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# Effect of *Azotobacter salinestris* on soil microbiological parameters and cucumber yield in integrated and organic farming systems

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Abstract. The aim of this study was to evaluate the effect of *Azotobacter salinestris* bacteria applied in the form of Rhizosum N plus preparation on the yield of cucumber cv. Gala F1 grown in organic and integrated systems. In addition, the effect of this preparation on soil microbiological parameters – *Pseudomonas* spp. and *Azotobacter* spp. number, dehydrogenases activity, biodiversity (Shannon coefficient) and microbial activity (AWCD) analyzed by BIOLOG tests – were studied.

In both cropping systems, the experiment included three treatments: control (full nitrogen fertilization); half dose of nitrogen fertilization; Rhizosum N plus preparation + half dose of nitrogen fertilization. The Rhizosum N plus formulation was applied at a rate of 25 g/ha as a spray on plants at the stage of 2–4 proper leaves. Cucumbers were harvested and segregated into the following fractions: canned, pickling and overgrown. Soil microbiological analyses and leaf analyses for *Azotobacter* spp. abundance were performed in three terms.

It was found that the abundance of *Azotobacter* sp. bacteria on cucumber leaves increased after spraying with Rhizosum N plus. The bacteria maintained a high abundance at least until 14 days after application. The preparation Rhizosum N plus had a slight effect on the increase of *Pseudomonas* bacteria number in the soil. No changes were found in dehydrogenases activity or in the biodiversity and functional activity of microorganisms in the soil in the treated combinations. In both cultivation systems, it was shown that the application of Rhizosum N plus had a positive effect on the yield of cucumbers. First of all, it accelerated fruit setting, which resulted in a higher yield at the first harvest. In addition, a positive effect on pickling and canning fractions was observed – a higher yield was obtained in the treatment with Rhizosum N plus + 1/2 N than in the "1/2 N fertilization" treatment.

These findings show that foliar spraying of *Azotobacter salinestris* in cucumber cultivation could be a sustainable way to promote plant growth and improve soil microbiological diversity.

Keywords: microbial activity, plant growth promoting bacteria, Cucumis sativus L.

### INTRODUCTION

The application of sustainable crop production methods is required by consumers. Chemical protection is being reduced for the reason of environmental protection and human health. The use of product containing plant growth promoting bacteria can be regarded as an alternative proposal for a more sustainable and environmental-friendly development of agriculture (Chiaranunt, White, 2023; Pylak et al., 2025). Special attention is also focused on improving soil biodiversity. Microbial biodiversity in the soil can be improved by use of beneficial microbes into cultivation practices. Increasing soil biodiversity through the application of biopreparations results in the multiplication of beneficial microorganisms and reducing population growth of microorganisms harmful to crops. Microorganisms are an important part of sustainable agriculture since their role is crucial for plant growth, soil fertility, and maintaining



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ecosystem stability. They exhibit numerous beneficial impacts on plants including nutrient fixation and solubilization, phytohormone production and growth improvement, plant protection against pathogens, abiotic stress management. The selected microorganisms are used in agriculture as commercial preparations, e.g. *Bacillus* spp., *Lactobacillus* spp., *Pseudomonas* spp., *Streptomyces* spp., *Arthrobacter* spp. Especially using alternatives methods of plant protection or improving plant yield is important in organic farming in which there are a limited number of approved agents for use and chemical agents are prohibited (Kowalska et al., 2020; Mącik et al., 2020; Antoszewski et al., 2022; Ptaszek et al., 2023).

Plant growth promoting rhizobacteria (PGPR) proved that they stimulate the growth and development of plants and conserving essential properties of soil ecosystem. PGPR is the alternate method in replacing the synthetic fertilizer for different food crops by fixing the atmospheric nitrogen and by also producing different growth promoting compounds (e.g. indole acetic acid, gibberellic acid). Among PGPR genera, species of Azotobacter play a vital role and are known to produce a wide variety of plant growth-stimulating secondary metabolites and directly influence vigor of the plant. Further, Azotobacter species produce certain antimicrobial compounds, which help limiting the plant diseases caused by different group of phytopathogens. Azotobacter species are known to tolerate and degrade synthetic pesticides and are potential bioagents for sustainable agriculture and maintain the ecological balance in the environment (Hindersah et al., 2020; Aasfar et al., 2021; Antoszewski et al., 2022; Minut et al., 2022; Shahrajabian et al., 2023).

