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Root and Rhizosphere Microbiome of Tomato Plants Grown in the Open Field in the South of West Siberia under Mineral Fertilization

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Abstract: Agricultural practices can affect root-associated microbiota, but the effect of fertilization is still poorly examined. The aim of this study was to obtain 16S and ITS metagenomic profiles of tomato rhizosphere and root endosphere under mineral (NPK) fertilization in the open field experiment in the south of West Siberia. We found 6 bacterial and 3 fungal phyla in the roots and 24 bacterial and 16 fungal phyla in the rhizosphere. *Proteobacteria* and *Actinobacteria* together contributed 90% of the total number of sequence reads in roots and 50% in the rhizosphere, whereas *Ascomycota* ultimately prevailed in OTUs' richness and abundance in both biotopes. Fertilization changed the relative abundance of 32 bacterial and 14 fungal OTUs in the rhizosphere and of 7 bacterial and 3 fungal OTUs in roots. The revealed root bacteriobiome response to conventional mineral NPK fertilization by the dominant taxa at the high taxonomic level (class) illustrates well the role of NPK-changed plant metabolism in shaping endophytic microbiota and hence fertilization potential in enhancing plant growth-promoting microorganisms and mitigating plant pathogens. Using fertilization rate gradient in further research may bring a more detailed understanding of how to modify and even fine-tune root-associated microbiomes in order to enhance crops' health and yields.

Keywords: bacteriobiome; mycobiome; Phaeozem; microplot experiment; mineral fertilizer

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1. Introduction

Agricultural practices can affect soil microbiota, but how such practices, and in particular, fertilization, can affect rhizosphere and root endospheric microbiota in different agricultural contexts is still poorly studied.

Plants, like other eucaryotic organisms, harbor a plethora of microorganisms inside their bodies. A complicated network of diverse above- and below-ground interactions between plants, environment, and microbes determine the establishment of microbial assemblages in plants [1]. This microbiota may be beneficial, harmful, or neutral for the host's growth and development. The importance of plant-associated microorganisms cannot be overestimated in all types of ecosystems, from natural to agricultural and technogenic, as well as in all kinds of artificially constructed plant-growing environments, as they increase plant resilience, improve plant nutrition, enhance stress tolerance and defense and, consequently, sustain plant growth and production [2] and enable more sustainable agriculture [3]. Pathogenic microbial endophytes can cause serious diseases, sometimes devastating natural or agricultural ecosystems. However, the endophytic microbiome has been poorly investigated even in agriculturally important crops, and researchers still have to work hard to obtain a better insight into "the black box of ecological and evolutionary interactions across phytobiomes" [4], as currently there is very little knowledge on plant–endophyte interactions and mechanisms shaping microbial assemblages in plants [5]. Fertilization treatments were shown to clearly influence

the endophytic community structure of potato, for example, [6], and a study with wheat found no effect of mineral N and P fertilization on either bacterial community diversity or bacterial phyla abundance in the rhizosphere soil [7]. Thus, there exists a lack of information on how fertilizers act on the plant-associated microbial communities, not only endophytic but rhizosphere ones as well [8].

Tomato (*Licopersicon esculentum* L.) is globally one of the most important agricultural crops, with annual production reaching 187 mln tons in 2020 [9] and steadily increasing during the last years. Mineral fertilization, for many years, has been one of the most common fertilization practices for producing tomatoes both in protected conditions and in open fields. However, there is a lack of reports about the effect of conventional mineral fertilization on the microbiome of the rhizosphere and roots of tomato plants, although various organic and synthetic fertilizers seem to have been receiving research attention in this respect [10]. Some publications provide detailed information about the composition, diversity, and influential factors shaping the rhizospheric, phyllospheric, and endophytic bacterial communities of tomato plants [11] yet do not inform at all about the fertilizers used to stimulate plants' growth and production, briefly mentioning the fertilization was accomplished "following the recommendation of the seed company" [11] (p. 3), the latter also not being specified.

The aim of this research was to obtain 16S and ITS metagenomic profiles of tomato rhizosphere and root endosphere under mineral (NPK) fertilization in the open field experiment in the south of West Siberia, Russia.

2. Materials and Methods

2.1. Experimental Site

To study the microbiome in tomato roots, a microplot field experiment was carried out at the experimental station during the 2021 growing season in the forest-steppe zone in the south of West Siberia (54°58′ N, 83°13′ E). The climate of the region is classified as continental (Table S1) with a 119-day frost-free period. The experiment was conducted on the loamy arable soil classified as Luvic Greyzemic Phaeozem, according to the World Reference Base for Soil Resources [12]; or as gray agricultural soil, according to the Russian Soil Classification [13]. Phaeozem, together with Chernozem, are the most common soil types used in the region for agricultural production. The soil in our study has been in agricultural use for more than 40 years.

2.2. Experimental Setup

The microplot open field experiment was started at the beginning of the growing season (May 2021) and finished at the end of the growing season (September 2021). One cultivar of *Licopersicon esculentum* L. "Zyryanka", included in the Russian State Crop Register and recommended for the region, was used. Tomato seedlings were grown in cassettes in a peat substrate in the Central Siberian Botanical Garden SB RAS (Novosibirsk, Russia) and, at the age of 50 days, planted out on 12 May 2021 in the open field microplots at a density of one plant per 0.25 m^2. The experiment included two fertilization treatments: no fertilization (No) and mineral fertilization (NPK). Fertilizer application was started one week after planting out and continued throughout the season every fortnight. Mineral fertilizer (Nitrofoska, Agrosintez LLC, Kemerovo, Russia) was applied at the rate commonly recommended for vegetables in the region, i.e., equivalent to 60 kg N, 60 kg P, and 60 kg K per hectare during the first 2/3 of the growing season, and at the half of the rate during the last 1/3. Each treatment was performed in 5 randomized replicates, i.e., altogether, there were 15 microplots with 5 plants.

2.3. Soil Sampling and Chemical Analyses

The soil was sampled at the beginning of May 2021, prior to planting out the tomato seedlings, from the 0–15 cm layer in 3 individual replicates from the plot that were bulked together for further analyses. Soil organic carbon (SOC) content was estimated by dichro-

mate digestion; soil organic nitrogen content (STN) was determined by the Kjeldahl method; the content of soil available nutrients (NO_3^-, NH_4^+, P_2O_5) and pH (H_2O) were measured by standard techniques [14]. Briefly, nitrate content was determined potentiometrically in 0.1% $AlK(SO_4)_2$ solution (soil–solution ratio 1:5 *w*/*v*); ammonium content was measured colorimetrically in 2M KCl extracts (1:10 *w*/*v*). Available soil P was extracted by 0.03 M K_2SO_4 (1:5 *w*/*v*) and determined colorimetrically. Soil pH was measured by equilibrating 10 g of field-moist soil with 25 mL of deionized water. All analyses were performed in triplicates, and the data were expressed on the oven (105 °C) dry basis.

2.4. Plant Sampling and Analyses

The growing season in the open field in West Siberia is short, with cool nights already occurring in August, which can prevent the majority of fruits from ripening in situ. Therefore, tomato fruits were collected repeatedly during the growing season, starting at the end of July, as soon as they stopped increasing in size and reached technical maturity. At the end of the experiment, all consumable fruits were collected. Above- and below-ground phytomass was also determined at the end of the experiment. Fruits and phytomass produced by every plant were counted and weighed in fresh form. Roots for the microbiome analysis were washed in distilled water, sterilized by shaking in the peroxide solution, air-dried, and stored at −20 °C until DNA extraction. All plant components were collected from one plant, i.e., one plot.

2.5. DNA Extraction and Sequencing

Total DNA was extracted from 0.40 g of roots using the DNeasy PowerSoil Kit (Qiagen, Hilden, Germany) as per the manufacturer's instructions. The bead-beating was performed using TissueLyser II (Qiagen, Hilden, Germany) for 10 min at 30 Hz. No further purification of the DNA was needed. The quality of the DNA was assessed using agarose gel electrophoresis.

The V3–V4 region of the 16S rRNA gene and ITS2 region were amplified with the primer pairs 343F/806R and ITS3_KYO2/ITS4, respectively, combined with Illumina adapter sequences [15]. PCR amplification was performed as described earlier [16]. A total of 200 ng PCR product from each sample was pooled together and purified through MinElute Gel Extraction Kit (Qiagen, Hilden, Germany). The obtained amplicon libraries were sequenced with 2 × 300 bp paired-ends reagents on MiSeq (Illumina, CA, USA) in the SB RAS Genomics Core Facility (ICBFM SB RAS, Novosibirsk, Russia). The read data reported in this study were submitted to the NCBI Short Read Archive under bioproject accession number PRJNA887478.

2.6. Bioinformatic Analysis

Raw sequences were analyzed with Usearch v.11.0.667 using the UPARSE pipeline [17], which included the merging of paired read s, read quality filtering (-fastq_maxee_rate 0.005), length trimming (remove less 350 nt), merging of identical reads (dereplication), discarding singleton reads, removing chimeras, and operational taxonomic unit (OTU) clustering using the UPARSE-OTU algorithm. The OTU sequences were assigned a taxonomy using the SINTAX [18] and 16S RDP training set v.16 [19], or fungi ITS UNITE USEARCH/UTAX v.2018.11.18 [20] as a reference. The taxonomic structure of thus obtained sequence assemblages, i.e., a collection of different species at one site at one time [21], was estimated by the ratio of the number of taxon-specific sequence reads (archaeal and non-fungal sequences were removed from the data matrices) to the total number of sequence reads, i.e., by the relative abundance of taxa, expressed as a percentage. A taxon was considered dominant if its relative abundance was equal to or exceeding 1.0%.

The OTUs datasets were analyzed by individual rarefaction (graphs are not shown) with the help of the PAST software [22]: the numbers of bacterial and fungal OTUs detected, reaching a plateau with an increasing number of sequences, confirmed that the sampling effort was close to saturation for all samples, thus being enough to compare diversity [23].

2.7. Statistical Analyses

Statistical analyses (descriptive statistics and ANOVA) were performed by using Statistica v.13.3 a (TIBCO Software Inc., Palo Alto, CA, USA), and PERMANOVA was performed with PAST [22] software packages. OTUs-based α-diversity indices were calculated using PAST. Factor effects and mean differences in post hoc comparisons by Fisher's LSD test were considered statistically significant at the $p \leq 0.05$ level.

3. Results

3.1. Rhizisphere and Root Bacteriobiome

3.1.1. General Pattern

After quality filtering, chimera, and non-bacterial sequences removal, a total of 206 bacterial OTUs were identified at 97% sequence identity level in the roots and 3655 OTUs in the rhizosphere. In total, 6 bacterial phyla were found in the roots, whereas 24 phyla were detected in the rhizosphere.

