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Assessment of the Rhizosphere Bacterial Community under Maize Growth Using Various Agricultural Technologies with Biomodified Mineral Fertilizers

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Abstract: Biomodified mineral fertilizers (BMFs) were produced by enriching the ammophos fertilizer with PGPR *Bacillus velezensis* BS89 with the use of two technologies: BMF 1, the ammophos fertilizer with the addition of spores of *Bacillus velezensis* BS89 on a dry carrier (diatomite); and BMF2, ammophos granules treated with spores of *Bacillus velezensis* BS89 in a cell suspension. The effects of BMFs on maize growth and productivity and the rhizosphere bacterial community were assessed. BMFs significantly increased maize growth, dry matter, minerals, starch and protein contents in maize grain. The application of biomodified mineral fertilizers resulted in the significant increase in the yield and some parameters of maize plants such as ear length and number of kernels in the row. The yield was increased by 7.5–7.6%, ear length by 9%, and number of kernels in the row by 6.7–7%, as compared with ammophos. However, we found no considerable differences in the composition of the bacterial community of the maize rhizosphere after the use of BMFs as compared with the use of ammophos at the level of the phyla, which was confirmed by the ecological indices of biodiversity: the Shannon index and the Simpson index. Comparison of the experimental variants with bulk soil showed differences in the microbiome composition of the dominant bacterial phyla. A greater abundance of *Proteobacteria* and *Bacteroidetes* and a lower abundance of *Chloroflexi* was registered in bulk soil as compared with the other experimental variants where maize plants were present. The highest percentage (5.3%) of unidentified taxonomic phyla was also found in bulk soil. Our studies showed that maize is the main structuring factor during formation of the microbiome composition in the rhizosphere. The application of biomodified fertilizers BMF1 and BMF2 considerably increased the abundance of bacteria representing the minority of the community, namely, those from the phyla *Verrucomicrobia*, *Chloroflexi*, *Planctomycetes*, *Proteobacteria*, *Firmicutes* and *Chlamydiae*, as compared with the use of ammophos. Thus, the application of biomodified mineral fertilizers is a promising agronomic and ecological strategy for boosting maize yield and the quality of grain under field conditions.

Keywords: plant microbial biosystems; PGPR; *Bacillus velezensis* BS89; biomodified mineral fertilizers

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1. Introduction

The microbial community of the rhizosphere is constantly involved in interactions with plants, providing a constant taxonomic composition of the microorganisms in biosystems

and freely altering their quantitative composition [1,2]. Plants release root exudates, such as amino acids, sugars, carboxylic acids, and various secondary metabolites. These exudates are used as energy sources by soil microorganisms, which provide plant growth and sustainability by production of a range of plant-growth-promoting substances [3,4]. In this way, an increased stability of biosystem transformation processes and a balance of biochemical flows between the partners of plant–bacterial association is achieved.

Maize (*Zea mays* L.) is one of the most significant crops in the world and it is necessary to provide both food and energy [5,6]. Maize is cultivated in many countries around the world and it is an important source of food, animal feed, and fuel [7,8]. Fertilization and the applied hybrid have a significant influence on maize yield [9,10].

Plant-growth-promoting rhizobacteria (PGPR) inhabit the plant rhizosphere and rhizoplane, interact with root exudates, and form strong associations with plants [11–13]. PGPR are promising candidates for applications in agriculture as biofertilizers and biocontrol agents [14]. They can also be used in the production of inoculants to promote plant growth and nutrition [15]. Certain PGPR employ not one but an entire array of mechanisms of plant-growth stimulation, including nitrogen fixation; the synthesis of phytohormones, such as indole-3-acetic acid (IAA) and organic acids; HCN production; the release of enzymes, such as soil dehydrogenase, phosphatase, and nitrogenase; the antagonism of pathogenic fungi; the production of siderophores; increasing the solubilization of phosphates; and the induction of systemic resistance [15,16].

A possible way towards the application of microbial preparations based on PGPR is their use for treating the granules of mineral fertilizers [17,18] or organic mineral fertilizers [19]. The mechanism of the action of these preparations is based on the fact that the PGPR used for treating the granules increase the availability of nutrients contained in mineral fertilizers and mobilize their reserves in soil as well. PGPR also produce amino acids, vitamins, hormones, and organic acids, which promote the growth of the plant and enhance its immunity. Last but not least, the microorganisms synthesize substances blocking the development of phytopathogens [20,21]. Thus, a "biocapsule" formed on the surface of pelleted mineral fertilizers after the treatment has multiple functions such as fertilization, protection, and stimulation. A combined beneficial effect may result in a considerably increased yield of agricultural crops and thus an increased pay-off of the mineral fertilizers [22].

It was reported that new bio-activated organic fertilizers have been successfully applied for maize cultivation [23,24]. The beneficial effects of bio-activated mineral fertilizers on maize physiology and yield have been reported [23], but the effect of these fertilizers on the maize rhizosphere community has not been described yet.

In our work, we used the PGPR strain *Bacillus velezensis* BS89 (previously identified as *Bacillus subtilis* Ch13) [25]. This strain was originally isolated from the roots of winter wheat plants, cv Lira, growing on chernozem soil in the Republic of Moldova [26]. In vitro studies aimed at identifying potential plant-beneficial metabolites of this strain have shown that it produces a mix of auxins, hydrolytic enzymes, and vitamins. The genome analysis of strain BS89 revealed the presence of gene clusters responsible for the synthesis of plant-growth-promoting metabolites (indole 3-acetic acid [IAA] and volatiles), numerous hydrolytic enzymes, vitamins, and antimicrobial compounds (surfactin, fengycin, bacilysin, macrolactin, difficidin, bacillaene, and plantazolicin) [27]. It was also demonstrated that strain *Bacillus velezensis* BS89 can enhance the assimilation of minerals such as Ca and P [28].

Two forms of biomodified mineral fertilizers (BMFs) were used for this study. BMF1 was the variant with ammophos fertilizer with the addition of spores of *Bacillus velezensis* BS89 on a dry carrier (diatomite) and BMF2 was the variant with ammophos granules treated with a spore suspension of *Bacillus velezensis* BS89.

The aim of this study was to evaluate the effect of biomodified ammophos on the maize yield and grain quality and the changes in the microbial community of the maize rhizosphere under the influence of biomodified mineral fertilizers.

2. Materials and Methods

2.1. Plant Material, Soil, Cultivation, and Experimental Design

The field cultivation of the maize plant (*Zea mays* L.) cv. Pioneer P 9578 hybrid was conducted in 2017 in the Krasnoarmeisky Settlement named after Maistrenko of Kuban State Agrarian University, Krasnodar, Russia (45.209483° N, 38.300953° E) (Figure S1). Maize cv. Pioneer P 9578 is a mid-late variety for grain purpose. It is the most popular Pioneer hybrid in the Krasnodar Region. The average plant height is 160–180 cm. The spadix is slightly conical and abundantly grained, characterized by good moisture transfer. Kernels are tooth-shaped and yellow. The average weight of 1000 kernels is 270 g. The Pioneer P 9578 hybrid has good tolerance to corn borer and flying soot and is resistant to blister and dusty smut and helminthosporiosis.