One of the most active microorganisms is bacterium Azotobacter salinestris. The bacterium is a compound of commercial products, e.g. Rhizosum N Plus available in the Polish market. Rhizosum N Plus according to manufacturer, is a soil improver for agricultural, vegetable, orchard, horticultural and lawn crops. It contains the nitrogenous bacterium Azotobacter salinestris, which fixes nitrogen from the air, making it available to plants. The benefits of using the product include: stimulating plant growth, improving the biological activity of the soil, reducing nitrogen fertilizer rates by 20-50%, saving time and money, and making the product environmentally friendly. Rhizosum N Plus can be applied to all soil types, both before and after sowing, as well as to grassland, orchards and vegetable and ornamental crops. The product can be combined with foliar fertilizers and most herbicides and fungicides. There is limited information in the scientific literature on what effect the use of this product has on vegetable crops. In the study cucumber was selected because of its economic value in Polish production.

The aim of the study was to evaluate the effect of using commercial product Rhizosum N Plus containing bacterium *Azotobacter salinestris* on microbiological parameters of soil and on the yield of cucumber (*Cucumis sativus* L. variety Gala F1) in organic and integrated farming systems.

### MATERIALS AND METHODS

The experiments were carried out parallel at the certified organic field  $(51^{\circ} 96' 41.9'' \text{ N}; 20^{\circ} 17' 85.7'' \text{ E})$  using organic system and at the conventional field  $(51^{\circ} 96' 35.7'' \text{ N}; 20^{\circ}16' 17.3'' \text{ E})$  using integrated system of the National Institute of Horticultural Research in Skierniewice, Poland in 2022. In both farming systems the experimental area soil was luvisols.

At the certified organic field, before cucumber sowing (25.05.2022) the soil was weeded mechanically and fertilized with Fertilan PROFI (Poltops Ltd., Poland) at a dose 2.5 t/ha. Fertilan PROFI is an organic fertilizer, granulated, made from the biomass of small-seeded bean plants and ground pure sheep's wool (6–6.5% N, 0.15–0.20% P, 1.2–1.25% K and Mg, and microelements Fe, Mn, Cu, Zn, B). In conventional field adequate mineral fertilization (fertilizer chalk, ammonium nitrate and potassium sulfate) was applied before sowing seeds according to recommendation on the based chemical analysis. Drip irrigation was used as needed.

The experiment was established in randomized complete block design in 4 replications. The experimental plot area was 4 m<sup>2</sup> (1 m × 4 m). In each plot 40 cucumber seeds were sown in one row. The experiment included the following treatments:

- 1. control full nitrogen fertilization
- 2. half dose of nitrogen fertilization
- 3. preparation Rhizosum N plus + half dose of nitrogen fertilization.

Preparation Rhizosum N plus (Agrosimex company, Poland), containing bacteria *Azotobacter salinestris* ( $1.3 \times 10^6$  cfu/g) was applied at a rate of 25 g/ha as a spray on cucumber plants at the stage of 2–4 proper leaves (24.06.2022).

The following analyses were performed during the experiment:

- microbiological analysis of soil for the abundance of *Pseudomonas* spp. and *Azotobacter* spp.
- microbiological analysis of cucumber leaves for the abundance of *Azotobacter* spp.
- dehydrogenases enzyme activity in soil
- soil microbial biodiversity assessed by BIOLOG tests.

Microbiological analysis were performed by sowing soil suspensions or homogenized leaves on media selective for particular groups of microorganisms: the Gould medium (Gould et al., 1985) for fluorescent *Pseudomonas* enumeration in soil and medium according to Martyniuk and Martyniuk (2003) for *Azotobacter* enumeration in soil and in cucumber leaves. Surface sowing and flood sowing were conducted for *Pseudomonas* and *Azotobacter*, respectively. The analysis of soil were conducted in three terms, one week, one month and two months after Rhizosum N plus application, for each treatment in four replications. One sample from each experimental replication (approx. 1–1.5 kg of soil) was combined of six subsamples taken with an Egner's tool from depth of about 0–15 cm. The soil samples were thoroughly mixed. Serial dilution method was used and the suspension was sowing.