Most of the total number of bacterial OTUs in roots belonged to the *Proteobacteria* phylum (85, or 41% of the OTU richness), with *Bacteroidetes* (49 OTUs) and *Actinobacteria* (42 OTUs) being the second and third most OTU-rich phyla, accounting for 23 and 20% of the total number of OTUs, respectively. The *Firmicutes* and Candidatus *Saccharibacteria* phyla contributed 10 and 11 OTUs, respectively, accounting for 5% of the total species richness in the study; *Deinococcus-Thermus* was represented by just 3 OTUs. As for the rhizosphere bacteriobiome, most of the OTUs also belonged to *Proteobacteria* (890, or 24% of the OTU richness), with *Firmicutes* (815 OTUs) and *Actinobacteria* (435 OTUs) being second and third most OTU-rich phyla, accounting for 22 and 12% of the total number of OTUs, respectively. Such phyla as *Acidobacteria*, *Chloroflexi*, and *Verrucomicrobia* were represented by 325, 153, and 97 OTUs, accounting for 9, 4, and 3% of the OTUs number, respectively.

As for the relative abundance, the ultimate dominants in both biomes were the *Proteobacteria* and *Actinobacteria* phyla, together contributing more than 50% of the total number of sequence reads in the rhizosphere and about 90% in the roots (Figure 1).

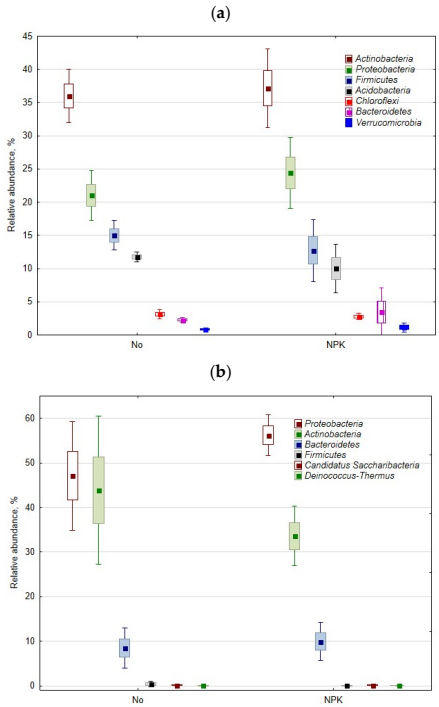

Figure 1. The relative abundance of bacterial phyla in the rhizosphere (only dominants (**a**)) and roots (all phyla (**b**)) of tomato plants grown under different fertilization on Phaeozem in the open field in the south of West Siberia. Squares denote means, boxes denote standard errors, and whiskers denote standard deviations. A taxon was considered dominant if its relative abundance was ≥1.0%.

3.1.2. The Effect of Mineral Fertilization on the Rhizosphere and Root Bacteriobiome

Mineral fertilization slightly decreased the relative abundance of one of the minor dominants of the rhizosphere bacteriobiome (Table 1). Overall, though, 32 bacterial OTUs showed differential fertilization-related abundance at $p \leq 0.05$ significance level, and 28 more OTUs showed differential abundance at the $0.05 \leq p \leq 0.10$ level; PERMANOVA, performed with bacterial OTUs, did not reveal statistically significant ($p = 0.58$) effect of fertilization on the rhizosphere bacteriobiome. The same was true for the root bacteriobiome at the OTU level ($p = 0.26$, PERMANOVA). The relative abundance of bacterial phyla did not show statistically significant changes; however, two of the dominant classes, namely *Alphaproteobacteria* and *Flavobacteriia* (of *Bacteroidetes*), demonstrated NPK-related changes. At the lower taxonomic levels, these changes translated to the changes in *Alphaproteobacteria/Sphingomonadales/Sphingomonadaceae/Sphingomonas* and *Flavobacteriia/Flavobacteriales/ Weeksellaceae/Chryseobacterium/Chryseobacterium* sp., increasing their relative abundance. The dominant actinobacterial taxa showed a tendency to decrease in roots under fertilization, but one of the dominant OTUs, attributed to *Actinobacteria/Actinobacteria/Micrococcales/Microbacteriaceae/ Rathayibacter/Rathayibacter* sp., was twice as abundant in the roots of the fertilized plants as compared with the non-fertilized ones (Tables 1 and 2). Seven OTUs, including four dominant ones, were differentially abundant in roots at the $p \leq 0.05$ significance level, with eight more OTUs at the $0.05 \leq p \leq 0.10$ level.

Table 1. The relative abundance (%, mean ± standard deviation) of the dominant bacterial taxa in the rhizosphere and roots of tomato plants, grown under different fertilization on Phaeozem in the open field in the south of West Siberia (a taxon was considered dominant if its relative abundance was ≥1.0%).

Taxon	Rhizosphere		Roots	
	No	NPK	No	NPK
Class level				
Actinobacteria	13.8 ± 3.4	18.5 ± 8.3	43.8 ± 16.6	33.6 ± 6.7
Alphaproteobacteria	11.5 ± 2.6	13.8 ± 3.4	28.6 ± 6.8 a [1]	41.9 ± 6.3 b
Thermoleophilia	8.4 ± 0.9	7.0 ± 2.9	0.03 ± 0.04	0.01 ± 0.01
Bacilli	7.8 ±1.1	7.3 ± 2.2	0.3 ± 0.6	0.0 ± 0.1
Acidimicrobiia	7.8 ± 1.0	6.8 ± 1.9	n.d.[2]	n.d.
un. [3] *Actinobacteria*	5.9 ± 1.0	4.9 ± 2.3	0.1 ± 0.1	0.0 ± 0.0 [2]
Clostridia	5.7 ± 1.1	4.3 ± 1.8	0.1 ± 0.1	0.0 ± 0.0
Acidobacteria_Gp6	5.4 ± 0.9	4.1 ± 1.6	n.d.	n.d.
Gammaproteobacteria	1.9 ± 0.6	3.0 ± 1.5	9.0 ± 5.4	7.6 ± 4.2
Betaproteobacteria	4.7 ± 1.0	4.5 ± 1.3	9.4 ± 8.2	6.7 ± 1.7
Deltaproteobacteria	2.6 ± 0.4	2.7 ± 0.7	n.d.	n.d.
Anaerolineae	1.3 ± 0.5	0.8 ± 0.6	n.d.	n.d.
Caldilineae	1.2 ± 0.2	0.9 ± 0.3	n.d.	n.d.
Cytophagia	0.9 ± 0.2	1.1 ± 0.7	6.9 ± 3.8	5.7 ± 3.3
Flavobacteriia	0.4 ± 0.1	1.6 ± 2.8	1.3 ± 1.0 a	3.6 ± 1.4 b
Order level				
Micrococcales	4.9 ± 4.0	7.4 ± 6.0	40.8 ± 16.6	31.8 ± 6.1
Rhizobiales	8.8 ± 2.2	9.5 ± 2.6	16.8 ± 7.2	24.0 ± 4.0
Bacillales	7.8 ± 1.1	7.3 ± 2.2	0.6 ± 0.3	0.04 ± 0.07
Acidimicrobiales	7.8 ± 1.0	6.8 ± 1.9	n.d.	n.d.
Gaiellales	6.0 ± 0.9	4.6 ± 1.9	n.d.	n.d.
Acidobacteria_Gp6	5.4 ± 0.9	4.1± 1.6	n.d.	n.d.
Clostridiales	5.2 ± 1.0	3.9 ± 1.5	0.00 ± 0.01	0.00 ± 0.00
Sphingomonadales	0.3 ± 0.2	0.6 ± 0.4	11.2 ± 1.0 a	17.4 ± 3.9 b
Burkholderiales	1.7 ± 0.4	1.6 ± 0.5	9.4 ± 8.2	6.7 ± 1.7

Table 1. *Cont.*

Taxon	Rhizosphere		Roots	
	No	**NPK**	**No**	**NPK**
Pseudomonadales	0.1 ± 0.2	0.1 ± 0.2	8.1 ± 5.1	7.3 ± 4.3
Cytophagales	0.9 ± 0.2	1.1 ± 0.7	6.9 ± 3.8	5.7 ± 3.3
Kineosporiales	n.d.	n.d.	2.1 ± 1.5	1.1 ± 0.6
Flavobacteriales	0.4 ± 0.1	1.6 ± 2.8	1.3 ± 1.0 a	3.6 ± 1.4 b
Family level				
Gaiellaceae	6.0 ± 0.9	4.6 ± 1.9	n.d.	n.d.
Acidobacteria_Gp6	5.4 ± 0.9	4.1 ± 1.6	n.d.	n.d.
un. *Rhizobiales*	5.0 ± 0.9	4.7 ± 1.0	n.d.	n.d.
Ilumatobacteraceae	4.3 ± 0.7	3.5 ± 1.1	n.d.	n.d.
Micrococcaceae	3.6 ± 3.4	3.7 ± 3.6	0.3 ± 0.3	0.0 ± 0.0
Hyphomicrobiaceae	2.9 ± 0.7	2.4 ± 0.5	n.d.	n.d.
Acidobacteria_Gp16	2.2 ± 0.7	2.5 ± 1.1	n.d.	n.d.
Iamiaceae	2.2 ± 0.2	2.0 ± 0.5	n.d.	n.d.
Planococcaceae	1.7 ± 0.4	1.6 ± 0.5	n.d.	n.d.
Bacillaceae1	1.6 ± 0.1	1.3 ± 0.5	n.d.	n.d.
Nocardioidaceae	1.6 ± 1.4	4.9 ± 4.8	0.1 ± 0.2	0.03 ± 0.03
Caldilineaceae	1.2 ± 0.2	0.9 ± 0.3	n.d.	n.d.
Paenibacillaceae1	1.0 ± 0.1	1.0 ± 0.4	0.03 ± 0.04	0.01 ± 0.01
Clostridiaceae1	1.0 ± 0.2	0.8 ± 0.4	n.d.	n.d.
Rhodobacteraceae	1.0 ± 0.2	1.9 ± 1.1	0.2 ± 0.2	0.1 ± 0.1
Microbacteriaceae	0.5 ± 0.3	1.6 ± 1.9	40.0 ± 17.1	31.6 ± 6.2
Sphingomonadaceae	0.2 ± 0.1	0.4 ± 0.2	11.2 ± 1.0 a	17.1 ± 3.5 b
Methylobacteriaceae	0.1 ± 0.2	0.1 ± 0.3	8.9 ± 4.0	13.4 ± 5.0
Pseudomonadaceae	0.1 ± 0.2	0.1 ± 0.2	8.1 ± 5.1	7.3 ± 4.3
Oxalobacteraceae	0.04 ± 0.03	0.10 ± 0.11	6.7 ± 7.4	3.5 ± 0.7
Hymenobacteraceae	0.2 ± 0.1	0.1 ± 0.1	6.4 ± 3.4	5.1 ± 3.1
Rhizobiaceae	n.d.	n.d.	4.5 ± 1.8	4.4 ± 1.9
Aurantimonadaceae	n.d.	n.d.	3.2 ± 2.1	5.9 ± 3.2
Comamonadaceae	n.d.	n.d.	2.7 ± 1.4	3.2 ± 1.2
Kineosporiaceae	n.d.	n.d.	2.1 ± 1.5	1.1 ± 0.6
Weeksellaceae	0.01 ± 0.01	0.01 ± 0.00	1.3 ± 1.0 a	3.6 ± 1.4 b
Genus level				
Gaiella	6.0 ± 0.9	4.6 ± 1.9	n.d.	n.d.
Acidobacteria_Gp6	5.8 ± 1.0	4.3 ± 1.7	n.d.	n.d.
un. Rhizobiales	5.0 ± 0.9	4.7 ± 1.0	0.1 ± 0.2	0.0 ± 0.0
Clavibacter	n.d.	n.d.	36.3 ±18.8	23.9 ± 6.2
Sphingomonas	0.1 ± 0.1	0.2 ± 0.1	11.1 ± 1.0 a	17.1 ± 3.4 b
Methylobacterium	n.d.	n.d.	8.9 ± 4.0	13.4 ± 5.0
Pseudomonas	0.1 ± 0.2	0.1 ± 0.2	8.1 ± 5.1	7.3 ± 4.3
Massilia	n.d.	n.d.	6.6 ± 7.4	3.5 ± 0.6
Hymenobacter	n.d.	n.d.	6.4 ± 3.4	5.1 ± 3.1
Agrobacterium	n.d.	n.d.	4.5 ± 1.8	4.4 ± 1.9
Aureimonas	n.d.	n.d.	3.2 ± 2.1	5.9 ± 3.2
Rathayibacter	n.d.	n.d.	2.9 ± 1.8 a	6.3 ± 3.5 b
un. [2] *Comamonadaceae*	0.2 ± 0.1	0.2 ± 0.2	2.7 ± 1.3	3.2 ± 1.2
Kineococcus	n.d.	n.d.	1.9 ± 1.5	1.0 ± 1.2
Chryseobacterium	n.d.	n.d.	1.3 ± 1.0 a	3.6 ± 1.4 b