The soil type is meadow low-humus loamy clay with agrochemical properties for top soil (0–20 cm): humus 2.78%, pH_{KCl}—6.25, $N\text{-}NO_3$—16.7 mg kg^{-1}, $N\text{-}NH_4$—11.9 mg kg^{-1}, P_2O_5—52.0 mg kg^{-1}, K_2O—460 mg kg^{-1}, clay fraction content (<0.01 mm) 67%. The base saturation (V) is 94.4%, hydrolytic acidity (Ha) is 1.96 mg Eq/100 g, and cation exchange capacity (CEC) is 35.8 mg Eq/100 g.

Sowing was carried out with precision seeding drills with a seeding rate of 5 seeds per linear meter. The row spacing width is 70 cm and the seed depth is 10 cm. The plot area of the experiment is 168 m^2 in 4-fold replication. The placement of variants is systematic. The vegetation period from germination to ripening and harvesting lasted 143 days (19 April 2017 sowing, 6 May 2017 germination, 20 September 2017 harvesting). Harvesting was carried out in the phase of full ripeness of grain.

Pre-sowing soil fertilization was conducted with ammophos (EuroChem Group) (100 kg ha^{-1}). Then, maize plants were fertilized with ammonium nitrate (120 kg ha^{-1}) at the stage of 3–4 leaves and at the stage of 5–6 leaves (80 kg ha^{-1}) (Figures S2 and S3). Total fertilization rate NPK was 228 kg ha^{-1}. Before the second cultivation, herbicides were treated with Dianat 0.4 L ha^{-1} + Titus, 44 g ha^{-1} + PVA Trend 90, 250 mL ha^{-1}.

The experimental design consisted of four variants: C was the control variant without any fertilizers; AF was the variant treated with ammophos fertilizer; BMF1 was the variant with ammophos fertilizer with the addition of spores of *Bacillus velezensis* BS89 on a dry carrier (diatomite); and BMF2 was the variant with ammophos granules treated with a spore suspension of *Bacillus velezensis* BS89.

2.2. Chemical Analysis of Plants and Soil

Nitrogen content. Dried samples of maize were finely ground, and 0.1 g of samples was moved to the digestion tubes for digestion using H_2SO_4 and H_2O_2. Nitrogen was determined from plant filtrate using the Kjeldhal method [29]. The protein proportion was measured by multiplying N by a conversion factor of 6.25.

Phosphate content was determined according to state standard GOST 51420-99 [30]. Briefly, 0.2 g of raw leaves was mixed with sulfuric and nitric acids during heating. The aliquot part of the acidic solution was mixed with a molybdenum phosphate reagent and the absorption of the resulting yellow solution was measured at a wavelength of 430 nm on a BioMate 160 Spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA).

Potassium content. Dry plant tissues were digested with ultra-pure concentrated nitric acid. The concentrations of potassium were measured by an emission flamephotometer PFA-378 (Unico, Dayton, NV, USA) following [31]. Standard solutions of K were used as reference [31].

Starch content in maize grains was determined as follows [32]. Glucose extraction was prepared with addition of 2 mL 80% ethanol to 50 mg grain powder twice. After removing ethanol, the starch in pellets was decomposed by the addition of 4 mL $HClO_4$. The amount of soluble sugar was determined with the anthrone reagent using glucose as the standard. Absorbance (A) was measured at 630 nm on a BioMate 160 Spectrophotometerv (Thermo Fisher Scientific, Waltham, MA, USA). Starch content (SC) was calculated using the formula $SC = A \times 0.9/DW \times 100\%$.

Exchangeable N-NH_4 in soil. Preparation of soil samples and extraction of N-NH_4 was conducted as follows [33]. Exchangeable N-NH_4 in soil samples was determined according to GOST 26489-85 [34]. The essence of the method is the acquisition of a colored indophenol compound formed by the interaction of ammonium with hypochlorite and sodium salicylate in an alkaline medium and a subsequent photometry of the colored solution. The procedure was as follows: 2 cm^3 of filtrates and reference solutions were put into conical flasks. Then, 40 cm^3 of the working staining solution was added to the samples, after which 2 cm^3 of sodium hypochlorite solution with a mass fraction of 0.125% was added. After one hour, optical densities of the color solutions were determined with the use of the BioMate 160 Spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA) at a wavelength of 655 nm.

Exchangeable N-NO_3 was determined with the help of selective electrodes according to GOST 26951-86 [35]. Nitrates were detected in a salt suspension of a 1% solution of potassium alum [$Al_2(SO4)_3 \cdot K_2SO_4 \cdot 24H_2O$] at a ratio of soil: solution = 1:2.5.

2.3. Meteorological Data

Data on precipitation and air temperature were obtained from the Kuban estuarine hydrometeorological station "Temryuk". In the first half of the growing season (April–July), the air temperature was similar to the long-term average values or (in some periods) was lower than average ones. Starting from August and until the end of the growing season of maize plants, the air temperature exceeded its average long-term values. During the growing season, the amount of precipitation was only 87.3% of the long-term average. The precipitation was distributed extremely unevenly: 79.2% of the norm in April, 87.3% in May, 22.0% in August, 24.0% in September, 176.7 of the norm in May, and 115.4% of the norm in July (Figure S4).

2.4. Bacteria and Biomodified Mineral Fertilizers

The strain BS89 *Bacillus velezensis* was stored in a freezer (Sanyo, Japan) at −80 °C as cell suspensions from a single colony in 20% glycerol. Potato dextrose agar (PDA; Sigma Aldrich, St. Louis, MO, USA) plates were used for passages. Bacteria were cultured in liquid potato dextrose broth (PDB, Sigma Aldrich, St. Louis, MO, USA) for 2 days at 28 °C on a rotary shaker at 200 rpm to obtain a final concentration of the bacterial spore suspension of 1.5×10^9 cfu/mL. Biomodification of ammophos was conducted two different ways. BMF1 was the variant with ammophos fertilizer with the addition of spores of *B. velezensis* BS89 on a dry carrier (diatomite). Diatomite was ground to the particle size of 50 µm and sterilized in closed glass cans at 121 °C for 3 h. Aseptically in the clean bench, 20 mL of bacterial spore suspension with a cell number of 1.5×10^9 cfu/mL was added to 1000 g of sterile diatomite and thoroughly stirred for 5 min to obtain the final product with a cell number not less than 15×10^6 cfu/g. Five grams of the final product was used to treat 1000 g of ammophos in a glass flask to obtain a cell number not less than 50×10^3 cfu/g of granules. BMF2 was the variant with ammophos granules treated with a spore suspension of *B. velezensis* BS89. An amount of 100 mL of bacterial spore suspension 1.5×10^9 cfu/mL was added to the anticaking agent (conditioner and industrial oil), heated to 85 °C in a ratio of 2:1, and kept for one hour at 85 °C. Ammophos granules were treated with the prepared mixture of the bacterial suspension and anticaking agent (cells number 500×10^6 CFU/mL) at the rate of 3 mL per 1000 g to obtain a cell number of not less than 50×10^3 CFU/g of granules.

