ethyl ether, AOAC: 920.39A) was analyzed with the use of a BUCHI Extraction System B-811 (BÜCHI, Flawil, Switzerland).
- e) Crude fiber (CF) was analyzed by the Hennenberg-Stohman method (AOAC: 978.10) using a Fibertec Tecator Foss apparatus for laboratory analysis.

Mineralization of plant material was completed using sulfuric acid and perhydrol in an electric furnace at 400 °C.

#### Soil samples

Soil samples were taken from the 0–20 cm layer after the end of the soybean growing season (October 2023) using a Egner stick. Soil from three locations was then mixed to form a subsample. We have also sampled the soil before experiment (March 2023) and analyzed according the normal procedure to test the changes of macroelements content in the soil and after harvest. Soil samples were airdried, disaggregated using a porcelain pestle and mortar, and sieved to < 2 mm. A portion of each sample was further finely ground for analysis. The soil was sieved through a sieve with a mesh diameter of 2 mm. Laboratory analyses included the following:

- determination of pH by potentiometric method. Soil pH was measured with a glass pH electrode (1:5 soil:deionized water, measurements after 30 min) and conductivity was assessed using a conductivity meter (the conductivity method).
- determination of  $P_2O_5$  and  $K_2O$  by Egner-Riehm method; Phosphorus and potassium compounds in a soil according to the Egner-Riehm method were extracted with a lactate buffer consisting of calcium lactate and lactic acid. The extraction solution used had pH = 3.55 (at this level of acidity, extraction conditions are maintained regardless of the initial soil reaction) (Egner et al., 1960).

 determination of bioavailable Mg by the FAAS method (Flame Atomic Absorption Spectrometry).

# Statistical methods

The analysis was performed using STATISTICA 13.3. According to the central limit theorem, the obtained results were assumed to have a normal distribution. The analysis of variance (ANOVA) was performed at a significance level of p < 0.05 using Statistica 13.1 (StatSoft, Kraków, Poland). Homogeneous groups were determined by Tukey's multiple range test using consecutive letters starting from "a"– the lowest to "d"– the highest value in terms of analyzed traits. Correlations were performed using Spearman's rho coefficient. Significance was assumed at  $\alpha = 0.05$  (p  $\leq 0.05$ ).

#### RESULTS

# Soybean development in 2023

The growth and development of soybeans can be divided into 9 stages depending on morphological characteristics. The length of the soybean growing season is the result of the interaction of the genetic factor as well as moisture and thermal conditions during soybean development. In the soybean development cycle, there are two critical periods concerning slightly higher thermal requirements, i.e. the moment from sowing to full emergence, during which

Table 3. Development of soybean cultivar Magnolia PZO in 2023.

Stage/phase	BBCH	Date of phase start
Sowing	00	05.05.2023
Emergence	10	13.05.2023
Flowering start	51	22.06.2023
Flowering	61	27.06.2023
Pod development	71	05.07.2023
Soybean harvest	99	10.10.2023

the occurrence of excessively low temperatures can extend this period up to 45 days (Szyrmer, Szczepańska, 1982). Emergence of soybean seeds began 13.05.2023, i.e. 8 days after sowing. Flowering of soybeans began in June 2023, while pods appeared in late July–August Plants were harvested in October 2023 (Table 3).

There were no significant (p > 0.05) differences in selected traits of soybean biometric measurements: plant height, first pod height, number of first-row branches, number of seeds per plant depending on the inoculants used (Table 4).

There were no significant (p > 0.05) differences in terms of root weight, stem weight and seed weight per plant depending on the inoculant used (Table 5).

Significant (p < 0.05) differences were shown in terms of yield depending on the inoculant used. Based on post hoc tests, it was presented that yield was significantly higher when Liquifix Glycine 120 was used compared to control and other inoculants (Table 6). The lowest yield was obtained using the inoculation with Rhizobium Soi. There were no statistically significant differences in WGT under innocluation applied.

Significant (p < 0.05) differences were found in terms of nodule weight depending on the inoculants used. It was shown that the weight of nodules was significantly higher when Liquifix Glycine 120 was used compared to control and other inoculants (Table 6).