[1] Values in rows, followed by different letters, differ significantly ($p \leq 0.05$, Fisher's LSD test). The absence of letters indicates no difference. [2] n.d. stands for "not detected", meaning that not a single sequence was found, whereas zero values mean that the respective sequences were found, but in numbers much less than [3] un. stands for unclassified.

Table 2. The relative abundance (%, mean ± standard deviation) of the dominant bacterial OTUs in the rhizosphere and roots of tomato plants grown under different fertilization on Phaeozem in the open field in the south of West Siberia (An OTU was considered dominant if its relative abundance was ≥1.0%).

No.	OTU	Rhizosphere		Roots	
		No	NPK	No	NPK
4	*Clavibacter* sp.	n.d. [1]	n.d.	36.3 ± 18.8	23.9 ± 6.2
7	*Pseudarthrobacter*	3.6 ± 3.4	3.6 ± 3.5	n.d. [1]	n.d.
9	*Sphingomonas* sp.	n.d.	n.d.	7.2 ± 3.0	7.4 ± 3.5
15	*Methylobacterium* sp.	n.d.	n.d.	8.4 ± 4.4	13.1 ± 4.9
16	*Aureimonas* sp.	n.d.	n.d.	2.9 ± 2.2	4.9 ± 3.8
26	*Pseudomonas* sp.	n.d.	n.d.	6.5 ± 3.1	6.5 ± 3.2
27	un. [2] *Rhizobiales*	3.6 ± 0.6	3.0 ± 0.6	n.d.	n.d.
28	*Agrobacterium* sp.	n.d.	n.d.	4.5 ± 1.8	4.4 ± 1.9
29	*Rathayibacter* sp.	n.d.	n.d.	2.9 ± 1.8 a [3]	6.3 ± 3.5 b
31	*Chryseobacterium* sp.	n.d.	n.d.	1.2 ±1.0 a	3.6 ± 1.4 b
43	*Sphingomonas* sp.	n.d.	n.d.	0.9 ± 1.0 a	3.4 ± 1.5 b
56	un. *Hyphomicrobiaceae*	1.5 ± 0.3	1.3 ± 0.3	n.d.	n.d.
57	un. *Actinobacteria*	2.2 ± 0.5	1.5 ± 0.9	n.d.	n.d.
58	*Sphingomonas* sp.	n.d.	n.d.	1.6 ± 1.0 a	4.3 ± 1.2 b
60	*un. Comamonadaceae*	n.d.	n.d.	2.1 ±1.0	1.8 ± 0.5
65	*Kineococcus* sp.	n.d.	n.d.	1.9 ± 1.5	1.0 ± 0.5
68	*Massilia* sp.	n.d.	n.d.	6.6 ± 7.4	3.5 ± 0.6
84	un. *Acidobacteria_Gp6*	1.6 ± 0.3 b	1.1 ± 0.2 a	n.d.	n.d.
88	un. *Gaiella*	1.7 ± 0.2	1.3 ± 0.8	n.d.	n.d.
95	un. *Nocardioides*	0.5 ± 0.4	1.5 ± 1.4	n.d.	n.d.
101	*Hymenobacter* sp.	n.d.	n.d.	5.0 ± 4.0	3.7 ± 2.9
107	un. *Actinobacteria*	1.3 ± 0.3	0.9 ± 0.4	n.d.	n.d.
122	un. *Nocardioides*	0.4 ± 0.4	1.2 ± 1.1	n.d.	n.d.
188	un. *Desertimonas*	1.0 ± 0.2	0.7 ± 0.3	n.d.	n.d.
1027	*Gaiella occulta*	1.0 ± 0.2	0.8 ± 0.2	n.d.	n.d.
1099	*Hymenobacter* sp.	n.d.	n.d.	1.0 ± 1.0	0.6 ± 0.4
3987	*Sphingomonas* sp.	n.d.	n.d.	1.0 ± 0.7	1.4 ± 0.9
6043	*Aureimonas* sp.	n.d.	n.d.	0.3 ± 0.3	1.0 ± 1.3

[1] n.d. stands for "not detected". [2] un. stands for unclassified. [3] Values in rows, followed by different letters, differ significantly ($p \leq 0.05$, Fisher's LSD test). The absence of letters indicates no difference.

The number of dominant OTUs, i.e., OTUs contributing more than 1% to the total number of sequence reads, was 17 in the root bacteriobiome and 11 in the rhizosphere (Table 2).

3.1.3. Alpha-Biodiversity in the Rhizosphere and Root Bacteriobiome

Neither rhizosphere nor root bacteriobiome α-biodiversity indices changed under fertilization, their values being very similar (Table 3) between the treatments. As expected, all indices indicated much greater bacteriobiome biodiversity in the rhizosphere. Noteworthy, though, is the fact that the *p*-value for the comparison of root bacteriobiome species evenness was 0.059 (Fisher's LSD test), i.e., very close to the 0.05 threshold of statistical significance.

Table 3. Alpha-biodiversity indices (mean ± standard deviation) of the rhizosphere and root bacteriobiome of tomato plants grown in the open field in the south of West Siberia.

Taxon	Rhizosphere		Roots	
	No	NPK	No	NPK
Richness	1338 ± 167	1381 ± 203	90 ± 47	76 ± 13
Chao-1	1909 ± 218	1923 ± 234	100 ± 42	84 ± 15
Simpson (1-D)	0.99 ± 0.00	0.99 ± 0.00	0.80 ± 0.13	0.89 ± 0.03
Shannon	6.0 ± 0.2	6.0 ± 0.2	2.4 ± 0.6	2.7 ± 0.1
Evenness	0.31 ± 0.00	0.29 ± 0.04	0.14 ± 0.03	0.21 ± 0.03
Equitability	0.84 ± 0.01	0.83 ± 0.02	0.55 ± 0.09	0.63± 0.02
Dominance (D)	0.01 ± 0.00	0.01 ± 0.00	0.20 ± 0.13	0.11 ± 0.03
Berger-Parker	0.05 ± 0.01	0.05 ± 0.02	0.36 ± 0.19	0.24 ± 0.06

3.2. Rhizosphere and Root Mycobiome

3.2.1. General Pattern

After quality filtering, chimera and all plant sequences removal and subsequent removal of non-fungal sequences (just five OTUs in the root mycobiome and 648 OTUs in the rhizosphere one), a total of 387 and 2718 fungal OTUs were identified at 97% sequence identity level in the root and rhizosphere mycobiomes, respectively. Altogether, 16 fungal phyla were detected: all of them in the rhizosphere and only 3 (namely, *Basidiomycota*, *Ascomycota,* and *Chytridiomycota*) in the roots. In both mycobiomes one cluster was attributed to *Fungi* but not classified below the domain level.

Most of the total number of fungal OTUs belonged to the *Ascomycota* phylum: 231, or 60% of the OTU richness, in the roots and 1159, or 43%, in the rhizosphere. In both mycobiomes, *Basidiomycota* was the second OTU-rich phylum with 150 OTUs, or 39% of the OTUs richness in the roots, and 438, or 16%, in the rhizosphere. In the root mycobiome, the other two clusters, i.e., *Chytridiomycota* and unclassified *Fungi*, together contributed six OTUs, being negligible in terms of OTUs richness. In the rhizosphere, *Chytridiomycota* and *Mortierellomycota* contributed respectively 5 and 2% of the total number of OTUs, the other 12 phyla together accounting for about one-third of the OTUs' richness.

As for the relative abundance (Figure 2), the *Ascomycota* phylum showed 40% in both treatments in the roots, whereas in the rhizosphere, the phylum accounted for 80% of the total number of sequence reads, as averaged over both treatments. *Basidiomycota* accounted for 5.8% as averaged over both treatments. The third-abundant *Zygomycota* contributed 4.4% in the rhizosphere mycobiome but was not detected at all in the root one. In the root mycobiome *Chytridiomycota* and unclassified *Fungi* were virtually non-present with their less than 0.01%, whereas in the rhizosphere, their contribution was 2% and 3.3%, respectively.