2.5. Extraction of DNA, PCR, and Sequencing

The sampling was carried out in early August before the maize harvest. DNA was extracted from 0.5 g of frozen soil using a commercial Quick-DNA Fecal/Soil Microbe Miniprep Kit (Zymo Research, Irvine, CA, USA). DNA was purified by electrophoresis in 1% agarose gel and then isolated from the gel by sorption on silicon oxide [36]. During construction and sequencing of amplicon libraries, a purified DNA preparation (10–15 ng

each time) was used as a matrix in the PCR reaction (temperature profile: 95 °C—30 s, 50 °C—30 s, 72 °C—30 s; a total of 30 cycles) with the addition of Encyclo polymerase (Evrogen, Russia) and universal primers to the variable V4 site of the 16S rRNA genes F515 (GTGCCAGCMGCCGGCGGTAA) and R806 (GGACTACVSGGGTATCTAAT) [37]. In addition, oligonucleotide identifiers for each probe and service sequences required for pyrosequencing according to the Roche (Switzerland) protocol were inserted into the primers. Sample preparation and sequencing were performed on a GSJunior instrument (Roche, Switzerland) according to the manufacturer's recommendations.

The sequenced sequences were analyzed using QIIME 1.8.0 software [38]. The following steps were performed during the analysis: separation of libraries by identifiers, sequencing quality check and filtering of nucleotide sequences, combining sequences into operational taxonomic units (OTUs) using the de novo method with a 97% similarity threshold, alignment of representative nucleotide sequences using Uclust, and construction of genetic distance matrix and phylogenetic tree using Fasttree. Taxonomic identification of OTUs was performed with the help of the RDP algorithm (http://rdp.cme.msu.edu/ (accessed on 6 June 2023) and the Greengenes database [39].

2.6. Systemic Assessment of the Rhizosphere Microbial Community

The microbial community was assessed with the help of ecological biodiversity indices: the Shannon index and the Simpson index, using Biodiversity calculator [40]. The shared OTUs for different agricultural treatments were determined using a Venn diagram [41,42]. The Venn diagram was constructed using an internet resource [43].

The functional components of bacterial communities were estimated using the Kyoto Encyclopedia of Genes and Genomes (KEGG) dataset [44] Principal coordinate analysis (PCoA) was conducted to determine the overall differences in community compositions and beta (β) diversity analysis was used to assess the richness and diversity of soil microbial diversity.

2.7. Statistical Analysis

The results are presented as the means of four replicates with standard error (SE). The data were analyzed using one-way and two-way analysis of variance (ANOVA) in Statistics v 10 (SPSS, Inc., Chicago, IL, USA). The means were separated using Duncan's multiple range tests. Overall differences in the composition of the communities were revealed with the help of principal coordinate analysis (PCoA). We also assessed the richness and diversity of the soil microbial diversity with the help of beta diversity analysis.

3. Results

3.1. Influence of Biomodified Fertilizers on the Growth Parameters and Quality of the Maize Crop

Presowing application of ammophos and both forms of biomodified ammophos contributed to a statistically significant ($p \leq 0.05$) increase in plant height (Figure S5). For instance, at the seedling stage, maize plants in the variants with ammophos and both forms of BMF were 38% and 54% higher than the control plants, respectively. This difference became less pronounced in the course of growth, and at the phase of full ripeness, fertilized plants were 6–9% higher than the control ones. No differences in the effect of BMF1 and BMF2 on plant growth compared with ammophos were observed.

In the seedling phase, plants from fertilized variants exceeded the control plants by 35.7–50.0% in dry weight (Figure S6). This indicates that they were not only higher but were also better developed. As the subsequent stages of ontogenesis progressed, the differences gradually decreased, remaining as significant as before. At the end of ontogenesis, their dry mass was 12.8–13.9% higher than in the control. Noticeable differences between plants that received biomodified fertilizers were noted only at the initial stages of ontogenesis—in the seedling phase—and amounted to 5.3–10.5% compared with ammophos. After this phase, the differences noted were insignificant.

Regarding the NPK content in the plants, it should be noted that a significant increase in the proportion of NPK after fertilizer application was observed at all the phases of vegetation, except the phase of full ripeness (Table 1). Application of fertilizers significantly increased the accumulation of nitrogen by 31–54% and increased the accumulation of phosphorus at the seedling, tassel emergence, and ear emergence stages by 50–85%. The greatest increase in nitrogen content by 54% and in phosphorus content by 85% occurred at the stage of tassel emergence. In general, the greatest levels of nitrogen, phosphorus, and potassium in plants were observed at the seedling stage. No differences in the effect of BMF1 and BMF2 on the NPK content in the plants as compared with ammophos have been observed.

Table 1. Dynamics of macro-element content in the maize plants (Pioneer P 9578 hybrid) at different vegetation stages.

Variants	Vegetation Stage			
	Seedlings	**Tassel Emergence**	**Ear Emergence**	**Full Ripeness**
	Nitrogen, % FW			
C	2.05 ± 0.15 b	1.01 ± 0.07 b	1.05 ± 0.07 c	0.66 ± 0.05 a
AF	2.87 ± 0.17 a	1.54 ± 0.09 a	1.24 ± 0.07 b	0.68 ± 0.06 a
BMF1	2.87 ± 0.15 a	1.55 ± 0.08 a	1.27 ± 0.09 ab	0.69 ± 0.06 a
BMF2	2.90 ± 0.15 a	1.56 ± 0.11 a	1.38 ± 0.10 a	0.67 ± 0.09 a
	Phosphorus, % FW			
C	0.35 ± 0.05 b	0.20 ± 0.04 b	0.14 ± 0.03 b	0.10 ± 0.03 a
AF	0.54 ± 0.04 a	0.35 ± 0.04 a	0.26 ± 0.03 a	0.12 ± 0.04 a
BMF1	0.56 ± 0.05 a	0.36 ± 0.04 a	0.27 ± 0.04 a	0.14 ± 0.02 a
BMF2	0.56 ± 0.05 a	0.37 ± 0.04 a	0.28 ± 0.03 a	0.14 ± 0.03 a
	Potassium, % FW			
C	2.98 ± 0.19 a	1.24 ± 0.07 b	0.75 ± 0.05 b	1.32 ± 0.08 a
AF	3.02 ± 0.17 a	1.34 ± 0.08 a	0.90 ± 0.05 a	1.34 ± 0.09 a
BMF1	3.06 ± 0.18 a	1.35 ± 0.08 a	0.89 ± 0.06 a	1.34 ± 0.11 a
BMF2	3.07 ± 0.20 a	1.36 ± 0.10 a	0.90 ± 0.06 a	1.35 ± 0.11 a

Notes: C—control plants (grown on fertilizer-free soil), AF—plants grown on soil fertilized with ammophos, BMF1—plants grown on soil fertilized with BMF1 (ammophos with spores of *Bacillus velezensis* BS89 on dry carrier), BMF2—plants grown on soil fertilized with BMF2 (ammophos granules treated with spore suspension of *Bacillus velezensis* BS89). All data are presented as the mean ± SE (standard error). Different letters indicate significant differences between treatments at $p < 0.05$ in the Duncan test.