Significant (p < 0.05) differences were found also in terms of the number of nodules depending on the inoculants used. It was shown that the number of nodules was significantly higher after Liquifix Glycine 120 application (Table 6).

Overall (regardless of the inoculants used), it was shown that the higher the plant, the higher the first pod set, the number of pods per plant and their weight. The number of pods and their weight also increased with the number of branches. In addition, the greater the number of pods on the plant, the greater their weight was also. On the other hand, with the weight of roots, the weight of stems increased. The greater the weight of pods was, the more the weight of stems decreased, as well as the number and weight of

Table 4. Morphological traits of soybean plants before harvest in 2023 (average and standard error).

Constitution	Control		Innocu	lants		
Specification	Control	Liquifix Glycine 120	Turbosoy	Rhizobium Soi	Bi Soya	— p value
Plant height [cm]	63.33 (±5.76)	62.93 (±5.24)	62.73 (±2.62)	63.90 (±3.44)	60.60 (±3.67)	ns
Number of branches of first row	1.06 (±0.06)	$0.73 \\ (\pm 0.08)$	0.90 (±0.10)	0.83 (±0.37)	1.30 (±0.11)	ns
Height of first pod [cm]	15.63 (±0.33)	16.27 (±0.23)	17.47 (±1.24)	17.37 (±0.84)	16.10 (±0.82)	ns
Number of pods per plant	18.87 (±0.91)	16.10 (±2.32)	15.60 (±1.02)	15.63 (±1.73)	15.70 (±7.1)	ns
Number of seeds per plant	49.9 (±2.6)	37.4 (±6.4)	37.3 (±3.2)	37.7 (±4.3)	37.4 (±1.4)	ns

Mean value (± standard error), ns - not significant difference

Creation Com		Inoculants					
Specification	Control	Liquifix Glycine 120	Turbosoy	Rhizobium Soi	Bi Soya	– p value	
		Weight of ur	nderground plant p	arts [g]			
Roots	11.17	14.55	14.13	14.31	19.76	na	
KOOIS	(±1.62)	(±0.59)	$(\pm 1.71)$	(±3.48)	(±1.79)	ns	
		Weight of ab	ove-ground plant p	oarts [g]			
Seeds	7.98	7.23	6.57	6.79	7.23	ns	
	(±5.97)	(±5.22)	(±4.56)	(±4.78)	(±5.22)		
Pods	1.10	1.01	1.00	0.94	1.04		
Pous	(±1.07)	(±1.50)	(±0.50)	(±1.39)	$(\pm 0.40)$	ns	
<u>C</u>	4.27	5.84	5.19	5.21	5.94		
Stems	(±2.99)	(±4.56)	(±2.30)	(±9.15)	(±4.59)	ns	
All together	13.35	14.08	12.76	12.94	14.21		

Table 5. Weight of selected elements of soybean plant in 2023 (average and standard error).

Mean value (± standard error), ns - not significant difference

Table 6. Seed yield, nodule weight and number depending on the inoculation used.

Specification	Control		Inocul	ants		- n voluo
Specification	Control	Liquifix Glycine 120	Turbosoy	Rhizobium Soi	Bi Soya	— p value
Soybean seed yield [t ha-1]	5.59 a (±0.10)	6.90 c (±0.12)	6.29 b (±0.17)	5.19 a (±0.16)	6.60 bc (±0.06)	0.011
TSW [g]	173.6 (±7.0)	186.0 (±7.0)	171.6 (±7.0)	170.0 (±7.0)	184.6 (±7.0)	ns
Nodule weight [g per 5 plants]	0.12 a (±0.01)	2.97 c (±0.26)	1.86 b (±0.08)	0.65 a (±0.03)	1.75 b (±0.11)	0.011
Number of nodules per plant	1.33 a (±0.33)	27.33 d (±0.23)	10.00 b (±0.57)	6.00 ab (±1.52)	17.00 c (±0.57)	0.009

Mean value ( $\pm$  standard error ), ns – not significant difference; Values in the rows with different letters differ significantly at  $p \le 0.05$ 

nodules. The number of seeds per plant, on the other hand, positively correlated with the seed weight of the plant. A positive correlation has been found between the number and weight of nodules and the yield size. The higher the number of nodules, the higher their weight (Table 7). There was also a significant positive relation between TSW, root weight and stem weight as well as roots weight and number of nodules. Stem weight was also positive correlated with seed yield, TSW, number and weight of nodules.