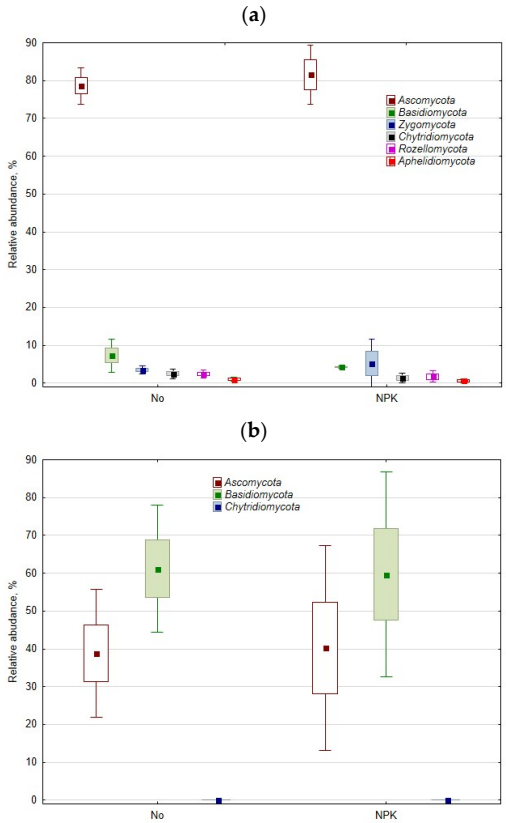

Figure 2. The relative abundance of fungal phyla in the rhizosphere (only dominants (**a**)) and roots (all phyla (**b**)) of tomato plants grown under different fertilization on Phaeozem in the open field in the south of West Siberia. Squares denote means, boxes denote standard errors, and whiskers denote standard deviations. A taxon was considered dominant if its relative abundance was ≥1.0%.

3.2.2. The Effect of Mineral Fertilization on the Rhizosphere and Root Mycobiome

In the rhizosphere mycobiome, fertilization-related differences were not revealed at the phylum, class, and order levels for the dominant taxa (Table 4). However, such differences were found at the lower taxonomic levels: three families (*Microascaceae, Plectosphaerellaceae,* and *Lasiosphaeriaceae)* showed decreased abundance, whereas two genera (*Plectosphaerella* and *Fusarium*) had increased abundance, which at the OTU level translated to the increased abundance of *Plectosphaerella plurivora/Plectosphaerella/Plectosphaerellaceae/Sordariomycetidae* _incertae sedis/*Sordariomycetes/Ascomycota* and *Fusarium domesticum/Nectriaceae/Hypocreales/Sordariomycetes/Ascomycota* (Table 5). Besides these two, twelve more minor or rare fungal OTUs showed NPK-related differential abundance changes; and PERMANOVA, performed with OTUs, revealed a statistically significant ($p = 0.04$) difference between the fertilization treatments.

Table 4. The relative abundance (%, mean ± standard deviation) of the dominant fungal taxa in the rhizosphere and roots of tomato plants, grown under different fertilization on Phaeozem in the open field in the south of West Siberia (a taxon was considered dominant if its relative abundance was ≥1.0%).

Taxon	Rhizosphere		Roots	
	No	NPK	No	NPK
Class level				
Tremellomycetes	3.0 ± 3.9	1.7 ± 0.7	55.6 ± 16.3	57.3 ± 26.7
Dothideomycetes	13.4 ± 7.9	20.1 ± 1.6	34.3 ± 17.8	30.3 ± 24.8
Microbotryomycetes	0.3 ± 0.2	0.3 ± 0.1	4.0 ± 3.0	1.6 ± 1.1
Cystobasidiomycetes	0.05 ± 0.03	0.09 ± 0.12	1.1 ± 1.6	0.3 ± 0.3
Leotiomycetes	8.2 ± 1.4	5.5 ± 3.1	0.1 ± 0.1	0.4 ±0.6
Sordariomycetes	42.6 ± 6.2	44.8 ± 7.1	0.1 ± 0.3	1.1 ± 2.4
Pezizomycetes	4.5 ± 1.3	3.6 ± 0.9	n.d. [1]	n.d.
Eurotiomycetes	3.0 ± 0.6	2.5 ± 0.9	0.003 ± 0.002	0.04 ± 0.04
Agaricomycetes	3.6 ± 3.5	2.0 ± 0.9	0.1 ± 0.1	0.02 ± 0.02
Aphelidiomycetes	1.0 ± 0.6	0.6 ± 0.5	n.d.	n.d.
Order level				
Tremellales	0.7 ± 0.5	0.8 ± 0.4	51.9 ± 14.8	53.4 ± 24.6
Pleosporales	4.0 ± 1.7	13.0 ± 11.2	21.3 ± 12.2	23.1 ± 24.0
Capnodiales	0.8 ± 1.8	1.8 ± 3.7	12.6 ± 8.3	6.9 ± 3.7
Cystofilobasidiales	0.3 ± 0.2	0.2 ± 0.1	3.7 ± 2.7	3.9 ± 4.2
Leucosporidiales	0.05 ± 0.08	0.01 ± 0.00	3.2 ± 3.0	1.1 ± 0.9
Cystobasidiomycetes_is	0.004 ± 0.003	0.01 ± 0.01	1.0 ± 1.6	0.2 ± 0.1
Helotiales	7.2 ± 1.2	4.3 ± 2.8	0.1 ± 0.1	0.4 ± 0.6
Glomerellales	0.5 ± 0.3	1.0 ± 0.3	0.0 ± 0.0 [1]	1.1 ± 2.4
Hypocreales	13.0 ± 2.2	19.1 ± 9.1	0.1 ± 0.3	0.001 ± 0.002
Microascales	15.7 ± 5.9	10.0 ± 5.7	n.d.	n.d.
Mortierellales	4.0 ± 1.2	5.1 ± 6.6	n.d.	n.d.
Pezizales	4.3 ± 1.4	3.5 ±0.9	n.d.	n.d.
Sordariales	4.4 ± 1.5	3.1 ±1.6	0.01 ± 0.00	0.01 ± 0.00
*Sordariomycetidae*_is [2]	5.2 ± 0.7	8.9 ±4.0	n.d.	n.d.
*Dothideomycetes*_is	4.4 ± 1.6	3.5 ± 2.2	n.d.	n.d.
Eurotiales	1.7 ± 0.7	1.7 ± 1.1	n.d.	n.d.
Agaricales	2.3 ± 3.4	0.5 ± 0.2	0.01 ± 0.01	0.01 ± 0.01
Coniochaetales	2.0 ± 0.4	1.4 ± 1.0	n.d.	n.d.
Onygenales	1.1 ± 0.5	0.6 ± 0.4	n.d.	n.d.
Family level				
Pleosporaceae	0.2 ± 0.1	1.8 ± 3.1	17.4 ± 12.0	20.7 ± 22.4
un. *Tremellales*	0.0 ± 0.0	0.0 ± 0.0	15.9 ± 7.6	23.1 ± 13.2
Tremellaceae	0.5 ± 0.4	0.5 ± 0.2	14.9 ± 17.7	8.0 ± 6.2
Bulleribasidiaceae	0.2 ± 0.3	0.3 ± 0.5	14.4 ± 10.1	17.7 ± 16.1

Table 4. *Cont.*

Taxon	Rhizosphere		Roots	
	No	**NPK**	**No**	**NPK**
Mycosphaerellaceae	0.8 ± 1.8	1.8 ± 3.7	12.6 ± 8.3	6.9 ± 3.7
*Tremellales*_is	0.0 ± 0.0	0.0 ± 0.0	6.4 ± 4.3	4.3 ± 5.8
Cystofilobasidiaceae	0.2 ± 0.2	0.1 ± 0.1	4.1 ± 2.7	3.6 ± 4.1
Leucosporidiaceae	0.0 ± 0.1	0.0 ± 0.0	3.6 ± 3.0	1.1 ± 0.9
Symmetrosporaceae	0.0 ± 0.0	0.0 ± 0.0	2.1 ± 1.6	0.1 ± 0.1
Phaeosphaeriaceae	0.1 ± 0.1	0.0 ± 0.0	0.9 ± 0.8	0.7 ± 0.5
Sclerotiniaceae	1.0 ± 0.5	0.8 ± 0.5	0.0 ± 0.0	0.0 ± 0.0
Microascaceae	10.2 ± 3.6	7.0 ± 3.5	n.d.	n.d.
Nectriaceae	6.2 ± 1.2	9.3 ± 5.2	0.03 ± 0.08	0.00 ± 0.00
Plectosphaerellaceae	5.5 ± 0.9 a [3]	9.9 ± 3.9 b	0.0 ± 0.0	1.1 ± 2.4
Mortierellaceae	3.9 ± 1.2	5.1 ± 6.6	n.d.	n.d.
Pseudeurotiaceae	2.9 ± 1.1	2.3 ±1.5	n.d.	n.d.
Psathyrellaceae	1.9 ± 3.4	0.3 ± 0.1	0.01 ± 0.01	0.01 ± 0.00
Ascodesmidaceae	1.9 ± 0.6	1.4 ± 0.9	n.d.	n.d.
Clavicipitaceae	1.7 ± 1.0	0.9 ± 0.7	0.00 ± 0.00	0.00 ± 0.00
Trichosporonaceae	1.6 ± 3.5	0.4 ± 0.5	n.d.	n.d.
Lasiosphaeriaceae	1.6 ± 0.5 b [3]	0.8 ± 0.5 a	n.d.	n.d.
Chaetomiaceae	1.6 ± 0.5	1.7 ± 0.6	0.01 ± 0.00	0.01 ± 0.00
Pyronemataceae	1.5 ± 0.9	0.9 ± 0.4	n.d.	n.d.
Aspergillaceae	0.7 ± 0.3	1.2 ± 1.2	n.d.	n.d.
Didymellaceae	1.0 ± 0.6	8.8 ± 11.9	2.1 ±1.7	1.0 ± 0.8
Trichocomaceae	1.0 ± 0.5	0.5 ± 0.4	n.d.	n.d.
Sclerotiniaceae	1.0 ± 0.5	0.8 ± 0.6	0.00 ± 0.00	0.00 ± 0.00
un. [4] *GS16*	1.0 ± 0.5	0.6 ± 0.5	n.d.	n.d.
Ascobolaceae	0.8 ± 0.4	1.0 ± 0.4	n.d.	n.d.
	Genus level			
Alternaria	0.01 ± 0.01	1.6 ± 3.2	17.3 ±12.0	20.7 ± 22.4
un. *Tremellales*	0.00 ± 0.00	0.00 ± 0.00	15.9 ± 7.6	23.1 ± 13.2
Cryptococcus	0.5 ± 0.4	0.5 ± 0.2	14.2 ± 17.9	7.4 ± 5.9
Vishniacozyma	0.2 ± 0.4	0.3 ± 0.5	13.9 ± 10.1	17.2 ± 16.4
Davidiella	0.8 ± 1.8	1.8 ± 3.7	12.4 ± 8.3	6.8 ± 3.7
Dioszegia	0.00 ± 0.00	0.00 ± 0.01	6.9 ± 4.7	4.8 ± 6.3
Cystofilobasidium	0.00 ± 0.00	0.00 ± 0.00	3.6 ± 2.7	3.6 ± 4.1
Leucosporidium	0.01 ± 0.01	0.01 ± 0.00	3.2 ± 3.0	1.1 ± 0.9
un. *Didymellaceae*	0.2 ± 0.2	0.4 ± 0.4	1.3 ± 1.6	0.7 ± 0.5
Botryotinia	1.0 ± 0.5	0.8 ± 0.6	0.0 ± 0.0	0.0 ± 0.0
Lectera	0.3 ± 0.2	0.3 ± 0.2	0.0 ± 0.0	1.1 ± 2.4
Wardomyces	5.2 ± 2.4	2.6 ± 2.5	n.d.	n.d.
Tetracladium	4.9 ± 1.4	2.6 ± 1.7	n.d.	n.d.
Plectosphaerella	4.0 ± 0.8 a	8.7 ± 4.0 b	n.d.	n.d.
Mortierella	4.0 ± 1.2	5.1 ± 6.6	n.d.	n.d.
Gibberella	3.7 ± 1.1	6.1 ± 4.6	0.0 ± 0.1	0.0 ± 0.0
Pseudogymnoascus	1.9 ± 1.1	1.7 ± 1.1	n.d.	n.d.
Metarhizium	1.6 ± 1.0	0.8 ± 0.6	n.d.	n.d.
Apiotrichum	1.6 ± 3.5	0.4 ± 0.5	n.d.	n.d.
Parasola	1.5 ± 3.3	0.0 ± 0.0	n.d.	n.d.
Cephaliophora	1.4 ± 0.5	1.1 ± 0.8	n.d.	n.d.
Gibellulopsis	1.1 ± 0.3	0.9 ± 0.2	n.d.	n.d.
Dokmaia	1.1 ± 0.6	1.8 ± 0.5	0.0 ± 0.1	0.0 ± 0.0
un. *GS16*	1.0 ± 0.6	0.6 ± 0.5	n.d.	n.d.
Didymella	0.5 ± 0.5	8.3 ± 12.0	0.2 ± 0.4	0.5 ± 0.5
Fusarium	0.7 ± 0.3 a	2.0 ± 1.1 b	0.000 ± 0.001	0.000 ± 0.001
Penicillium	0.6 ± 0.4	1.2 ± 1.2	n.d.	n.d.

