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Exploring the Influence of Soil Salinity on Microbiota Dynamics in *Vitis vinifera* cv. "Glera": Insights into the Rhizosphere, Carposphere, and Yield Outcomes

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Abstract: In a world grappling with the severe effects induced by climate change, one of the most significant concerns affecting agriculture is the gradual decline in water quality for irrigation associated with reduced rainfalls and the consequent increase in soil salinity. This issue is particularly crucial for grapevine cultivation (*Vitis vinifera* L.) and the associated winemaking industry. The aroma of the resulting wines and the yield parameters can be influenced both directly by water quality and indirectly due to the effects exerted by salinity on the microbiota, which directly impacts plant health. To gain insights into this topic, our study aimed to analyse the changes induced in the microbiota of both the rhizosphere and the carposphere due to salt stress using a metabarcoding approach, focusing on *Vitis vinifera* cv. Glera. The control plants were irrigated with rainwater, while the treated plants were irrigated with water containing salt (NaCl). Our findings revealed significant differences in the microbiota (both fungi and bacteria) of the rhizosphere and carposphere between the two treatments. For instance, the Shannon diversity index (i.e., alpha diversity) was lower in the treated plants compared to the control not-treated ones, whilst the beta diversity did not show any differences. Several microbial phyla exhibited better resilience to this abiotic stress (e.g., *Ascomycota*, *Saccharomycetes*, *Acidobacteria*, *Proteobacteria*, *Bacteroidetes*), shedding light on their impact on crucial bacterial and fungal groups essential for the subsequent winemaking stages. Additionally, the salt stress negatively affected the yield parameters. This study contributes valuable insights to the viticultural community, providing a deeper understanding of the complex interplay between soil characteristics, microbial communities, and their influence on productivity.

Keywords: climate change; salinity; soil; microbiota; rhizosphere; carposphere; wine

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1. Introduction

Due to environmental changes linked to global warming, the agricultural value chain is faced with the challenge of managing increasing soil salinity levels. Changes in precipitation cause environmental modifications such as lowering water tables and salt-water intrusion as sea levels rise. These, combined with the ever-increasing need to reuse degraded water, are some of the factors that will influence the salinity levels of agricultural soils in the near future [1,2].

Soluble salts, which are naturally present in the soil or are derived from fertilization and also from irrigation water, can have a significant impact on grapevine physiology. Salts are essential for plant nutrition, influencing key physiological processes such as water

uptake and nutrient absorption. Their balanced presence is critical for plants, as an increase in salts makes it difficult to absorb water and minerals due to increased osmotic pressure. As a result, while certain concentration levels can enhance grape flavours, an excessive salt content causes stress to the plant, leading to stunted grapevine development, decreased harvests, and inferior fruit quality [3,4].

In this regard, climate change is consistently reducing precipitation in certain regions, necessitating an increased reliance on irrigation with water containing salts and minerals as opposed to rainwater [5–7]. Furthermore, climate change will have an impact on the aforementioned irrigation waters, reducing their availability while also altering their chemical balance and enriching them with salts [8–10].

In addition to the direct effects on the plant, the soil microbiota must also be considered. Microbes play an important role in shaping the vineyard's terroir, the unique environmental factors that contribute to the taste and character of grapes and wines, as well as interacting with grapevine roots, influencing nutrient cycling and disease resistance and thus directly influencing the yield and aromatic profile of the resulting wines [11,12]. Understanding this intricate relationship is then critical to unravelling the complexities of the terroir and producing wines with distinct character and quality.

Some bacteria and fungi have adapted more than others to thrive in these increased saline concentrations, forming a distinct microbial community that is critical to the overall health of the soil, influencing biodiversity and microbial composition characteristics in saline soils [13,14]. Particular attention should be paid to the study of halophilic mycorrhizal fungi, which, under these stress conditions, may improve grapevine salinity tolerance by enhancing water and nutrient uptake via their symbiotic relationships with plant roots [15]. For instance, the relative abundances of several plant-growth-promoting bacteria, including *Planctomycetes*, *Bacteroidetes*, *Verrucomicrobia*, *Cyanobacteria*, *Gemmatimonadetes*, *Chloroflexi*, and *Firmicutes*, increased with salinized soils, and their metabolic function exerted in the soil might play vital roles in the mitigation of salt stress on grapevines [16].

Bringing the soil salinity and microbiota together allows us to investigate a dynamic interplay in which the saline composition of the soil influences the composition and function of the microbial community not only in the soil but also in the berry carposphere. Differences in the chemical–physical characteristics of the berries can alter the ecology and health status of the carposphere [17], resulting in differences in microbial populations, particularly those of indigenous yeasts, which, along with their metabolites during the first stages of fermentation, are important in modelling the aromatic profile of the obtained wines [18,19].