The use of ammophos and two biomodified forms of ammophos (in combination with the application of potassium chloride at the rate of 100 kg/ha before sowing and fertilizing with ammonium nitrate at the rate of 150 kg/ha) affected the content of mineral nitrogen in the soil under maize cultivation (Table S1). In the tassel emergence phase, nitrate nitrogen in the soil was higher than in the control by 8.2–16.8%, in the ear emergence phase by 39.8–53.7%, and in the full ripeness phase by 64.0–84.0%, and ammonium nitrogen was higher by 29.5–45.9%, 9.6–27.4%, and 4.9–65.1%, respectively. It should be noted that biomodification significantly ($p \leq 0.05$) increased the nitrate nitrogen in the soil as compared with ammophos by 7.5–8% in the tassel emergence phase, by 9.3–9.9% in the ear emergence phase, and by 11.0–12.2% in the full ripeness phase (Table S1). The more pronounced effect of the biomodification of ammophos has been observed in content of ammonium nitrogen in the soil. So, biomodification significantly ($p \leq 0.05$) increased the ammonium nitrogen in the soil as compared with ammophos by 12.0–12.7% in the tassel emergence phase, by 15.0–16.2% in the ear emergence phase, and by 16.4% in the full ripeness phase. No significant differences in the effect of biomodification on phosphorus content in the soil have been observed.

A statistically significant positive effect of fertilizers on grain quality was observed in the experiment. Application of ammophos, BMF1, and BMF2 increased dry matter in the

grain by 13–14% (Figure S7). In the variants with ammophos, BMF1, and BMF2, the grain was formed with an increased content of nitrogen and phosphorus by 17–18% and 24–34%, respectively. The potassium content also increased, but the difference was not statistically significant (Figure S8).

Ammophos, BMF1, and BMF2 had a different effect on some parameters of maize plants such as ear length and the number of kernels in the row and in the ear (Table 2). For instance, BMF1 and BMF2 increased ear length by 33.3%, while ammophos increased ear length only by 22.2%. Ammophos increased the number of kernels in the row and in the ear by 18.6% and 14.3%, respectively, while BMF1 and BMF2 increased the number of kernels in the ear by 26.5–26.9% and 16.1–16.7%, respectively. The effect of fertilizers on the 1000 kernel weight also differed, but not statistically significantly; in general, it was increased by 12–15% when compared with the control.

Table 2. Growth parameters of maize plants (Pioneer P 9578 hybrid).

Variants	Number of Plants per m^2, pcs	Number of Maize Ears, pcs per Plant	Maize Ear Length, cm	Number of Kernels in a Row, pcs	Number of Kernels in an Ear, pcs	Mass of 1000 kernels, g
C	3.9 ± 0.6 b	1.0 ± 0.3 a	18 ± 0.4 c	26.4 ± 0.4 c	384 ± 16 c	289 ± 9 b
AF	4.2 ± 0.9 a	1.2 ± 0.4 a	22 ± 0.7 b	31.3 ± 0.8 b	439 ± 18 b	329 ± 11 a
BMF1	4.2 ± 0.9 a	1.3 ± 0.4 a	24 ± 1.1 a	33.4 ± 0.8 a	446 ± 20 a	338 ± 13 a
BMF2	4.2 ± 0.9 a	1.3 ± 0.4 a	24 ± 1.1 a	33.5 ± 0.9 a	448 ± 20 a	339 ± 11 a

Notes: C—control plants (grown on fertilizer-free soil), AF—plants grown on soil fertilized with ammophos, BMF1—plants grown on soil fertilized with BMF1 (ammophos with spores of *Bacillus velezensis* BS89 on dry carrier), BMF2—plants grown on soil fertilized with BMF2 (ammophos pellets treated with spore suspension of *Bacillus velezensis* BS89). All data are presented as the mean ± SE (standard error). Different letters indicate significant differences between treatments at $p < 0.05$ in Duncan the test.

Full provision of plants with elements of mineral nutrition by application of ammophos, BMF1, and BMF2 increased the yield of maize by 25.1–30.1 c/ha or 76.6–90.1% as compared with the control (Table 3). The use of two biomodified forms of ammophos allowed for a yield of 4.4–4.5 c/ha to be obtained, or 7.5–7.6% more than using the ammophos. In particular, the use of mineral fertilizers improved crop quality, increasing starch and protein content by 4–5% and 16–21%, respectively (Table 3).

Table 3. Crop yield parameters of maize plants (Pioneer P 9578 hybrid).

Variants	Yield, Centner ha^{-1}	Starch, %	Protein, %
C	53.4 ± 2.9 c	62.1 ± 1.3 b	8.7 ± 0.5 b
AF	59.0 ± 3.2 b	64.9 ± 1.5 a	10.1 ± 0.6 ab
BMF1	63.4 ± 3.5 a	64.4 ± 1.4 a	10.5 ± 0.7 a
BMF2	63.5 ± 3.4 a	64.4 ± 1.6 a	10.5 ± 0.6 a

Notes: C—control plants (grown on fertilizer-free soil), AF—plants grown on soil fertilized with ammophos, with BMF1 (ammophos with spores of *Bacillus velezensis* BS89 on dry carrier), BMF2—plants grown on soil fertilized with BMF2 (ammophos granules treated with spore suspension of *Bacillus velezensis* BS89). All data are presented as the mean ± SE (standard error). Different letters indicate significant differences between treatments at $p < 0.05$ in the Duncan test.

Our experiments demonstrated a positive effect and the effectiveness of the studied forms of biomodified fertilizers (BMF1 and BMF2). The yield and some parameters of maize plants such as ear length and the number of kernels in the row and in the ear significantly increased after their application as compared with the use of ammophos. This indicates that BMF1 and BMF2 can be considered as promising fertilizers for maize cultivation.

3.2. Analysis of Microbial Community Composition in the Maize Rhizosphere

An analysis of the uncultured microbiome showed the presence of forty-three phyla, including unidentified bacteria (Figure 1 and Figure S9, Table S2). It was demonstrated that the microbial community of all experimental variants studied was dominated by the phyla Actinobacteria (27.8–32.3%), Proteobacteria (15.1–20.0%), Archaea Crenarchaeota

(12.8–18.9), Chloroflexi (4.0–10.5%), Firmicutes (5.6–10.1), Acidobacteria (3.5–5.2), Verrucomicrobia (2.5–4.9), Bacteroidetes (1.9–4.0%), Planctomycetes (1.9–2.4%), and Gemmatimonadetes (1.0–1.5%).

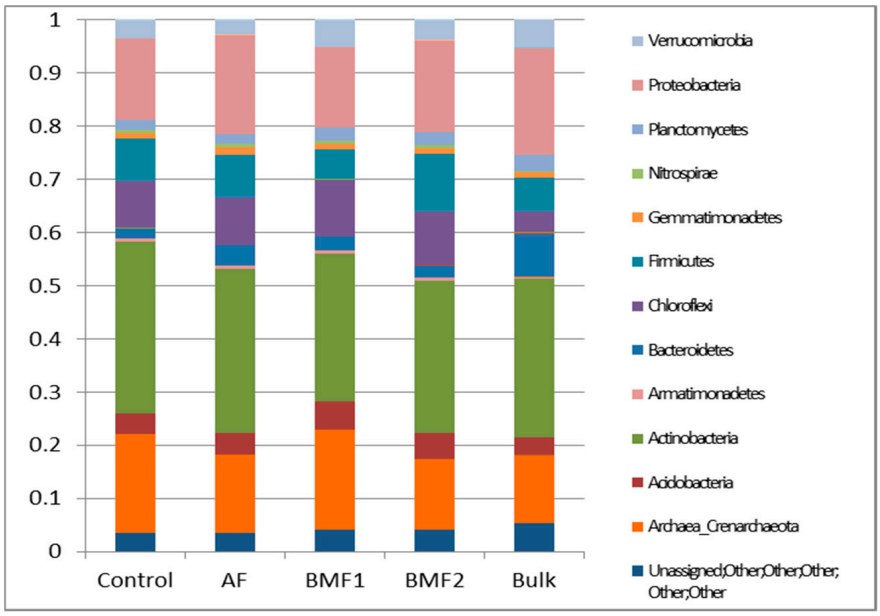

Figure 1. Taxonomic profile of the bacterial community of maize rhizosphere in variants with the use of various agricultural technologies. Notes: C—control plants (grown on fertilizer-free soil), AF—plants grown on soil fertilized with ammophos, BMF1—plants grown on soil fertilized with BMF1 (ammophos with spores of *Bacillus velezensis* BS89 on dry carrier), BMF2—plants grown on soil fertilized with BMF2 (ammophos granules treated with spore suspension of *Bacillus velezensis* BS89).