The soil pH value did not change under the influence of the applied inoculants. A 0.1 decrease in pH value for control, inoculant Liquifix Glycine 120 and Turbosoy is not significant for a given cultivation site. There were no differences between the pH values of the control plots and the sites where the inoculants were applied (Figure 2). The pH was stable for one season.

Soil phosphorus content increased only in the soil where Rhizobium Soi inoculation was applied, but this was a slight change ( $0.2 P_2O_5 mg/100 g$  of soil). The remaining plots showed a decrease in phosphorus content. The largest difference from the result before the establishment of the

experiment occurred on the soil of application of inoculant Bi Soya – a decrease of 7.6 mg/100 g of soil. There was also a decrease in phosphorus content in terms of application of inoculant Bi Soya relative to the control object –  $4.9 \text{ P}_2\text{O}_5 \text{ mg/100 g of soil (Figure 3)}$ .

Soil potassium content decreased after the experiment. The largest difference was observed in the control – 14.9 mg  $K_2O/100$  g of the soil, while the smallest difference was observed in the plots of Rhizobium Soi application – 9.3. There were no significant differences between the potassium content of the control and plots with inoculation (Figure 4).

Soil magnesium content increased after the experiment. The largest increase was observed in the area where inoculant Liquifix Glycine 120 was applied - 8.0 Mg mg/100 g of soil. There were no differences between the magnesium content of control and remain inoculants (Figure 5).

Chemical analyses did not show directional changes in nutrients and macroelement content in harvested seeds as effect of inoculation (Table 8).

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Biometric traits	Plant height [cm]	Height of first pod [cm]	Number of branches of first row [pcs]	Number of pods per plant [pcs]	Root weight [g]	Pod weight [g]	Stem weight [g]	Number of seeds per plant	Weight of seeds per plant	Seed yield [t/ha]	Weight of nodules of 5 plants [g]
Height of first pod [cm]	0.182*										
Number of branches of first row [pcs]	0.105	-0.029									
Number od pods [pcs]	0.393***	-0.158	0.608***								
Root weight [g]	0.027	-0.070	0.059	-0.224							
Pod weight [g]	0.387***	-0.038	0.574***	0.794***	-0.492						
Stem weight [g]	-0.131	-0.045	-0.188	-0.292	0.846***	-0.556*					
Number of seeds from plant [psc]	-0.084	0.029	-0.004	-0.335	0.036	0.045	0.043				
Weight of seeds from plant [g]	-0.059	660.0	-0.012	-0.278	0.118	0.050	0.111	0.971***			
TSW [g]	0.419	0.117	-0.056	0.007	0.584*	-0.259	0.543*	0.321	0.455		
Seed yield [t/ha]	0.084	0.032	-0.326	-0.317	0.493	-0.504	0.525*	-0.104	-0.032		
Weight of nodules of 5 plants [g]	0.068	-0.081	-0.328	-0.356	0.389	-0.520*	0.561*	-0.250	-0.250	0.821***	
Number of nodules [pcs] per plant	0.086	-0.023	-0.352	-0.370	0.547*	-0.663**	0.643**	-0.241	-0.191	0.881***	0.931***
* * / U OS ** * / U O1 *** * / U OU1	** * / 0 001										

Table 7. Correlation for biometric traits of soybean plants regardless of the inoculants used (total).

164

# Current Agronomy, 53/1, 2024

\* p < 0.05, \*\* p < 0.01, \*\*\* p < 0.001



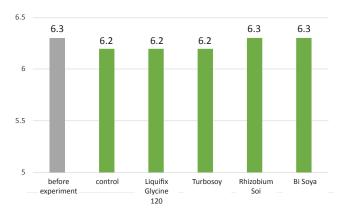


Figure 2. Soil pH value depending on the soybean bacterial inoculation (2023).

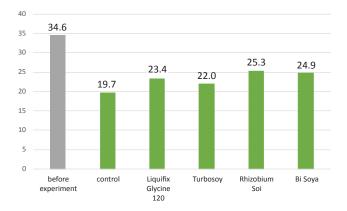


Figure 4. Potassium content [mg  $K_2O/100$  g of soil] depending on the soybean bacterial inoculation (2023).