In this sense, our study aimed to investigate the relationship between soil salinity and the microbiota within vineyards. Understanding how salt levels influence the composition of fungi and bacteria is critical not only for understanding the complex dynamics of the vineyard ecosystem but also for practical viticulture and winemaking applications. Identifying specific bacteria and fungi that thrive or struggle in saline conditions could help inform strategies for improving soil health, enhancing grapevine resilience, and potentially influencing the terroir of the resulting wines.

To that end, the research compared *Vitis vinifera* cv. Glera grown both in non-saline soil and in soil with added salts to increase salinity. Subsequently, the main growth parameters of the plant were evaluated, and the analysis of the rhizosphere and berry microbiota was conducted through metagenomic analyses.

2. Materials and Methods

2.1. Experimental Design

For this experiment, plants of *Vitis vinifera* cultivar Glera, were grafted onto 140 Ruggeri rootstocks (http://catalogoviti.politicheagricole.it/scheda.php?codice=622 accessed on 12 January 2024). The vine cuttings were cultivated over a 4-year period in a glasshouse within pots containing the same type and amount (100 L) of initial substrate. The plants in the control and treated groups were maintained under identical conditions. Temperature

and solar radiation in autumn, winter, and spring followed the outside conditions, whilst the greenhouse was set up at max 28 °C during summer, using fans to extract the excessive heat inside. Fertilization was conducted during early spring and autumn, utilizing an equal amount of a balanced fertilizer containing 15% nitrogen (N), 10% phosphorus (P), and 20% potassium (K), along with magnesium (Mg) and sulphur (S). The application of plant agrochemicals remained consistent across both treatments in each study year, with the same quantity and frequency of treatments maintained throughout the experiment. During the fifth year of growth, according to the needs of the plant, the control ones were irrigated using rainwater collected in designated containers, while the treated plants were irrigated with water obtained from an irrigation canal and the soil was amended with 0.1 L of a suspension 0.5 M of NaCl on a weekly basis (Figure 1). The soil conductivity was measured using a conductivity meter (Hanna Instruments, Smithfield, RI, USA).

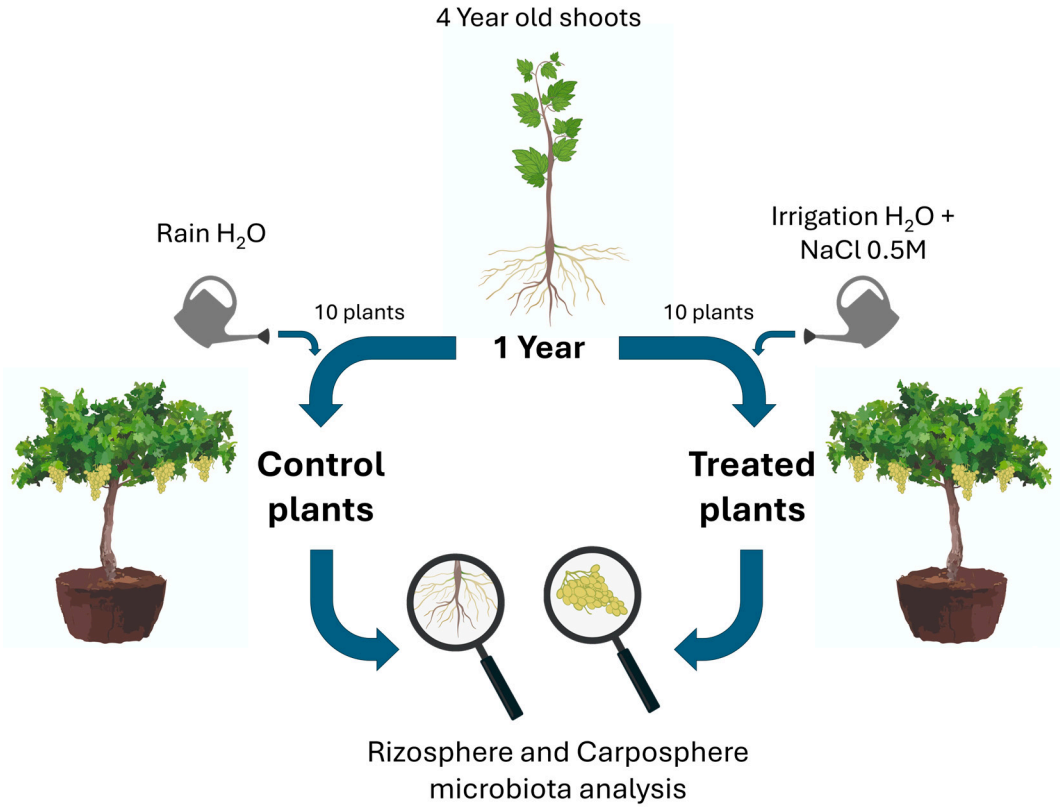

Figure 1. Graphic summary of the experimental design adopted in this work.