Table 4. *Cont.*

Taxon	Rhizosphere		Roots	
	No	**NPK**	**No**	**NPK**
Emericellopsis	1.0 ± 0.9	0.8 ± 0.7	n.d.	n.d.
Ascobolus	1.0 ± 0.4	0.7 ± 0.3	n.d.	n.d.

[1] n.d. stands for not detected, and 0.0 values mean that there were some sequences detected, but their relative abundance was extremely low. [2] "_is" stands for incertae sedis. [3] Values in rows, followed by different letters, differ significantly (≤0.05, Fisher's LSD test). The absence of letters indicates no difference. [4] un. stands for unclassified.

Table 5. The relative abundance (%, mean ± standard deviation) of the dominant fungal OTUs in the rhizosphere and roots of tomato plants grown under different fertilization on Phaeozem in the open field in the south of West Siberia (an OTU was considered dominant if its relative abundance was ≥1.0%).

No.	OTU	Rhizosphere		Roots	
		No	**NPK**	**No**	**NPK**
1	un. [1] *Alternaria*	0.0 ± 0.0 [2]	1.5 ± 3.0	5.8 ± 12.1	19.7 ± 21.7
2	un. *Tremellales*	n.d. [2]	n.d.	11.9 ± 7.8	17.3 ± 12.5
3	un. *Ascomycota*	3.9 ± 7.0	1.1 ± 1.4	4.1 ± 2.0	8.4 ± 7.4
4	*Davidiella* sp.	0.8 ± 1.8	1.8 ± 3.7	12.4 ± 8.3	6.8 ± 3.7
7	*Vishniacozyma victoriae*	n.d. [2]	n.d.	6.4 ± 2.6	13.9 ± 13.8
8	*Cryptococcus* sp.	n.d.	n.d.	9.0 ± 19.1	0.3 ±0.6
10	*Dioszegia crocea*	n.d.	n.d.	6.2 ± 4.4	4.3 ± 5.8
13	un. *Tremellales*	n.d.	n.d.	3.9 ± 2.5	5.7 ± 4.1
14	*Phoma exigua*	0.5 ± 0.5	8.3 ± 11.9	n.d.	n.d.
15	*Gibberella* sp.	2.0 ± 0.7	2.2 ± 1.8	n.d.	n.d.
18	*Cystofilobasidium macerans*	n.d.	n.d.	2.6 ± 1.2	3.8 ± 1.7
19	*Plectosphaerella cucumerina*	2.5 ± 0.5	3.0 ± 0.4	n.d.	n.d.
21	*Mortierella minutissima*	3.1 ± 1.0	4.8 ± 6.7	n.d.	n.d.
24	*Leucosporidium* sp.	n.d.	n.d.	3.2 ± 2.9	1.0 ± 0.9
25	un. *Hypocreales*	2.1 ± 2.3	1.1 ±0.6	n.d.	n.d.
27	un. *Alternaria*	0.0 ± 0.0	0.1 ±0.2	1.3 ±1.4	0.9 ± 0.7
31	*Wardomyces inflatus*	5.1 ± 2.4	2.6 ± 2.5	n.d.	n.d.
33	*Tetracladium* sp.	1.8 ± 0.5	1.3 ± 1.0	n.d.	n.d.
34	un. *Microascaceae*	7.1 ± 2.6	4.4 ± 2.6	n.d.	n.d.
35	un. *Ascomycota*	3.9 ± 3.3	1.8 ± 0.7	n.d.	n.d.
39	*Cryptococcus chernovii*	n.d.	n.d.	2.1 ±1.7	1.7 ±2.0
40	*Cryptococcus festucosus*	n.d.	n.d.	0.5 ± 0.2	1.6 ± 1.8
46	*Dokmaia monthadangii*	1.0 ± 0.6	1.7 ± 0.5	n.d.	n.d.
47	*Tetracladium maxilliforme*	2.8 ± 1.2	1.1 ± 0.9	n.d.	n.d.
57	*Botryotinia* sp.	1.0 ± 0.5	0.8 ± 0.6	0.0 ± 0.0	0.0 ± 0.0
65	*Fusarium cerealis*	1.3 ± 0.6	1.5 ± 1.3	n.d.	n.d.
66	*Gibellulopsis nigrescens*	1.1 ± 0.3	0.9 ± 0.2	n.d.	n.d.
67	*Cephaliophora* sp.	1.4 ± 0.5	1.0 ± 0.7	n.d.	n.d.
70	*Metarhizium* sp.	1.4 ± 1.0	0.7 ± 0.5	n.d.	n.d.
76	un. *Dothideomycetes*	1.1 ± 0.5	1.1 ± 0.8	n.d.	n.d.
78	*Pseudogymnoascus* sp.	1.0 ± 0.3	1.1 ± 0.6	n.d.	n.d.
82	*Cryptococcus tephrensis*	0.0 ± 0.0	0.1 ±0.1	1.9 ± 2.2	2.3 ± 2.4
85	un. *Hypocreales*	0.0 ± 0.1	3.0 ± 5.6	n.d.	n.d.
89	*Ascobolus* sp.	0.7 ± 0.3	1.0 ± 0.4	n.d.	n.d.
90	un. *Coniochaetales*	1.0 ± 0.3	0.9 ± 0.7	n.d.	n.d.
101	*Apiotrichum dulcitum*	1.6 ± 3.5	0.4 ± 0.5	n.d.	n.d.
103	*Emericellopsis microspora*	0.8 ± 0.7	1.0 ± 0.9	n.d.	n.d.
118	*Symmetrospora coprosmae*	n.d.	n.d.	1.0 ± 1.6	0.2 ± 0.1
142	*Fusarium domesticum*	0.2 ± 0.2 a[3]	1.1 ± 1.1 b	n.d.	n.d.
152	*Lectera capsica*	0.3 ± 0.2	0.3 ±0.2	0.0 ± 0.0	1.1 ± 2.4
175	*Plectosphaerella plurivora*	1.4 ± 0.5 a	5.1 ± 3.7 b	n.d.	n.d.
190	*Parasola kuehneri*	1.5 ± 3.3	0.0 ± 0.0	n.d.	n.d.
643	*Vishniacozyma heimaeyensis*	0.2 ± 0.3	0.3 ± 0.5	7.4 ± 10.1	3.2 ± 2.5
1095	un. *Didymellaceae*	0.2 ± 0.2	0.4 ± 0.4	1.3 ± 1.5	0.7 ± 0.5
3321	*Cryptococcus magnus*	n.d.	n.d.	0.3 ± 0.1	1.1± 1.3

[1] un. stands for unclassified. [2] n.d. stands for no sequences detected, and 0.0 values mean that there were some sequences detected, but their relative abundance was extremely low [3] Values in rows, followed by different letters, differ significantly (≤0.05, Fisher's LSD test). The absence of letters indicates no difference.

The dominant OTUs numbered 22 (summarily 49% of the relative abundance) and 25 (56%) in No and NPK treatments, respectively, with a few variations between them, which resulted in the overall pool of prevailing OTUs totaling 31.

In the root mycobiome, there were no statistically significant differences in the relative abundance of the dominant taxa at all taxonomic levels due to a rather high variation within the NPK treatment (Tables 4 and 5), which was also confirmed by PERMANOVA (p = 0.70), performed with the entire set of root mycobiome OTUs. Overall, only three very rare OTUs, i.e., with maximal relative abundance of 0.004% among them, revealed NPK-related changes at the $p \leq 0.05$ level. The dominant OTUs numbered 16 in each treatment, totaling 20 OTUs and not showing any NPK-related differential abundance.

3.2.3. Alpha-Biodiversity in the Rhizosphere and Root Mycobiome

As for the α-biodiversity indices in the rhizosphere and root mycobiome, there were no statistically significant differences between the treatments (Table 6), although altogether, the indices showed a tendency for the α-biodiversity to decrease in the NPK treatment as compared to the other one. As expected, the root mycobiome was drastically less diverse than the rhizosphere one.

Table 6. Alpha-biodiversity indices (mean ± standard deviation) of the mycobiome in the tomato roots grown under different fertilization on Phaeozem in the open field in the south of West Siberia.

Taxon	Rhizosphere		Roots	
	No	NPK	No	NPK
Richness	928 ± 55	790 ± 174	171 ± 40	144 ± 34
Chao-1	1044 ± 73	909 ± 146	203 ±52	179 ± 40
Simpson (1-D)	0.97 ± 0.01	0.95 ± 0.04	0.85 ± 0.04	0.81 ±0.10
Shannon	4.8 ± 0.1	4.4 ±0.7	2.5 ± 0.2	2.3 ± 0.4
Evenness	0.13 ± 0.02	0.11 ± 0.04	0.08 ± 0.01	0.08 ±0.03
Equitability	0.70 ± 0.02	0.66 ± 0.09	0.49 ± 0.03	0.47 ±0.07
Dominance (D)	0.03 ± 0.01	0.04 ± 0.03	0.15 ± 0.04	0.19 ±0.10
Berger-Parker	0.10 ± 0.03	0.14 ±0.08	0.31 ± 0.09	0.34 ± 0.16

3.3. Tomato Production Properties

As for the biological and consumable tomato plant production, there were no statistically significant differences (Table 7), although, under the NPK treatment, the plants showed a tendency to produce better.