We found forty-three OTUs, including representatives of unidentified bacteria (see Figure S9 and Table S2). The microbial community of all experimental variants was dominated by Actinobacteria and Proteobacteria, as well as Archaea Crenarchaeota. Low-abundance phyla (1–10)% were presented by Chloroflexi, Firmicutes, Acidobacteria, Verrucomicrobia, Bacteroidetes, Planctomycetes, and Gemmatimonadetes (Table 4).

Table 4. Composition of bacterial community of maize (Pioneer P 9578 hybrid) rhizosphere after application of different fertilizers and in bulk soil.

Variant	Control	AF	BMF1	BMF2	Bulk Soil
Dominant phyla (>10%)					
Actinobacteria	32.3 ± 2.1 a	30.9 ± 3.0 a	27.7 ± 2.7 a	28.5 ± 1.7 a	29.7 ± 3.9 a
Archaea Crenarchaeota	18.5 ± 2.1 a	14.8 ± 2.2 ab	18.9 ± 2.1 a	13.3 ± 5.7 b	12.8 ± 1.3 b
Proteobacteria	15.3 ± 1.2 b	18.6 ± 4.6 ab	15.1 ± 1.3 b	17.2 ± 1.8 ab	20.0 ± 2.6 a
Low-abundance phyla (1–10%)					
Chloroflexi	8.7 ± 0.8 a	8.9 ± 0.2 a	10.5 ± 0.7 a	10.1 ± 0.8 a	4.0 ± 1.9 b
Firmicutes	7.8 ± 1.8 b	7.5 ± 1.0 b	5.6 ± 1.8 b	10.6 ± 2.3 a	6.0 ± 0.6 b
Acidobacteria	4.0 ± 0.7 a	4.0 ± 0.8 a	5.3 ± 1.6 a	5.0 ± 1.1 a	3.5 ± 0.4 a
Verrucomicrobia	3.3 ± 1.0 ab	2.5 ± 0.2 b	4.8 ± 1.3 a	3.6 ± 0.2 ab	5.0 ± 1.7 a
Bacteroidetes	1.9 ± 0.6 b	3.8 ± 2.6 b	2.7 ± 2.1 b	2.4 ± 0.8 b	8.2 ± 4.7 a
Planctomycetes	1.8 ± 0.4 b	1.9 ± 0.3 b	2.3 ± 0.2 ab	2.4 ± 1.0 ab	3.0 ± 1.0 a
Gemmatimonadetes	1.0 ± 0.2 a	1.4 ± 0.4 a	1.0 ± 0.1 a	1.0 ± 0.3a	1.1 ± 0.4 a
Other	5.4 ± 0.8b	5.7 ± 0.3b	6.1 ± 0.5ab	5.9 ± 0.5 ab	6.7 ± 0.6 a
Total	100	100	100	100	100

Notes: C—control plants (grown on fertilizer-free soil), B—bulk soil, AF—plants grown on soil fertilized with ammophos, with BMF1 (ammophos with spores of *Bacillus velezensis* BS89 on dry carrier), BMF2—plants grown on soil fertilized with BMF2 (ammophos granules treated with spore suspension of *Bacillus velezensis* BS89). All data are presented as the mean ± SE (standard error). Different letters indicate significant differences between treatments at $p < 0.05$ in the Duncan test.

Analysis of the beta diversity of the microbial community showed that the bulk soil formed a well-separated cluster and that rhizosphere communities were grouped into separate clusters (Figure 2). The control variant without fertilizers had the most isolated position. The grouping was confirmed by correlation coefficients, which were 0.98–0.99 for all variants with maize cultivation and 0.90 for bulk soil (Table 5).

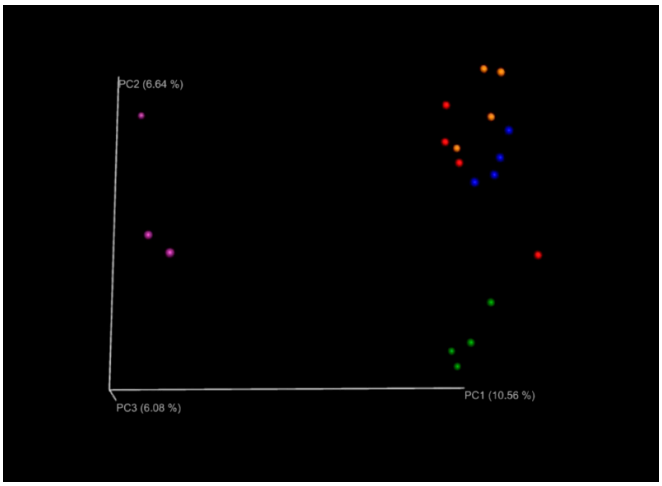

Figure 2. Beta diversity of rhizosphere communities of maize plants in experimental variants with application of ammophos, BMF1, and BMF2. Variants: bulk soil—lilac circles, control—green circles, ammophos—red circles, BMF1—blue circles, BMF2—orange circles. C—control plants (grown on fertilizer-free soil), AF—plants grown on soil fertilized with ammophos, BMF1—plants grown on soil fertilized with BMF1 (ammophos with spores of *Bacillus velezensis* BS89 on dry carrier), BMF2—plants grown on soil fertilized with BMF2 (ammophos granules treated with spore suspension of *Bacillus velezensis* BS89).

Table 5. Correlation coefficients between experimental variants.

Variants	Correlation Coefficients
C-BMF1	0.99
AF-BMF2	0.99
C-BMF2	0.98
C-AF	0.98
AF-BMF1	0.97
BMF1-BMF2	0.98
BMF1-B	0.91
BMF2-B	0.89
AF-B	0.90
C-B	0.90

Notes: B—bulk soil, C—control plants (grown on fertilizer-free soil), AF—plants grown on soil fertilized with ammophos, BMF1 (ammophos with spores of *Bacillus velezensis* BS89 on dry carrier), BMF2—plants grown on soil fertilized with BMF2 (ammophos granules treated with spore suspension of *Bacillus velezensis* BS89).

In the microbiome of the bulk soil, the abundance of representatives of the phyla Bacteroidetes and Planctomycetes was 4.3 and 1.7 times higher than that in the control variant of the rhizosphere soil. On the contrary, the abundance of representatives of the phylum Chloroflexi was two times higher in the rhizosphere soil (Table 4). The highest number (6.7%) of unidentified taxonomic phyla was also registered in the variant with the bulk soil. Based on the biodiversity indices, it was shown that the use of ammophos, BMF 1, and BMF2 did not show statistically significant changes in the number of dominant types of microbial communities compared to the control variant (Figure 3).