The analysis of leaves was conducted three times -24 h, 7 days and 14 days after Rhizosum N plus application for each treatment in four replications. One sample (10 leaves) was taken from 10 randomly selected plants from each plot. The leaves were cut and 10 g with 100 mL of 0.85% NaCl was homogenized in a stomacher BagMixer 400P, Interscience, France (8 strokes/s, 10 min). Serial dilution method was used and the suspension was flood sowing on medium for *Azotobacter*.

The abundance of studied microorganisms was expressed as the number of colony-forming units per 1 g of dry weight of soil or plant material (cfu/1 g).

Dehydrogenases activity (DHA) was measured in the soil samples taken in three different terms: one week, one month and two months after Rhizosum N plus application, according to Brzezińska and Włodarczyk (2005). In brief, 3 g of sieved soil was placed in a 15 mL dark tube, and added successively with 1.8 mL of sterile deionized water, 600 µL of 1% glucose suspension, and 600 µL of 3% water solution of 2,3,5-triphenyltetrazolium chloride (TTC). The mixture was incubated for 24 h at 30 °C in the darkness. The reaction was stopped by the addition of 12 mL ethanol (96%), and the mixture was agitated for one hour in the dark. After centrifugation (12,000 rpm/min, 8 min, 4 °C) the TPF concentration in the supernatant was measured spectrophotometrically at  $\lambda = 485$  nm (UviLine<sup>®</sup> 9600). The mean dehydrogenases activity was expressed in units of dehydrogenases activity, as the amount of TPF produced by 1 g of dry soil during 24 h [µmol TPF/g d.m. 24 h].

The biodiversity of bacteria populating the soil samples (Shannon coefficient H') and their microbial activity (AWCD) were analyzed using EcoPlate BIOLOG plates (Biolog Inc., USA) according to Ortiz-Burgos (2016) in two terms - one week and one month after Rhizosum N plus application. The system enables to evaluate the microbial activity towards 31 different potential carbon sources. A soil suspension from each sample was obtained by submerging 1 g of soil in 9 mL of sterile distilled water. The suspension was mixed on a rotary shaker for 20 min, and then serially diluted three times. The wells in the plates were inoculated with 100 µL of diluted soil suspension and incubated in the dark at 26 °C for 72 h. After incubation, the color development in each well was recorded measuring the absorbance at 590 nm (OD). The activity of microorganisms was evaluated calculating the average well color development (AWCD). Microbial diversity was calculated using the Shannon coefficient (H') formula.

Cucumber harvesting was conducted twice a week. The fruits were harvested from 22 July to 13 September, 15 times and 13 in the integrated and organic production, respectively. During harvesting, cucumbers were sorted into the following fractions: canned, pickling and overgrown. The fruit weight of each cucumber fraction was determined.

The obtained results were statistically analyzed using one-way analysis of variance with Tukey test for microbial analysis and Duncan test for yield analysis (P < 0.05), using the statistical program Statistica 13.1. Values not significantly different from each other were marked with the same letters.

### RESULTS

During the experiments in organic and conventional fields the number of *Pseudomonas* spp. and *Azotobacter* spp. bacteria in the soil was analyzed three times. In the conventional field with integrated system the number of *Azotobacter* bacteria in the I term was comparable in all treatments. In the II term – increased number of *Azotobacter* was observed compared to the I term, and was significantly higher in the "1/2 N fertilization" treatment and slightly higher in treatment with Rhizosum N plus + 1/2 N compared to the "full N fertilization" combination. In the III term, the abundance of these bacteria was the highest in the "1/2 N fertilization" treatment in the "1/2 N fertilization" combination. In the trem, the abundance of these bacteria was the highest in the "1/2 N fertilization" combination – 14.5 × 10<sup>5</sup> cfu/g. The number of *Pseudomonas* bacteria increased in the III term, being highest in the combination with Rhizosum N plus – 29.8 × 10<sup>3</sup> cfu/g (Table 1).