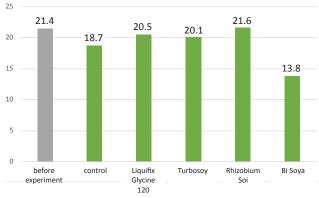


Figure 3. Phosphorus content [mg  $P_2O_5/100$  g of soil] in the soil depending on the soybean bacterial inoculation (2023).

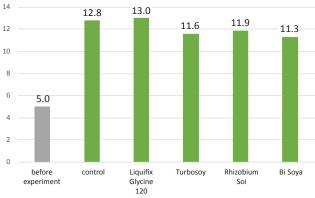


Figure 5. Soil magnesium content [mg Mg/100 g of soil] depending on the soybean bacterial inoculation (2023).

Guardi Caratian	Constant.		Inoculants				
Specification	Control	Liquifix Glycine 120	Turbosoy	Rhizobium Soi	Bi Soya		
		Nutrient	s [%]				
Dry matter	92.71	92.51	92.90	92.69	92.81		
Crude protein	30.27	28.08	30.93	31.38	31.56		
Crude fat	21.44	20.61	21.77	21.42	21.59		
Crude fiber	7.66	7.86	7.45	8.09	8.09		
Crude ash	6.73	6.70	6.72	6.17	6.19		
		Macroelem	ents [%]				
Nitrogen (N)	4.84	4.49	4.95	5.02	5.05		
Phosphorus (P)	0.79	0.75	0.66	0.65	0.71		
Potassium (K)	1.78	1.71	1.68	1.62	1.55		
Magnesium (Mg)	0.24	0.23	0.23	0.31	0.31		
Calcium (Ca)	0.07	0.07	0.08	0.07	0.07		

Table 8. Chemical composition of soybean seeds depending on the inoculation used.

# DISCUSSION

For the purpose of this study, the effects of application of four inoculates: Liquifix Glycine 120, Turbosoy, Rhizobium Soi and Bi Soya, were evaluated. In the available literature there are no studies evaluating these inoculants in soybean cultivation. Therefore, the studies cited address other inoculants and crop species, such as peas and lupins. Many studies have shown that the negative response of plants to various factors can be reduced by using biostimulants to increase plant yields (Kocira et al., 2018; Calvo et al., 2014; Koleska et al., 2017; Rymuza et al., 2023). In our study, there were no statistically significant differences in terms of plant height, first pod height, number of firstrow branches, number of pods per plant, number of seeds per plant, root weight, pod weight, stem weight and seed weight per plant depending on the inoculants used, but this does not mean that inoculants have no effect on plants.

In a study conducted by Sulewska et al. (2018) Rhizobium was used for yellow lupin seeds inoculation and it was shown that through it the number of pods formed on the plant significantly increased, and this difference compared to the control object was 1.1 and statistically significant. In contrast, in our own study, there were significant differences in yield depending on the type of inoculant used. Post hoc analysis revealed that the use of Liquifix Glycine 120 resulted in a significantly higher yield compared to Rhizobium Soi. On the other hand, it was noted that a higher number of branches translated into an increase in the number of pods per plant and their weight. In addition, a positive correlation was observed between the number of pods per plant and their weight. The results obtained in the experiment showed that the use of bacterial inoculants leads to an increase in soybean yields. The highest yields were obtained using Liquifix Glycine 120. Similar observations were made by Wei et al. (2023) in China, where it was found that inoculation of soybeans with appropriate rhizobium strains significantly increased seed yield by improving symbiotic nitrogen fixation. Inoculation led to an increase in the number and weight of nodules on the roots, which directly resulted in better plant growth and higher yields. Increased yield due to inoculation is also confirmed by a study conducted in Bangladesh (Alam et al., 2015). The experiment showed that rhizobium inoculation in soybean farming systems can increase soil microbial diversity. This, in turn, improves plant health and increases yields.