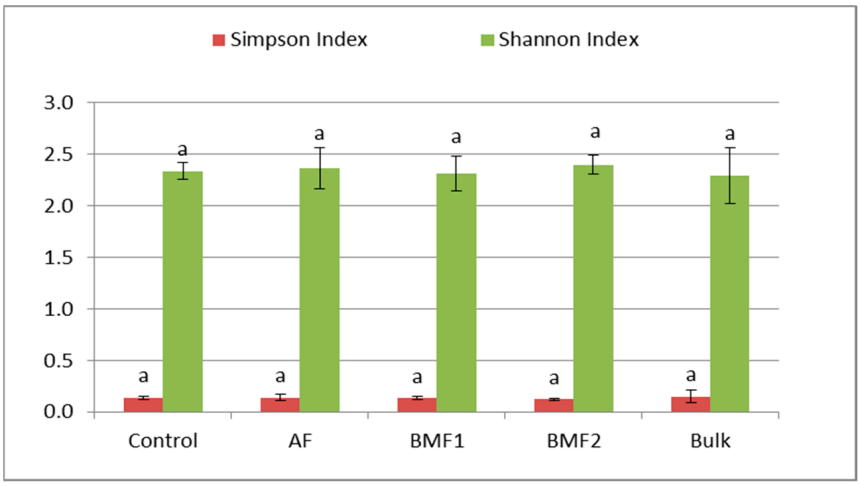

Figure 3. Biodiversity indices of the microbiome (calculated for forty-three phyla). Notes: C—control plants (grown on fertilizer-free soil), B—bulk soil, AF—plants grown on soil fertilized with ammophos, BMF1—plants grown on soil fertilized BMF1 (ammophos with spores of *Bacillus velezensis* BS89 on dry carrier), BMF2—plants grown on soil fertilized with BMF2 (ammophos granules treated with spore suspension of *Bacillus velezensis* BS89). All data are presented as the mean ± SE (standard error). Different letters indicate significant differences between treatments at $p < 0.05$ in the Duncan test.

The Venn diagram shows that the variant with BMF2 forms the most diverse rhizosphere community, with fifty-one unique bacterial OTUs (Figure 4). The diagram also shows that the variants with rhizosphere microbiomes have the greatest similarity, sharing eighty-three OTUs.

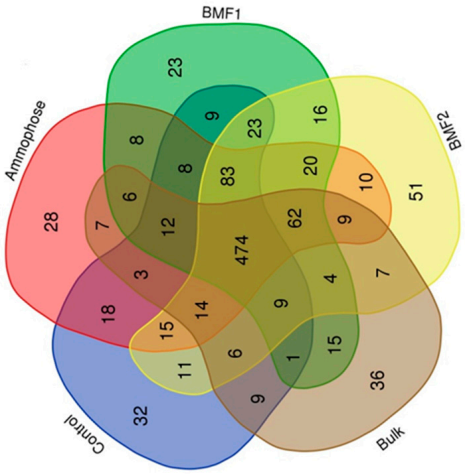

Figure 4. Venn diagram of rhizosphere microbiomes under the maize cultivation and the bulk soil. Notes: C—control plants (grown on fertilizer-free soil), B—bulk soil, AF—plants grown on soil fertilized with ammophos, BMF1—plants grown on soil fertilized with BMF1 (ammophos with spores of *Bacillus velezensis* BS89 on dry carrier), BMF2—plants grown on soil fertilized with BMF2 (ammophos granules treated with spore suspension of *Bacillus velezensis* BS89.

A curious feature of the soil was the presence of significant numbers of archaebacteria, most of which belonged to the phylum Crenarchaeota (from 12.8 to 18.9%). Minority components of soil communities (0.1–1%) were represented by the phyla Nitrospirae, Armatimonadetes, Cyanobacteria, and Tenericutes as well as some incompletely identified bacteria. Representatives of the phyla Chlamydiae, Chlorobi, Elusimicrobia, Fibrobacteres, and some others were present in insignificant amounts (less than 0.1%). An overview of the taxonomic profile of the microbial communities from the maize rhizosphere is shown

in Figures 1 and 5. When comparing the control and the variants with the application of ammophos, BMF1, and BMF2, no statistically significant changes in microbial communities at the phyla level were found. However, an increase in the abundance of phylum Chloroflexi was recorded in the variant with BMF1 as compared to the control; in the variant with BMF2, an increase in abundance of phyla Proteobacteria and Chloroflexi as well as a decrease in abundance of phylum Actinobacteria was noted as compared to the control.

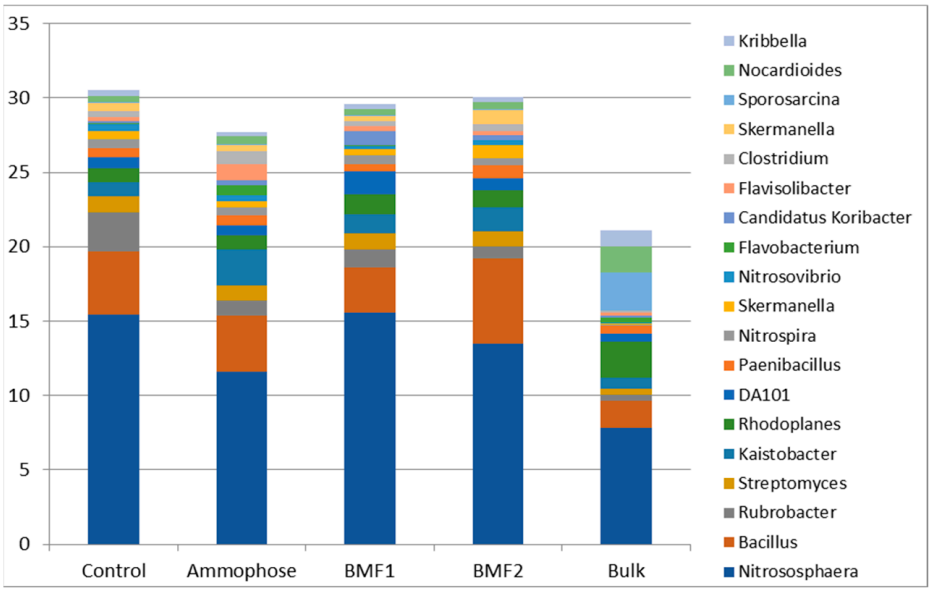

Figure 5. Taxonomic profile of the bacterial communities of maize rhizosphere grouped from the identified genera of microorganisms and ranked by abundance. Notes: C—control plants (grown on fertilizer-free soil), B—bulk soil, AF—plants grown on soil fertilized with ammophos, BMF1—plants grown on soil fertilized with BMF1 (ammophos with spores of *Bacillus velezensis* BS89 on dry carrier), BMF2—plants grown on soil fertilized with BMF2 (ammophos granules treated with spore suspension of *Bacillus velezensis* BS89).