In the organic system the population of *Azotobacter* bacteria in the I term was comparable in all treatments, in the II term was the highest in the treatment with Rhizosum N plus, however, any statistical differences were not observed. The number of *Pseudomonas* bacteria was variable depending on the date and the treatment (Table 1). Leaf analysis obtained from integrated systems showed the highest number of *Azotobacter* sp. in the treatment with Rhizosum N plus + 1/2 N compared to the other two treatments. This relationship was observed at all three tested terms (Table 2). It can be concluded that these bacteria maintained a high abundance at least until 14 days after spraying. In organic system the phenomena was observed only in I term.

Measurements were made of dehydrogenases enzyme activity in soil taken at the three dates. There was no significantly different effect of Rhizosum N plus + 1/2 N treatment on this parameter. However, in the II term the activity in Rhizosum N plus treatment was much higher than in the "full N fertilization" treatment. In the III term the activity was similar in all treatments (Table 3).

During the experiments microbial activity and biodiversity were studied using BIOLOG tests. It was found that the Rhizosum N plus application caused increase the bio-

| Treature   | I term     |         | II term    |         | III term   |         |  |  |  |  |
|--|------------|---------|------------|---------|------------|---------|--|--|--|--|
| Treatment  | integrated | organic | integrated | organic | integrated | organic |  |  |  |  |
| Azotobacter sp. $(cfu \times 10^5/g)$                    |            |         |            |         |            |         |  |  |  |  |
| Full nitrogen fertilization                              | 0.45 a     | 1.1 a   | 7.2 b      | 5.6 a   | 5.7 b      | 3.2 a   |  |  |  |  |
| 1/2 nitrogen fertilization                               | 0.75 a     | 1.6 a   | 13.1 a     | 4.5 a   | 14.5 a     | 0.5 b   |  |  |  |  |
| Rhizosum N plus<br>+ 1/2 nitrogen fertilization          | 0.68 a     | 0.6 a   | 10.4 b     | 9.2 a   | 3.0 b      | 1.6 b   |  |  |  |  |
| <i>Pseudomonas</i> sp. (cfu $\times$ 10 <sup>3</sup> /g) |            |         |            |         |            |         |  |  |  |  |
| Full nitrogen fertilization                              | 13.3 a     | 29.3 a  | 1.3 a      | 7.0 b   | 8.7 b      | 21.4 a  |  |  |  |  |
| 1/2 nitrogen fertilization                               | 7.3 a      | 8.0 b   | 1.7 a      | 14.8 ab | 10.9 b     | 20.2 a  |  |  |  |  |
| Rhizosum N plus<br>+ 1/2 nitrogen fertilization          | 12.0 a     | 5.3 b   | 0.2 b      | 30.4 a  | 29.8 a     | 28.8 a  |  |  |  |  |

Table 1. The number of Azotobacter sp. and Pseudomonas sp. in soil in the integrated and organic production of cucumber.

Values marked with the same letters in each column within each group of microorganisms are not significantly different according to Tukey test, P < 0.05.

Table 2. The number of Azotobacter sp. on the cucumber leaves in integrated and organic production.

| Treatment                                       | I term     |         | II term    |         | III term   |         |  |  |  |
|---|------------|---------|------------|---------|------------|---------|--|--|--|
|   | integrated | organic | integrated | organic | integrated | organic |  |  |  |
| Azotobacter sp. (cfu $\times 10^2$ /g)          |            |         |            |         |            |         |  |  |  |
| Full nitrogen fertilization                     | 5.3 b      | 16.7 b  | 5.0 b      | 6.0 b   | 23.7 b     | 15.1 a  |  |  |  |
| 1/2 nitrogen fertilization                      | 5.3 b      | 16.7 b  | 3.6 b      | 60.9 a  | 37.6 b     | 35.0 a  |  |  |  |
| Rhizosum N plus<br>+ 1/2 nitrogen fertilization | 17.5 a     | 22.4 a  | 45.9 a     | 10.4 b  | 95.2 a     | 34.9 a  |  |  |  |

Values marked with the same letters in each column are not significantly different according to Tukey test, P < 0.05.