Table 7. Production characteristics of tomato plants grown under different fertilization on Phaeozem in the open field in the south of West Siberia (mean ± standard deviation).

	No	NPK	*p*-Value
Fruits, pcs/plant	30 ± 12	31 ± 16	0.773
Yield (Y), kg/plant	1.28 ± 0.48	1.65 ± 0.92	0.312
Average fruit mass, g	43 ± 10	49 ± 13	0,261
Aboveground phytomass [1] (A), g/plant	224 ± 173	340 ± 275	0.334
Belowground phytomass (B), g/plant	15.6 ± 7.2	27.8 ± 17.1	0.226
A/B	13.1 ± 6.1	11.1 ± 6.0	0.625
Total phytomass, g/plant	1.66 ± 0.67	2.24 ± 1.28	0.279

[1] Without fruit yield.

4. Discussion

4.1. Rhizosphere and Root Bacteriobiome

It is universally accepted that rhizosphere and roots harbor distinct bacterial assemblages, and our results showed the same. The strong prevalence of *Actinobacteria* and *Proteobacteria* in the rhizosphere soil and roots of tomato plants complies with the results of

other studies on tomato [3,24,25], although in the rhizosphere *Proteobacteria* abundance was two times lower in our study as compared, for instance, with [24]. The ultimate dominance of the *Proteobacteria* and *Actinobacteria* phyla in roots agrees with the results of López et al., 2020 [26] and other results reviewed by Bulgarelli et al. (2013) [1] and Trivedi et al. (2021) [3]. They concluded that most plant species harbor an enrichment of bacterial taxa belonging to the phyla *Proteobacteria*, *Bacteroidetes*, *Firmicutes,* and *Actinobacteria* and that the root endosphere shows an overwhelming dominance of bacteria belonging to the *Proteobacteria* phylum, also by culture-dependent methods [27]. Our finding that the *Acidobacteria* phylum, being one of the major dominants in the rhizosphere soil, was not even detected in the root endosphere agrees with the drastically decreased phylum's abundance in *Arabidopsis thaliana* roots as compared with the bulk or rhizosphere soil [28,29]. The fact that in our study, other minor dominants in the rhizosphere, i.e., *Chloroflexi* and *Verrucomicrobia*, like *Acidobacteria*, were not even detected in the roots, implies that these phyla are either "actively excluded by the host immune system, outcompeted by more successful root colonizers or metabolically unable to colonize" [28] (p. 90), the root endosphere. However, some studies reported the presence, albeit not prominent, of *Chloroflexi* and *Verrucomicrobia* sequence reads in tomato root endosphere bacteriobiome [30]. Such discrepancy implies either the failure of the plant immune system, lack of competition from other root colonizers, strains with some metabolic features facilitating colonization and establishment inside roots, or mere contamination of root endosphere by bacteria from the rhizoplane. It is noteworthy that the three phyla, i.e., *Acidobacteria*, *Chloroflexi,* and *Verrucomicrobia*, are common in similar soils of the region [31]. As for the *Bacteroidetes* phylum, in our study, it was a moderate dominant in the roots and a minor dominant in the rhizosphere, although in a recent study with tomatoes grown on a soil substrate in a greenhouse, *Bacteroidetes* was one of the major dominant members of the community in the roots [25]. As for *Firmicutes*, in our study, the phylum's representatives displayed negligible abundance in the roots, where the latter was drastically lower as compared with the rhizosphere, where the *Firmicutes* phylum ranked third in abundance (12–15%). Such a pattern, i.e., significantly decreased presence of *Firmicutes* in tomato roots as compared with the rhizosphere soil, was reported by Lee et al. (2019) [23]. This is in contrast with the results of some studies of culturable bacteria in tomato plants: the *Firmicutes* representatives, mainly belonging to the *Bacillus* genus, were prominent in roots [25,32].

There exists a general belief that a subset of rhizospheric microorganisms penetrates into the plant roots and colonizes the endosphere depending on the plant's innate immune system [1,28]. Our attempt to identify soil-type-specific OTUs within the root-inhabiting bacterial assemblages rendered extremely few ones: of the rhizosphere bacteriobiome totaling more than 3000 OTUs, only 15 OTUs, each with less than 1% abundance, were detected in the root bacteriobiome. Yet there is also a possibility that the bacteria, compatible with the endophytic lifestyle, might have entered inside the roots from the rhizosphere but were simply below the limit of detection in soil, but it seems unrealistic to spread this assumption for all bacteria detected in roots. Thus, many tomato root endophytes could have entered plants via other routes [28,29].

The finding of much greater α-biodiversity in the root endosphere was to be expected; on average OTUs' richness was 17 times greater in the rhizosphere samples, with 1360 OTUs per sample. This greater richness complies with a prominent (7% on average) share of the sequence reads, not attributed below the domain level, in the rhizosphere bacteriobiome and a negligible share in the root one.

Mineral fertilization for many years has been one of the most common fertilization practices for producing tomatoes both in protected conditions and in open fields. We showed that such fertilization can change the rhizosphere and root bacteriobiome taxonomic profile, albeit mostly affecting minor or rare taxa. We want to emphasize that we did not correct for multiple comparisons, mainly because our goal was to examine if simple NPK fertilization may bring about some microbiome changes and indicate putative taxa worth focusing attention on in further research.

Our finding that *Sphingomonas* sp./*Sphingomonadaceae*/*Sphingomonadales*/*Alphaproteobacteria* was 1.5 times more abundant in roots under mineral fertilization implies its role in promoting plants' performance under fertilization: for instance, some endophytic representatives of the genus, producing gibberellins and indole-acetic acid, were shown to promote tomato growth [33]. Some other representatives of the genus seem to be associated with tomato: novel *Sphingomonas* species was recently isolated from the soil of a tomato garden [34].

Our finding that a *Bacteroidetes* species, namely *Chryseobacterium* sp./*Weeksellaceae*/*Flavobacteriales*/*Flavobacteriia*, increased its abundance due to fertilization, suggests the beneficial effect of fertilization: for instance, a representative of the *Chryseobacterium* was reported to be able to act as a biocontrol agent and a bio-fertilizer [35], and flavobacterial genome was far more abundant in the rhizosphere microbiome of the tomato plants resistant to the soil-borne pathogen *Ralstonia solanacearum* as compared with that of the susceptible plant [36].

Notably, one of the root bacteriobiome OTUs, namely *Clavibacter* sp., being the ultimate dominant in the roots in our study, seemed to decrease its abundance due to fertilization, although the decrease was not statistically significant (Fisher's LSD test, $p = 0.19$). The finding that this OTU was not even detected in the rhizosphere bacteriobiome strongly suggests that this endophyte is not soil-derived, which implies another route for the bacterium to colonize and proliferate in the root endosphere: indeed, *C. michiganensis* subsp. *michiganensis* was shown to invade tomato fruits and seeds through multiple entry routes [37]. Additionally, the majority of healthy root bacteria could be tracked from the soil, and only a very small portion could be tracked from the soil for diseased samples, as it was shown by Dastogeer et al. (2022) [38]. It is highly likely that in our study, the detected *Clavibacter* OTU represented the infamous pathogen *C. michiganensis* subsp. *michiganensis*, causing often devastating bacterial canker [39,40]: we observed some specific, albeit weakly manifested, disease symptoms, i.e., unilateral leaf wilting, scarce stem canker, and bird's-eye lesions on fruit in our experimental plants, although fruit yield per plant was similar to the values reported earlier for tomato plants grown in the open field in the same region [41]. Strictly speaking, however, one cannot be fully sure about the pathogenic nature of the *Clavibacter* sp., first-ranked in the relative abundance in the root bacteriobiome in our study, since (a) the metagenome-based pathogen identification at the strain level cannot be achieved because of the challenges inherent to assigning reads to specific strains [42], and (b) based on comparative genomics and phylogenetic analyses several novel species within the genus *Clavibacter* were suggested [43], including nonpathogenic tomato-associated strains, belonging to the *C. michiganensis* clade [44]. Yet, we cannot help but note that our result about the *Clavibacter* prevalence in the root bacteriobiome and its apparent decrease due to fertilization suggests the possibility of improving plant performance by supplying with extra macronutrients and hence boosting plants' immunity and defense against pathogens. Besides that, as some of the species from the *Sphingomonas* genus have been noted to improve plant growth during stress conditions such as drought, salinity, and heavy metals in agricultural soil [45], in our study, it could also help the plants stressed by a pathogen. We should add that the incidence of the visual disease manifestations was rather low, and since we did not expect any such damage to be substantial, we did not record all such data in a reportable form; it seems that high variation in the growth and production characteristics of the fertilized plants might have resulted from the plants' differential response to the putative pathogen.

Our finding that mineral fertilization affected the relative abundance of only one dominant (a) representative of *Acidobacteria_Gp6*) and several dozens of minor or rare OTUs of the rhizosphere bacteriobiome suggests two things: (a) that, despite the relatively short growing period, stimulation of tomato plants growth and production by fertilization also somewhat changed their rhizodeposition profile, bringing shifts in bacterial populations attracted by the rhizodeposition; and (b) that under the experimental conditions of our study the key taxa of the rhizosphere bacteriobiome were rather stable, and microbiota fine-tuning was effected by minor or rare members.

The fact that we did not detect any archaea in tomato roots complies with the general view that archaea are less abundant and diverse in association with eukaryotic hosts [46]: the primers we used, albeit not specific for archaea, usually render from several to dozens of archaeal sequences, especially from the environments where they are usually present, such as soil.

4.2. Rhizosphere and Root Mycobiome

Our finding that tomato root and rhizosphere mycobiome was dominated by *Ascomycota* and *Basidiomycota* phyla once again confirms their role as major players in diverse environments, ranging from the soil and subsoil [31] to plants [2], from the deep-sea sediments [47] and water [48] to the air [49]. However, the fact that the root mycobiome in our study was almost exclusively composed of these two phyla does not seem common for endophytic fungal communities, for which more diverse phyla profiles were reported [50].

Increased fertilization abundance of *Plectosphaerella plurivora* in the rhizosphere mycobiome in our study might be due to the increased attractiveness of the fertilized plants' roots for the pathogenic strains of the fungus [51], as members of the *Plectosphaerella* genus can be found in various habitats, including plants [52] and soil, are pathogens [53], causing large losses in agriculture. However, some strains can be beneficial for plants, for instance, by attacking plant-parasitic nematodes [54]. Thus, it is difficult to speculate about a putative ecophysiological mechanism and significance of more abundant *P. plurivora* under mineral fertilization.