In all experimental variants except the bulk soil, archaeans from phylum Nitrososphaera dominated, and their abundance ranged from 11.6 to 15.6% (groups with an abundance of more than 10% were considered as dominant) (Figure 5). In the variant with the bulk soil, no dominant genera of microorganisms were revealed, but Nitrososphaera showed a maximum abundance (7.8%) among the genera with a low abundance. Representatives of the genera *Sporosarcina*, *Rhodoplanes*, *Bacillus*, *Nocardioides*, and *Kribbella* were noted among the genera with a low abundance in the variant with the bulk soil. In the control variant, the genera with a low abundance were presented by *Bacillus* (4.2%), *Rubrobacter* (2.6%), and *Streptomyces* (1.04%). The same genera were noted in other variants with maize growing using different forms of fertilizers.

Figure 5 shows profiles of microbial communities of soils of the experimental variants grouped from the identified genera of archaeans and bacteria and ranked by abundance. The genus *Nitrososphaera* had the highest relative abundance. The application of ammophos reduced the abundance of bacteria of the genus *Rubrobacter* and increased the proportion of the genera *Flavisolibacter*, *Kaistobacter*, and *Clostridium* as compared with the other variants. The highest proportion of bacteria of the genus *Bacillus* was noted in the variant with BMF2. An increase in the abundance of bacteria of the genera *DA101* and *Candidatus Koribacter* was noted in the variant with BMF1. Table 6 shows the main functional groups of bacteria in the experimental variants as identified with the help of KEGG classification.

Table 6. KEGG functional classification statistics of soil bacteria in the rhizosphere of maize hybrid Pioneer.

Pathways	Control	Ammophos	BMF1	BMF2	Bulk
Nitrate assimilation	0.094 ± 0.019 b	0.086 ± 0.044 b	0.109 ± 0.172 b	0.128 ± 0.048 b	0.278 ± 0.108 a
Sulfate-sulfur assimilation	1.539 ± 0.206 b	1.664 ± 0.446 b	1.389 ± 0.218 b	1.896 ± 0.202 a	0.882 ± 0.059 c
Anoxygenic photosynthesis	0.590 ± 0.072 a	0.605 ± 0.334 a	0.421 ± 0.197 a	0.955 ± 0.238 a	0.194 ± 0.043 b
Methanogen	0.024 ± 0.011 a	0.008 ± 0.003 a	0.015 ± 0.012 a	0.029 ± 0.022 a	0

Note: All data are presented as the mean ± SE (standard error). Different letters in the same row indicate significant differences between treatments at $p < 0.05$ in the Duncan test. Results are presented as a percentage.

Nitrate-assimilating bacteria had the greatest abundance in the bulk soil and the lowest abundance in the variant with ammophos and in the control (Table 6). Sulfate-reducing bacteria were best represented in the variants with maize cultivation, especially in the variant with the application of BMF2. It was also shown (Table 6) that bacteria involved in the processes of methanogenesis completing the anaerobic destruction of organic matter were presented in the variants with the cultivation of maize plants. No methanogenic bacteria were noted in the bulk soil. The abundance of sulfate-reducing bacteria correlated with that of anoxygenic phototrophic bacteria.

When assessing the minority microbiome community of the experimental variants, we looked at the quantitative change in the bacterial diversity (Figures S9–S14). Ammophos had an inhibitory effect on some bacterial species, reducing the presence of some of them 6-29-fold as compared with the control variant. An increase (6.3-fold) was noted only for bacteria from the phylum Acidobacteria as compared with the control (Figure S10). Similarly, BMF1 reduced the abundance of some bacterial phyla in the minority community, 5-18-fold as compared with the control (Figure S11). In the variant with BMF2, a greater growth as compared to the control can be noted for the following identified genera from the minority community: *Vermamoeba*, *Ramlibacter*, *Methylobacter*, *Microbispora*, *Microlunatus*, *Conexibacter*, *Planomicrobium*, and *Parachlamydia* (Figure S12). However, BMF1 had a positive effect on the growth of four bacterial phyla, increasing their presence 5-9.2-fold as compared with ammophos (Figure S13). The bacteria positively affected by BMF1 belonged to the phyla Actinobacteria, Verrucomicrobia, Chloroflexi, and Planctomycetes.

In contrast to ammophos and BMF1, BMF2 had a significant effect on the minority community of the rhizosphere microbiome (Figures S14 and S15). The effect was registered for twenty-nine bacterial genera, which showed a 5–292-fold increase in abundance. The following increases in abundance were registered: 164-fold for c__Betaproteobacteria IS-44, 72-fold for f__Pseudomonadaceae, 292-fold for g__Tatlockia, 71-fold for c__Anaerolineae, 197-fold for o__Acidimicrobiales koll13, and 69-fold c__Gemmatimonadetes C114.

4. Discussion

In our study, the application of biomodified mineral fertilizers resulted in the significant increase in the yield and some parameters of maize plants such as ear length and number of kernels in the row. The yield was increased by 7.5–7.6%, ear length by 9%, and number of kernels in the row by 6.7–7%, as compared with ammophos. The same effect has been observed by other researchers. It has been shown before that the biomodified fertilizer BOZ4, containing Zn-solubilizing bacteria *Bacillus* sp. AZ6, increased maize yield by up to 25% as compared to the control [23]. Inoculation with *Azospirillum brasilense* increased maize yield by up to 9.7% in the field [45]. The increase in maize yield was accompanied by a 4% increase in protein accumulation in the grain. Recently, it was reported that after the application of biomodified nitrogen fertilizers, the accumulation of 15N in the barley plants increased by 2–5%, its incorporation in the soil decreased, and gaseous losses were decreased by 7 % as compared with the use of the usual forms of fertilizers [17]. In our study, biomodified fertilizers increased the available nitrogen and phosphorus in the soil and their uptake by plants, resulting in an increase in maize plant biomass, grain protein content, and plant yield. We suppose that this effect can be explained by the peculiarities

of the *Bacillus velezensis* BS89 strain. It was also shown that application of *Bacillus velezensis* BS89 on strawberries in three-year field trials demonstrated the same effect as the application of nitrogen fertilizers. Moreover, application of PGPR strain BS89 alone increased the yield of strawberries by 6.7–36.4% for cv. Rusich and 7.5–19.3% for cv. Troitskaya [46]. This was mainly associated with the plant-growth-promoting activity of *Bacillus velezensis* BS89, which was able to produce a high amount of IAA—494.1 μg/mL.

Thus, it is shown that the use of biomodified mineral fertilizers can have a significant impact on crop yields and improve product quality. In this regard, the question arises of what effect biomodified mineral fertilizers have on the microbiome of the rhizosphere of plants. It is known that inoculation causes some changes in the physiological activity of the rhizosphere microbiome. For instance, the abundance of microaerophilic nitrogen-fixing bacteria increased when maize plants were inoculated with the endophytic diazotrophic bacterium *Herbaspirillum seropedicae* strain HRC 54 [47]. Inoculation of wheat plants with non-nitrogen-fixing bacteria *Bacillus subtilis* V2 also resulted in an increase in the abundance of nitrogen-fixing rhizosphere microflora [48]. Plant roots releasing secondary metabolites and signaling molecules selectively attract various microorganisms from the surrounding soil [49]. It is known that carbohydrates make up 70% of maize root exudates [47]. Exudation of sugars and polyols by maize roots increased when nitrogen fertilizers were applied, with an increase in the abundance of bacteria involved in the nitrogen cycle in the rhizosphere [47]. So, root exudation can increase biological activity and rhizosphere microbiome abundance [49]. We demonstrated that the bacterial community of the experimental soil variants was represented by the phyla Actinobacteria, Proteobacteria, Chloroflexi, Firmicutes, Acidobacteria, Verrucomicrobia, Bacteroidetes, Planctomycetes, and Gemmatimonadetes. These phyla are known to be dominant and common microorganisms in soil communities [50]. Representatives of some other phyla were noted in the minority soil communities [51]. Another dominant group in the soil community which we observed were bacteria from the phylum Crenarchaeota. Although bacteria from the phylum Crenarchaeota are abundant and ubiquitous microorganisms, their metabolism is still studied incompletely [50]. Bacteria from the phylum Crenarchaeota are chemoautotrophic nitrificators [52], assimilators of amino acids [53] and carbon [54], and are also involved in methane metabolism [55].