Table 3. Dehydrogenases activity in the soil [ $\mu$ mol TPF/g d.m. of soil] in the integrated and organic production of cucumber (means  $\pm$  SE).

| Treatment -                                     | I term          |                  | II te         | erm           | III term       |              |  |
|---|-----------------|------------------|---------------|---------------|----------------|--------------|--|
|   | integrated      | organic          | integrated    | organic       | integrated     | organic      |  |
| Full nitrogen fertilization                     | $15.2\pm7.73$   | $17.6\pm4.41$    | $11.2\pm0.75$ | $18.0\pm2.72$ | $7.8\pm0.21$   | $4.7\pm2.03$ |  |
| 1/2 nitrogen fertilization                      | $14.8 \pm 3.84$ | $16.9 \pm 10.52$ | 26.9 ± 2.39   | $20.2\pm3.97$ | 7.4 ± 1.57     | 6.0 ± 1.41   |  |
| Rhizosum N plus<br>+ 1/2 nitrogen fertilization | 13.4 ± 4.29     | $17.3 \pm 6.06$  | 21.7 ± 2.87   | 24.1 ± 4.14   | $7.7 \pm 0.50$ | $4.8\pm0.70$ |  |

The data in each column are not significantly different according to Tukey test, P < 0.05.

## Table 4. Indices of microbial metabolic potential – Shannon (H') and biodiversity AWCD in soil in the integrated and organic production of cucumber (means $\pm$ SE).

|   | I term          |                 |                 |               | II term         |                 |                 |                 |
|---|-----------------|-----------------|-----------------|---------------|-----------------|-----------------|-----------------|-----------------|
| Treatment                                       | integrated      | organic         | integrated      | organic       | integrated      | organic         | integrated      | organic         |
|   | H'              |                 | AWCD            |               | H'              |                 | AWCD            |                 |
| Full nitrogen fertilization                     | $1.30\pm0.79$   | $2.82\pm0.13$   | $0.07\pm0.09$   | $0.78\pm0.15$ | $2.67\pm0.19$   | $2.79\pm0.20$   | $0.61\pm0.18$   | $0.74\pm0.17$   |
| 1/2 nitrogen fertilization                      | $2.35\pm0.36$   | $2.81 \pm 0.10$ | $0.31 \pm 0.16$ | 0.86 ± 0.11   | $2.63 \pm 0.33$ | 2.91 ± 0.10     | $0.57 \pm 0.21$ | $0.94 \pm 0.11$ |
| Rhizosum N plus +<br>1/2 nitrogen fertilization | $1.62 \pm 0.72$ | $2.90 \pm 0.08$ | $0.25 \pm 0.22$ | 0.96 ± 0.09   | $2.60 \pm 0.24$ | $2.92 \pm 0.09$ | $0.52 \pm 0.20$ | $0.84 \pm 0.12$ |

The data in each column are not significantly different according to Tukey test, P < 0.05.

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Figure 1. Cucumber yield harvesting in 15 terms in the integrated cultivation. Values marked with the same letters in each term are not significantly different according to Duncan test, P < 0.05.



Figure 2. Total cucumber yield and yield fractions in the integrated cultivation.

Values marked with the same letters in each fraction are not significantly different according to Duncan test, P < 0.05.

diversity and functional activity of microorganisms compared to control in the first term in both farming systems. The differences were not significant. In the II term, the values were very equal in all treatments, in both farming systems. However, it is important to mention that the parameters were higher in organic system than in the integrated system, in both the I and the II terms of analysis (Table 4).