As for the NPK-increased abundance of *Fusarium domesticum*, its detection in the rhizosphere soil is rather unexpected, as *Fusarium domesticum* is usually found as a part of the specific cheese surface microbiota [55], and we did not manage to find any reports about the fungus detection in the soil in general and tomato rhizosphere soil in particular. The *Fusarium* genus is versatile, distributed worldwide in soil, aquatic and semiaquatic environments, stored grain, and natural products, and its members are mostly pathogenic [56].

This study showed that mineral NPK fertilization can shift mycobiome towards the enhanced presence of pathogenic fungi in the rhizosphere. However, the fact that in our study, these fungi were not even detected inside roots suggests that (a) the longevity of the growth/fertilization period was not enough for the fungi to colonize and establish themselves inside roots, or (b) they lack such ability, being common soil commensals.

The fact that tomato phytomass production characteristics showed high variation due to the mineral NPK fertilization thus decreasing the statistical significance of the apparent increase, which we fully expected to be significant at the conventional rate for vegetable crops in the country, may have resulted from the plants' need to control and fight-off the bacterial pathogen. Therefore, the fact that we did not focus on and specifically record the incidence of disease manifestations, albeit seemingly scarce to attract serious attention, we regard as a drawback of the study.

The positive aspect is the fact that we studied root-associated microbiomes of tomato plants grown in the real-world environment in the soil with known genesis and history of agricultural use, with its properties measured and described, as all those provide a detailed set of environmental variables that shall be helpful in meta-analytical attempts to obtain better insights into the factors shaping tomato plant-associated microbiota and its effect on tomato fruit quantity and quality.

5. Conclusions

The results of our study showed very distinct microbiomes around and inside tomato roots. These distinct microbiome patterns, especially in terms of α-biodiversity, were to be expected due to the higher versatility of environmental niches in the rhizosphere, not so narrowly specific and prohibiting as in the endosphere: for instance, bacterial species richness was an order of magnitude higher in the rhizosphere than in the root endosphere. Yet tomato hosts a rather diverse root-associated microbiome composed of dozens of bacterial and fungal species, not all of them originating from rhizosphere soil. Our finding

that root bacteriobiome responded to conventional mineral NPK fertilization already at the high taxonomic level (class), and at the dominant ones at that, illustrates very well the role of NPK-changed plant metabolism in shaping endophytic microbiota and hence fertilization potential in mitigating plant pathogens. We did not examine different rates of NPK fertilization as separate treatments/factor levels in our study, but we believe that using such gradient in further research may bring a more detailed understanding of how to modify and even fine-tune phytobiomes in order to enhance crops' health and yields.

As for the rhizosphere bacteriobiome, the finding of only one minor dominant bacterial OTU, decreasing its presence by 0.5% due to mineral fertilization, indicates the robustness of the rhizosphere bacteriobiome, most likely because of the much more diverse and open microenvironment, where stronger forces are needed to cause greater shifts.

Root bacteriobiome and mycobiome differed in their response to mineral fertilization, most likely due to (a) different mechanisms of tomato roots' control of bacterial and fungal endophytes and (b) greater recalcitrance of fungal hyphae inside plants to any changes.

Knowledge about the fertilizer-induced microbiome shifts in tomato cultivating systems opens a window of opportunity for designing fertilizers targeted at supporting high quantity and quality of yield. This research field seems rather exciting, albeit agronomically and ecologically may turn out to be strongly contextual.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/horticulturae8111051/s1, Table S1 Climate (as averaged 1991–2000, available online: https://meteoinfo.ru/climatcities?p=1930, accessed on 31 October 2022) characteristics of the region where the experiment was performed; Table S2: Soil (Phaeozem) properties at the experimental site in the south of West Siberia, mean ± standard deviation.

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References

1. Bulgarelli, D.; Schlaeppi, K.; Spaepen, S.; ver Loren van Themaat, E.; Schulze-Lefert, P. Structure and functions of the bacterial microbiota of plants. *Ann. Rev. Plant Biol.* **2013**, *64*, 807–838. [CrossRef] [PubMed]
2. Pozo, M.J.; Zabalgogeazcoa, I.; de Aldana, B.R.V.; Martinez-Medina, A. Untapping the potential of plant mycobiomes for applications in agriculture. *Curr. Opin. Plant Biol.* **2021**, *60*, 102034. [CrossRef]
3. Trivedi, P.; Mattupalli, C.; Eversole, K.; Leach, J.E. Enabling sustainable agriculture through understanding and enhancement of microbiomes. *New Phytol.* **2021**, *230*, 2129–2147. [CrossRef]
4. Baltrus, D.A. Adaptation, specialization, and coevolution within phytobiomes. *Opin. Plant Biol.* **2017**, *38*, 109–116. [CrossRef] [PubMed]
5. Papik, J.; Folkmanova, M.; Polivkova-Majorova, M.; Suman, J.; Uhlik, O. The invisible life inside plants: Deciphering the riddles of endophytic bacterial diversity. *Biotechnol. Adv.* **2020**, *44*, 107614. [CrossRef]

6. Kracmarova, M.; Karpiskova, J.; Uhlik, O.; Strejcek, M.; Szakova, J.; Balik, J.; Demnerova, K.; Stiborova, H. Microbial Communities in Soils and Endosphere of *Solanum tuberosum* L. and their Response to Long-Term Fertilization. *Microorganisms* **2020**, *8*, 1377. [CrossRef] [PubMed]
7. Cangioli, L.; Mancini, M.; Napoli, M.; Fagorzi, C.; Orlandini, S.; Vaccaro, F.; Mengoni, A. Differential Response of Wheat Rhizosphere Bacterial Community to Plant Variety and Fertilization. *Int. J. Mol. Sci.* **2022**, *23*, 3616. [CrossRef]
8. Mehlferber, E.C.; McCue, K.F.; Ferrel, J.E.; Koskella, B.; Khanna, R. Temporally Selective Modification of the Tomato Rhizosphere and Root Microbiome by Volcanic Ash Fertilizer Containing Micronutrients. *Appl. Environ. Microbiol.* **2022**, *88*, e0004922. [CrossRef]
9. Food and Agriculture Organization of the United Nations. FAOSTAT. Data. Crops and Livestock Products. Available online: https://www.fao.org/faostat/en/#data/QCL (accessed on 27 August 2022).
10. Allard, S.M.; Walsh, C.S.; Wallis, A.E.; Ottesen, A.R.; Brown, E.W.; Micallef, S.A. *Solanum lycopersicum* (tomato) hosts robust phyllosphere and rhizosphere bacterial communities when grown in soil amended with various organic and synthetic fertilizers. *Sci. Total Environ.* **2016**, *573*, 555–563. [CrossRef]
11. Dong, C.J.; Wang, L.L.; Li, Q.; Shang, Q.M. Bacterial communities in the rhizosphere, phyllosphere and endosphere of tomato plants. *PLoS ONE* **2019**, *14*, e0223847. [CrossRef]
12. IUSS Working Group. *WRB, World Reference Base for Soil Resources 2014, Update 2015: International Soil Classification System for Naming Soils and Creating Legends for Soil Maps*; FAO: Rome, Italy, 2015.
13. Shishov, L.L.; Tonkonogov, V.D.; Lebedeva, I.I.; Gerasimoiva, M.I. (Eds.) *Classification and Diagnostics of Soils in Russia*, 1st ed.; Oykumena Pubs: Moscow, Russia, 2004. (In Russian)
14. Carter, M.R.; Gregorich, E.G. (Eds.) *Soil Sampling and Methods of Analysis*, 2nd ed.; CRC Press: Boca Raton, FL, USA, 2008.
15. Fadrosh, D.W.; Ma, B.; Gajer, P.; Sengamalay, N.; Ott, S.; Brotman, R.M.; Ravel, J. An improved dual-indexing approach for multiplexed 16S rRNA gene sequencing on the Illumina MiSeq platform. *Microbiome* **2014**, *2*, 6. [CrossRef] [PubMed]
16. Tyurin, M.; Kabilov, M.R.; Smirnova, N.; Tomilova, O.G.; Yaroslavtseva, O.; Alikina, T.; Glupov, V.V.; Kryukov, V.Y. Can Potato Plants Be Colonized with the Fungi Metarhizium and Beauveria under Their Natural Load in Agrosystems? *Microorganisms* **2021**, *9*, 1373. [CrossRef] [PubMed]
17. Edgar, R.C. UPARSE: Highly accurate OTU sequences from microbial amplicon reads. *Nat. Methods* **2013**, *10*, 996–998. [CrossRef]
18. Edgar, R.C. UNOISE2: Improved error-correction for Illumina 16S and ITS amplicon reads. *bioRxiv* **2016**, 081257. [CrossRef]
19. Wang, Q.; Garrity, G.M.; Tiedje, J.M.; Cole, J.R. Naïve Bayesian Classifier for Rapid Assignment of rRNA Sequences into the New Bacterial Taxonomy. *Appl. Environ. Microbiol.* **2007**, *73*, 5261–5267. [CrossRef] [PubMed]
20. Abarenkov, K.; Zirk, A.; Piirmann, T.; Pöhönen, R.; Ivanov, F.; Nilsson, R.H.; Kõljalg, U. *UNITE USEARCH/UTAX Release for Fungi*, Version 18.11.2018; UNITE Community: Tartu, Estonia, 2018. [CrossRef]
21. Fauth, E.; Bernardo, J.; Camara, M.; Resetarits, W.J., Jr.; Van Buskirk, J.; McCollum, S.A. Simplifying the Jargon of Community Ecology: A Conceptual Approach. *Am. Nat.* **1996**, *147*, 282–286. [CrossRef]
22. Hammer, O.; Harper, D.A.T.; Ryan, P.D. PAST: Paleontological Statistics Software Package for Education and Data Analysis. *Palaeontol. Electron.* **2001**, *4*, 9.
23. Hughes, J.B.; Hellmann, J.J. The Application of Rarefaction Techniques to Molecular Inventories of Microbial Diversity. *Methods Enzym.* **2005**, *397*, 292–308. [CrossRef]
24. Lee, S.A.; Kim, Y.; Kim, J.M.; Chu, B.; Joa, J.H.; Sang, M.K.; Song, J.; Weon, H.Y. A preliminary examination of bacterial, archaeal, and fungal communities inhabiting different rhizocompartments of tomato plants under real-world environments. *Sci. Rep.* **2019**, *9*, 9300. [CrossRef]
25. Anzalone, A.; Mosca, A.; Dimaria, G.; Nicotra, D.; Tessitori, M.; Privitera, G.F.; Pulvirenti, A.; Leonardi, C.; Catara, V. Soil and Soilless Tomato Cultivation Promote Different Microbial Communities That Provide New Models for Future Crop Interventions. *Int. J. Mol. Sci.* **2022**, *23*, 8820. [CrossRef]
26. López, S.; Pastorino, G.N.; Fernández-González, A.J.; Franco, M.; Fernández-López, M.; Balatti, P.A. The endosphere bacteriome of diseased and healthy tomato plants. *Arch. Microbiol.* **2020**, *202*, 2629–2642. [CrossRef] [PubMed]
27. Zuluaga, M.; Lima Milani, K.M.; Azeredo Gonçalves, L.S.; Martinez de Oliveira, A.L. Diversity and plant growth-promoting functions of diazotrophic/N-scavenging bacteria isolated from the soils and rhizospheres of two species of Solanum. *PLoS ONE* **2020**, *15*, e0227422. [CrossRef] [PubMed]
28. Bulgarelli, D.; Rott, M.; Schlaeppi, K.; ver Loren van Themaat, E.; Ahmadinejad, N.; Assenza, F.; Rauf, P.; Huettel, B.; Reinhardt, R.; Schmelzer, E.; et al. Revealing structure and assembly cues for *Arabidopsis* root-inhabiting bacterial microbiota. *Nature* **2012**, *488*, 91–95. [CrossRef]
29. Lundberg, D.S.; Lebeis, S.L.; Paredes, S.H.; Yourstone, S.; Gehring, J.; Malfatti, S.; Tremblay, J.; Engelbrektson, A.; Kunin, V.; Rio, T.G.; et al. Defining the core *Arabidopsis thaliana* root microbiome. *Nature* **2012**, *488*, 86–90. [CrossRef] [PubMed]
30. Colagiero, M.; Rosso, L.C.; Catalano, D.; Schena, L.; Ciancio, A. Response of Tomato Rhizosphere Bacteria to Root-Knot Nematodes, Fenamiphos and Sampling Time Shows Differential Effects on Low Level Taxa. *Front. Microbiol.* **2020**, *11*, 390. [CrossRef]
31. Naumova, N.B.; Belanov, I.P.; Alikina, T.Y.; Kabilov, M.R. Undisturbed Soil Pedon under Birch Forest: Characterization of Microbiome in Genetic Horizons. *Soil Syst.* **2021**, *5*, 14. [CrossRef]