Our study confirmed the observations that root exudates, depending on plant genotype, form the plant rhizosphere with its unique microbial community [56,57]. The experimental variants with the rhizosphere soil showed the greatest similarity in the composition of the microbiome, indicating the recruiting role of the maize plant in the formation of the rhizosphere microbiome. The microbiome of the bulk soil differed from that of the rhizosphere soil. The differences were mainly expressed in the minority composition of the microbial community, with the dominant and common bacteria being represented by the same phyla. The variant with the bulk soil was characterized by a higher abundance of the phyla Proteobacteria and Bacteroidetes and a lower abundance of the phylum Chloroflexi compared with the other variants with maize plants.

It was demonstrated that biomodified fertilizers BMF1 and BMF2 did not significantly affect the composition of dominant microbial communities of the maize rhizosphere. However, we observed differences between BMF1 and BMF2 for low-abundance bacterial groups and in minority communities of the rhizosphere microbiome. We demonstrated that biomodified fertilizers increased the abundance of representatives of the phyla Verrucomicrobia, Chloroflexi, Planctomycetes, Proteobacteria, Firmicutes, and Chlamydiae as compared with the variant with ammophos. BMF2 significantly increased the abundance of identified bacterial genera *Vermamoeba*, *Ramlibacter*, *Methylobacter*, *Microbispora*, *Microlunatus*, *Conexibacter*, *Planomicrobium*, and *Parachlamydia* as compared with the control. This increase was probably due to phytohormones produced by bacteria. BMF1, on the contrary, reduced the relative abundance of some genera as compared to the control, except for representatives of the families Thermogemmatisporaceae and Acidobacteriaceae.

The influence of the agricultural technology and the cultivated crop enhances the intensity of both the production and degradation of organic matter, which can be inferred from the quantitative assessment of microbial communities in the rhizosphere of plants. Inoculation with PGPR promotes the intensification of various physiological processes of the microbiome by means of increasing the microbial mass [58], leading to the accumulation or the decomposition of organic matter through their interactions with the resident microbiome [59] and mobilization of nutrients by the plants [17,47].

5. Conclusions

The application of biomodified mineral fertilizers resulted in a significant increase in the yield and some parameters of maize plants such as ear length and number of kernels in the row. The yield was increased by 7.57.6%, ear length by 9%, and number of kernels in the row by 6.7–7%, as compared with ammophos. Biomodified fertilizers stimulate growth and development of the plants by producing physiologically active substances and by stimulating the maize microbiome. The application of BMF1 and BMF2 increased the amount of nitrogen accessible to plants in the soil (both in the nitrate and in the ammonium forms), indicating an intensification of nitrification and ammonification processes.

We showed that maize formed a unique rhizosphere microbiome, differing from that of the bulk soil. The microbiome of the rhizosphere was characterized by a higher abundance of bacteria from the phylum Chloroflexi as compared with the bulk soil. On the contrary, the proportion of Proteobacteria and Bacteroidetes in the rhizosphere microbiome decreased. It was demonstrated that biomodified fertilizers BMF1 and BMF2 did not significantly affect the composition of dominant microbial communities of the maize rhizosphere, which was confirmed by the ecological indices of biodiversity: the Shannon index and the Simpson index. The application of biomodified fertilizers BMF1 and BMF2 considerably increased the abundance of bacteria representing the minority of the community, namely, those from the phyla Verrucomicrobia, Chloroflexi, Planctomycetes, Proteobacteria, Firmicutes, and Chlamydiae as compared with the use of ammophos. Thus, the application of biomodified mineral fertilizers is a promising agronomic and ecological strategy for boosting maize yield and the quality of grain under field conditions.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agronomy13071855/s1, Figure S1: Coordinates of the experimental field of experience 45.209483, 38.300953. Figure S2: Growth of maize (*Zea mays* L.) Pioneer 9578 hybrid in phase 5-6 leaves (27 May 2017). Figure S3: Growth of maize (Pioneer hybrid 9578) in flowering stage (16 July 2017). Figure S4: Meteorological data for the year of the field experiment (2017): A—air temperature, B—precipitation. Figure S5: Plant height of maize plants (Pioneer P 9578 hybrid) at seedling, tassel emergence, ear emergence and full ripeness vegetation stages. Figure S6: Dynamics of dry matter content in maize plants (Pioneer P 9578 hybrid) at seedling, tassel emergence, ear emergence and full ripeness vegetation stages. Figure S7: Dry matter content of maize (Pioneer P 9578 hybrid) kernels. Figure S8: NPK content of maize (Pioneer P 9578 hybrid) kernels. Figure S9: Taxonomic profile maize (Pioneer hybrid 9578) rhizosphere bacterial community when using various agricultural technologies. 1-4 control (C), 5-8 ammophos addition (AF),9-12 ammophos with spores of *Bacillus velezensis* BS89 on dry carrier (BMF1); ammophos granules treated with a spore suspension of *Bacillus velezensis* BS89 (BMF2). Figure S10: Changes in the composition of the minor microbial community in the control and ammophos (AF) experimental variants. Figure S11: Changes in the composition of the minor microbial community in the control and BMF1 experimental variants. Figure S12: Changes in the composition of the minor microbial community in the control and BMF2 experimental variants. Figure S13: Changes in the composition of the minor microbial community in the BMF1 and ammophos experimental variants. Figure S14: Changes in the composition of the minor microbial community in the BMF2 and ammophos experimental variants. Figure S15: Changes in the composition of the minor microbial community in the BMF1 and BMF2 experimental variants. Table S1: Effects of different fertilizers on the contents of available $N\text{-}NO_3$, $N\text{-}NH_4$ and P_2O_5 in rhizosphere soil of maize (Pioneer hybrid 9578) under different vegetative stages. Table S2: The

number of representatives of the main phyla of the microbiome of the rhizosphere of maize and bulk soil under different variants of fertilization.

Author Contributions: Conceptualization, V.K.C. and V.N.P.; methodology, E.P.C. and E.E.A.; software V.N.P., P.S.F. and N.I.V.; validation, V.K.C. and S.N.K.; formal analysis, V.N.P. and P.S.F.; investigation, E.P.C., O.V.K., M.E.B. and E.E.A.; resources, V.K.C., O.V.K. and M.E.B.; data curation, V.N.P. and E.E.A.; writing—original draft preparation, V.N.P. and P.S.F.; writing—review and editing, V.K.C., P.S.F. and V.N.P.; visualization, V.N.P. and P.S.F.; supervision, V.K.C.; project administration, V.K.C.; funding acquisition, V.K.C., E.P.C. and V.N.P. All authors have read and agreed to the published version of the manuscript.

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