Cucumber fruit harvesting was carried out twice a week starting from the  $22^{nd}$  July to the  $13^{th}$  September (Fig. 1–4). In the integrated system the overall fruit yield is as follows: 88.1 t/ha; 76.6 t/ha and 86.7 t/ha for the "full N fertilization" treatment, "1/2 N fertilization" and "1/2 N fertilization + Rhizosum N plus", respectively (Fig. 2). For the treatment with Rhizosum N plus total yield and yield of individual fractions were higher than for the "1/2 N fertilization" treatment. However significant differences were obtained only for pickling fraction. During 12 out of the 15 all harvests, a higher total yield was obtained with Rhizosum N plus than in the "1/2 fertilization" treatment.



Figure 3. Cucumber yield harvesting in 13 terms in the organic cultivation.

Values marked with the same letters in each term are not significantly different according to Duncan test,  $P \le 0.05$ .



Figure 4. Total cucumber yield and yield fractions in the organic cultivation.

Values marked with the same letters in each fraction are not significantly different according to Duncan test, P < 0.05.

However, significant differences were obtained in the first term when the highest yield was obtained in treatment with Rhizosum N plus (Fig. 1).

During the experiments in the organic system, unfavorable weather conditions occurred, especially some flood, which had a very negative impact on the emergence, growth and yield of the plants. The plots were flooded several times by violent downpours and storms. As a result, many seeds failed to germinate, some plants died during vegetation, and other plants were characterized by poor growth and low yield. Harvesting of cucumbers was carried out during 13 harvest. The overall fruit yield is as follows: 19.4 t/ha; 8.0 t/ha and 14.6 t/ha for "full N fertilization", "1/2 N fertilization", "Rhizosum N plus + 1/2 N fertilization", respectively (Fig. 4). In case of treatment with Rhizosum N plus total yield and yield of individual fractions were higher than in the "1/2 fertilization" combination, however the differences were not significant. Additionally, fruit yield at individual 13 harvest dates was always higher in Rhizosum N plus treatment than in the "1/2 fertilization" treatment (Fig. 3). The application of Rhizosum N plus in both systems had a positive effect on the yield of cucumbers, first of all, it accelerated fruit set, which was evident in higher yield during the first several harvests compared to "1/2 N fertilization" treatment.

### DISCUSSION

Interest in *Azotobacter* group is related to their properties that can be useful in agriculture. In view of the current situation when methods of improving the quality of plants that are safe for the environment and for humans are being sought, the possibility of using microorganisms for this purpose acquires particular importance.

Soil is a specific component of the natural environment. The abundance of *Azotobacter* spp. in soils varies widely, ranging from a few cells to several hundred thousand colony forming units in 1 gram of soil (Kozieł et al., 2021; Kozieł, 2023). In the presented studies the population of *Azotobacter* in soil in both cultivation systems was high. However, it is important that unpredictable weather condition in organic system had negative influence on these bacteria number which was occurred in the third term of study. These findings are in agreement with studies of Kozieł (2023) which show that *Azotobacter* are sensible to unfavorable weather condition and according to O'Callaghan et al. (2022) it could be a limitation of using microorganisms in agriculture.

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In the studies increased amount of *Pseudomonas* spp. in soil treated with Rhizosum N plus was observed especially two months after the product application. It could be very positive for plants because many *Pseudomonas* spp. fix an atmospheric nitrogen, solubilize phosphorus and potassium, and also produce phytohormones, lytic enzymes, volatile organic compounds, antibiotics, and secondary metabolites during stress conditions. These compounds stimulate plant growth by inducing systemic resistance and by inhibiting the growth of pathogens. Furthermore, pseudomonads also protect plants during different stress conditions like heavy metal pollution, osmosis, temperature, oxidative stress, etc. (Mehmood et al., 2023; Pylak et al., 2025).

Dehydrogenases activity in soil is an indicator of the intensity of respiratory metabolism of soil microorganisms, mainly bacteria and actinomycetes. The change in the oxygenation state of the soil significantly modifies the activity of this enzyme. In the presented studies dehydrogenases activity in soil in treatment with Rhizosum N plus increased compared to "full N fertilization" treatment in II term of analysis, in both cropping systems. However, in III term, there were not differences among all treatments. It is assumed that the Rhizosum N plus spray remained on the leaves and it did not affect significantly the change of the soil microbiome or it is possible that the effect would be observed at a later date.