32. Xia, Y.; DeBolt, S.; Dreyer, J.; Scott, D.; Williams, M.A. Characterization of culturable bacterial endophytes and their capacity to promote plant growth from plants grown using organic or conventional practices. *Front. Plant Sci.* **2015**, *6*, 490. [CrossRef]
33. Khan, A.L.; Waqas, M.; Kang, S.M.; Al-Harrasi, A.; Hussain, J.; Al-Rawahi, A.; Al-Khiziri, S.; Ullah, I.; Ali, L.; Jung, H.Y.; et al. Bacterial endophyte *Sphingomonas* sp. LK11 produces gibberellins and IAA and promotes tomato plant growth. *J. Microbiol.* **2014**, *52*, 689–695. [CrossRef]
34. Akter, S.; Lee, S.Y.; Moon, S.K.; Choi, C.; Balusamy, S.R.; Siddiqi, M.Z.; Ashrafudoulla, M.; Huq, M.A. *Sphingomonas horti* sp. nov., a novel bacterial species isolated from soil of a tomato garden. *Arch. Microbiol.* **2021**, *203*, 543–548. [CrossRef]
35. Yoo, S.J.; Weon, H.Y.; Song, J.; Sang, M.K. Effects of *Chryseobacterium soldanellicola* T16E-39 and *Bacillus siamensis* T20E-257 on biocontrol against phytophthora blight and bacterial wilt and growth promotion in tomato plants. *Int. J. Agric. Biol.* **2020**, *23*, 534–540.
36. Kwak, M.J.; Kong, H.G.; Choi, K.; Kwon, S.K.; Song, J.Y.; Lee, J.; Lee, P.A.; Choi, S.Y.; Seo, M.; Lee, H.J.; et al. Rhizosphere microbiome structure alters to enable wilt resistance in tomato. *Nat. Biotechnol.* **2018**, *36*, 1100–1109. [CrossRef] [PubMed]
37. Tancos, M.A.; Chalupowicz, L.; Barash, I.; Manulis-Sasson, S.; Smart, C.D. Tomato fruit and seed colonization by Clavibacter michiganensis subsp. michiganensis through external and internal routes. *Appl. Environ. Microbiol.* **2013**, *79*, 6948–6957. [CrossRef] [PubMed]
38. Dastogeer, K.; Yasuda, M.; Okazaki, S. Microbiome and pathobiome analyses reveal changes in community structure by foliar pathogen infection in rice. *Front. Microbiol.* **2022**, *13*, 949152. [CrossRef] [PubMed]
39. Ftayeh, R.; von Tiedemann, A.; Koopmann, B.; Rudolph, K.; Abu-Ghorrah, M. First Record of *Clavibacter michiganensis* subsp. *michiganensis* Causing Canker of Tomato Plants in Syria. *Plant Dis.* **2008**, *92*, 649. [CrossRef] [PubMed]
40. Ansari, M.; Taghavi, S.M.; Hamzehzarghani, H.; Valenzuela, M.; Siri, M.I.; Osdaghi, E. Multiple Introductions of Tomato Pathogen *Clavibacter michiganensis* subsp. michiganensis into Iran as Revealed by a Global-Scale Phylogeographic Analysis. *Appl. Environ. Microbiol.* **2019**, *85*, e02098-19. [CrossRef] [PubMed]
41. Naumova, N.; Nechaeva, T.; Smirnova, N.; Fotev, Y.; Belousova, S. Effect of Sapropel Addition on Selected Soil Properties and Field Tomato Yield in South West Siberia. *Asian J. Soil Sci. Plant Nutr.* **2017**, *1*, 1–11. [CrossRef]
42. Mechan Llontop, M.E.; Sharma, P.; Aguilera Flores, M.; Yang, S.; Pollok, J.; Tian, L.; Huang, C.; Rideout, S.; Heath, L.S.; Li, S.; et al. Strain-Level Identification of Bacterial Tomato Pathogens Directly from Metagenomic Sequences. *Phytopathology* **2020**, *110*, 768–779. [CrossRef]
43. Osdaghi, E.; Rahimi, T.; Taghavi, S.M.; Ansari, M.; Zarei, S.; Portier, P.; Briand, M.; Jacques, M.A. Comparative Genomics and Phylogenetic Analyses Suggest Several Novel Species within the Genus Clavibacter, Including Nonpathogenic Tomato-Associated Strains. *Appl. Environ. Microbiol.* **2020**, *86*, e02873-19. [CrossRef]
44. Jacques, M.A.; Durand, K.; Orgeur, G.; Balidas, S.; Fricot, C.; Bonneau, S.; Quillévéré, A.; Audusseau, C.; Olivier, V.; Grimault, V.; et al. Phylogenetic analysis and polyphasic characterization of *Clavibacter michiganensis* strains isolated from tomato seeds reveal that nonpathogenic strains are distinct from C. michiganensis subsp. michiganensis. *Appl. Environ. Microbiol.* **2012**, *78*, 8388–8402. [CrossRef]
45. Asaf, S.; Numan, M.; Khan, A.L.; Al-Harrasi, A. *Sphingomonas*: From diversity and genomics to functional role in environmental remediation and plant growth. *Crit. Rev. Biotechnol.* **2020**, *40*, 138–152. [CrossRef]
46. Jung, J.; Kim, J.S.; Taffner, J.; Berg, G.; Ryu, C.M. Archaea, tiny helpers of land plants. *Comput. Struct. Biotechnol. J.* **2020**, *18*, 2494–2500. [CrossRef] [PubMed]
47. Zhang, X.; Tang, G.; Xu, X.; Nong, X.; Qi, S. Insights into Deep-Sea Sediment Fungal Communities from the East Indian Ocean Using Targeted Environmental Sequencing Combined with Traditional Cultivation. *PLoS ONE* **2014**, *9*, e109118. [CrossRef] [PubMed]
48. Sen, K.; Sen, B.; Wang, G. Diversity, Abundance, and Ecological Roles of Planktonic Fungi in Marine Environments. *J. Fungi* **2022**, *8*, 491. [CrossRef]
49. Lu, Y.; Wang, X.; Almeida, L.C.S.d.S.; Pecoraro, L. Environmental Factors Affecting Diversity, Structure, and Temporal Variation of Airborne Fungal Communities in a Research and Teaching Building of Tianjin University, China. *J. Fungi* **2022**, *8*, 431. [CrossRef] [PubMed]
50. Schmidt, J.E.; Vannette, R.L.; Igwe, A.; Blundell, R.; Casteel, C.L.; Gaudin, A.C.M. Effects of agricultural management on rhizosphere microbial structure and function in processing tomato plants. *Appl. Environ. Microbiol.* **2019**, *85*, e01064-19. [CrossRef] [PubMed]
51. Carlucci, A.; Raimondo, M.L.; Santos, J.; Phillips, A.J. *Plectosphaerella* species associated with root and collar rots of horticultural crops in southern Italy. *Persoonia* **2012**, *28*, 34–48. [CrossRef]
52. Harrison Wright, A.; Ali, S.; Migicovsky, Z.; Douglas, G.M.; Yurgel, S.; Bunbury-Blanchette, A.; Franklin, J.; Adams, S.J.; Walker, A.K. A Characterization of a Cool-Climate Organic Vineyard's Microbiome. *Phytobiomes J.* **2022**, *6*, 69–82. [CrossRef]
53. Han, L.; Zhou, X.; Zhao, Y.; Wu, L.; Ping, X.; He, Y.; Peng, S.; He, X.; Du, Y. First report of *Plectosphaerella plurivora* causing root rot disease in *Panax notoginseng* in China. *Phytopathol. Z.* **2020**, *168*, 375–379. [CrossRef]
54. Sosa, A.L.; Rosso, L.C.; Salusso, F.A.; Etcheverry, M.G.; Passone, M.A. Screening and identification of horticultural soil fungi for their evaluation against the plant parasitic nematode *Nacobbus aberrans*. *World J. Microbiol. Biotechnol.* **2018**, *34*, 63. [CrossRef]

55. Bachmann, H.P.; Bobst, C.; Bütikofer, U.; Casey, M.G.; Dalla Torre, M.; Fröhlich-Wyder, M.T.; Fürst, M. Occurrence and significance of *Fusarium domesticum* alias *Anticollanti* on smear-ripened cheeses. *LWT Food Sci. Technol.* **2005**, *38*, 399–407. [CrossRef]
56. Patel, R.; Mehta, K.; Prajapati, J.; Shukla, A.; Parmar, P.; Goswami, D.; Saraf, M. An anecdote of mechanics for Fusarium biocontrol by plant growth promoting microbes. *Biol. Control* **2022**, *174*, 105012. [CrossRef]