Most studies confirm the benefits of inoculation, indicating an increase in the number of pods per plant, the number of seeds per plant, straw yield, and the aforementioned yield after the application of commercial products (Zimmer et al., 2016; Leggett et al., 2017). In our study the yield and the number of nodules and its weight was differentiated under inoculants application.

An interesting perspective is also introduced by a study conducted by Serafin-Andrzejewska et al. (2024). It shows that soybean seed inoculation combined with moderate nitrogen fertilization (30 kg/ha) can show a synergistic effect on crop productivity. Integration of microbial technology with appropriately adjusted synthetic fertilization may yield the best results in terms of overall soybean productivity.

The results of the present study indicate that there is a relationship between the microbial preparations used and soil characteristics. Optimizing soil pH through various practices is beneficial, as it can enhance soil chemical properties and increase the availability of essential mineral nutrients (Bagayoko et al., 2000). However, changes are observed in the content of some minerals, such as phosphorus, but under the application of Rhizobium Soi, the content of which increased, and magnesium, which also showed an increase. In contrast, the potassium content of the soil showed a decrease after the experiment. The study of Bambara and Ndakidemi (2010) showed that Rhizobium inoculation significantly influenced soil chemistry, particularly by raising soil pH and increasing the availability of key nutrients such as calcium (Ca) and sodium (Na). Additionally, the inoculated treatments demonstrated a notable improvement in the levels of important micronutrients, including iron (Fe), copper (Cu), zinc (Zn), and manganese (Mn), when compared to non-inoculated control groups. The increase in nutrient availability resulting from Rhizobium inoculation may be attributed to the enhanced, favorable pH conditions, approaching near-neutral levels. This study of Bambara and Ndakidemi (2010) demonstrated that Rhizobium inoculation led to a numerical, though not statistically significant, increase in phosphorus (P) and potassium (K) concentrations as was showed in our study in a case of Mg. The improved growth of soybean plants due to inoculation may promote better development of the root system, which can facilitate the uptake of magnesium from the soil. Plants with a stronger root system can more effectively absorb nutrients, including magnesium. Inoculation of soybean seeds with Rhizobium bacteria leads to an increase in the number of these microorganisms in the soil. These bacteria, responsible for fixing atmospheric nitrogen, can influence the biogeochemical cycle in the soil, including the mobilization of other mineral components, such as magnesium (Khoso et al., 2024).

Enhancing the process of symbiotic nitrogen fixation through inoculation can effectively contribute to improved nitrogen availability and increased availability of other nutrients. Exploiting the mechanism of biological nitrogen fixation is an important factor in the context of optimizing soybean crop productivity.

# CONCLUSIONS

Bacterial inoculation of soybean seeds affected phosphorus, potassium and magnesium content in soil, compared to control treatment or soil before experiment. The direction and range of changes depended on preparation used. No differences were observed in the results of chemical analyses of soybean seeds after the application of the tested inoculants. The bacterial inoculants had also no significant effect on most biometric traits of soybean plants. In the experiment conducted, the average yield of soybeans in the three tested sites, where different inoculants were used (Liquifix Glycine 120, TURBOSOY, Bi Soya), was always higher than that of the control. These results suggest that inoculation of soybean seeds with rhizobia significantly contributes to increased yields. Inoculation had a significant effect on the weight and number of nodules. Significantly higher values were found in plants treated with Liquifix Glycine 120, Turbosoy and Bi Soya compared to the control.

The effects of inoculants on soil chemistry, seed chemistry and biometric traits of soybeans require further, more detailed research both in Poland and internationally. Results to date suggest a positive effect of inoculations on soybean yield, but a full understanding of the mechanisms of action of inoculations, their long-term effects on soil health and the optimal conditions for their application requires further experiments. Continuing this research is crucial to developing effective agronomic practices that can contribute to sustainable agricultural growth. The research requires further field studies to verify the data collected so far and that enables us to find answers to the questions that have arisen.

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# 168

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Author Bernard Gałka Anna Szuba-Trznadel Rafał Ramut Anna Jama-Rodzeńska Wiesław Fialkiewicz ORCID 0000-0002-4555-9460 0000-0002-4304-667X 0000-0002-1060-476X 0000-0003-4864-7367 0000-0002-2517-5064 received 11 September 2024 reviewed 25 October 2024 accepted 15 November 2024

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