Microbial activity and biodiversity of soil samples were studied using BIOLOG tests. This method is based on physiological profiling of population levels. The use of different carbon sources by environmental microorganisms is assessed. The activity of whole populations of microorganisms from a given environment is assessed. Functional diversity of microorganisms is as important as structural diversity, because by performing different functions the microbiome participates in many processes in the soil environment. Microbial biodiversity and microbial activity in soil mainly depend on the tillage system, intercropping, rotations, cover crops and also the application of natural fertilizers or the use of other exogenous organic matter (Wolińska et al., 2017; Frac, 2019; Vincze et al., 2024). In the presented study microbial biodiversity and activity in soil did not change significantly after Rhizosum N plus application. It is assumed that the differences in these parameters could be observed at a later date, several months after the application of the product with *Azotobacter salinestris*. According to Jezierska-Tys et al. (2020) higher enzymatic activity and biodiversity of soil microorganisms could be noted in the next season after bioproduct application.

In the study *Azotobacter* bacteria applied as Rhizosum N plus commercial product, showed positive effects on cucumber yield in both cropping systems – integrated and organic, compared to "1/2 N fertilization" treatment. First of all, it accelerated fruit setting, which resulted in a higher yield at the beginning of harvesting. In addition, a positive effect on pickling and canning fractions was observed – a higher yield was obtained than in the "1/2 N fertilization" combination. In conventional system the differences were statistically significant for pickling fraction.

In the literature many reports are presented relating the effect of Azotobacter bacteria on the yield of different plants and their influence on differentiated growth parameters. Kurrey et al. (2018) showed positive effect of Azotobacter on growth and yield of onion and also biochemical properties of soil. Foliar application of these bacteria is often used with promising results (Razmjooei et al., 2022). Sagar et al. (2022) presented the positive effect of Azotobacter nigricans on the yield of maize (Zea mays L.). However, it was also noticed that soil application gave higher maize yield values with statistically significant differences compared to the foliar application (Efthimiadou et al., 2020). Based on the results we could assume that maybe an additional soil application of Rhizosum N plus could obtain greater increase in cucumber yield and perhaps improve soil microbial biodiversity.

On the other hand, there are many reports on the positive response of cucumber plants to growth-stimulating bacteria, especially in the extreme weather conditions (Kang et al., 2015; Kumar et al., 2018; Kartik et al., 2021; Zapata-Sifuentes et al., 2024). Weather condition and climate changes have impact on cultivated plants (Faber, Jarosz, 2024) and using biopreparations (Macik et al., 2020). In the presented study during experiments conducted in the organic system, unpredictable weather conditions appeared, mainly the flood which drastically affected the cultivation of cucumber, especially decreased yield. However, what is the most important in the study, Rhizosum N plus showed positive tendency on the increasing of the yield.

The role of *Azotobacter* is not only to increase plant growth and yield but also to reduce the chemical fertilizer level. In the relation to climate change issues, *Azotobacter* as biofertilizer is a potential bioagent to reduce ammonia and nitrous oxide emission and nitrate leaching. Using Rhizosum N plus is especially important in organic farming because in the system there is a limited number of registered preparations to support plant growth and yield. Organic farming helps reduce dependence on artificial plant protection inputs in crop production. Nowadays, the production and consumption of ecological food is still growing and organic agriculture is one of the fastest developing branches of agriculture.

### CONCLUSIONS

In general, based on the results obtained in this publication, foliar application of *Azotobacter* sp. can be used to promote plant growth, especially yield of *Cucumis sativus* L. variety Gala F1 in organic and integrated systems. It can reduce nitrate mineral fertilization of cucumber which could have positive effect on the natural environment. Further studies are needed to confirm positive effect on quality and quantity of macro- and microelements present in cucumber fruits. Moreover, it would be interesting to conduct similar experiments using other cucumber cultivars. Foliar spraying of *Azotobacter* could be a financially effective and sustainable substitute to promote plant growth and nutrition and reduce nitrate accumulation in vegetable crops. Finally, expanding the research to other major vegetable crops would also be valuable.

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