2.2. Field Sampling, DNA Extraction, and Amplification

For the analysis of the rhizosphere microbiota, the roots of 10 plants per treatment were up-taken and processed to obtain the rhizosphere, following the protocol of Mian et al., 2023 [20]. For each rhizosphere sample, three subsamples were placed into a 50 mL Falcon tube filled with 30 mL of epiphyte removal buffer adjusted to pH 6.5 (composed of 6.75 g of KH_2PO_4, 8.75 g of K_2HPO_4, and 1 mL of Triton X-100 per litre of deionized water). The samples underwent sonication at 600 Hz, alternating between 30 s of sonication and 30 s without sonication, for a total of 10 min at 4 °C. Following sonication, the tubes were centrifuged at 12,000× *g* for 10 min at 4 °C to pellet the cells. The pelleted cells were then separated from the supernatant and stored at −80 °C. For the rhizosphere, the total genomic DNA was extracted from 250 mg of rhizosphere from each individual sample using the Quick-DNA Fecal/Soil Microbe Miniprep kit (Zymo Research Corp., Irvine, CA, USA), following the manufacturer's instructions. As for the carposphere, both control and treated berries were sampled immediately before harvest time and handled under sterile conditions [21]. For each sample, the peels of 60 berries were placed into 50 mL

Falcon tubes, each containing 30 mL of epiphyte removal buffer, following the previously described method to obtain pellets. DNA extraction was conducted using the NucleoSpin Plant II kit (Carlo Erba, Cornaredo, Italy), following the manufacturer's instructions. The DNA sample's concentration and quality were assessed using the NanoDrop ND-1000 spectrophotometer (ThermoFisher Scientific, Waltham, MA, USA) to achieve 30 ng/µL diluted DNA samples. Polymerase Chain Reaction (PCR) was employed to amplify the ITS and 16S regions for fungi and bacteria, respectively, with 10 samples for the control group, 10 for the treated group, and the same number for ITS. For the bacterial 16S rRNA gene targeting the V3–V4 region amplification, PCR was performed using the following mixture: 10 µL of 2 × Dr. MAX Master Mix Solution (Doctor Protein Corp., Seoul, Korea), 1 µM of 341F (5′-CCTACGGGNGGCWGCAG-3′) and 805R (5′-GAC-TACHVGGGTATCTAATCC-3′) primer set, and 30 ng of extracted DNA as a template. For eukaryotic fungal ITS region amplification, the same protocol was followed with different primers (1 µM of ITS3 (5′-GCATCGATGAAGAACGCAGC-3′) and ITS4 (5′-TCCTCCGCTTATTGATATGC-3′)). PCR amplification involved initial denaturation at 95 °C for 7 min, followed by 30 cycles at 95 °C for 30 s, 55 °C for 30 s, and 72 °C for 30 s, with a final extension at 72 °C for 10 min. The amplification products were confirmed by electrophoresis on 1.5% agarose gel in TBE (tris-borate EDTA) buffer for 1.5 h, followed by purification using the GenElute™ Microbiome DNA Purification Kit (Merck, Darmstadt, Germany). The amplicons were subsequently sequenced using the Illumina MiSeq platform (Illumina Inc., San Diego, CA, USA) by an external company, with Nextera adapters introduced.

2.3. Bioinformatic and Statistical Analysis

Bioinformatics analyses were conducted via the QIIME2 software package (v2021.11) [22]. Raw paired-end sequences underwent filtering, trimming, denoising, and merging using Cutadapt [23] and DADA2 [24], resulting in the generation of Amplicon Sequence Variants (ASVs). Taxonomy was assigned to the ASVs using the sklearn naïve Bayes taxonomy classifier [25]. SILVA v.138 was employed for taxonomic analysis of the 16S reads to train the classifier based on the primer sequences used for PCR amplification [26], whilst the Unite v8.3 dynamic 2021-05-10 database encompassing all eukaryotic species was utilized for the ITS reads [27]. Sequences assigned to mitochondria, chloroplasts, those with frequencies < 2 (singletons), or unidentified ones were filtered out from the dataset. Subsequent statistical analyses were carried out using R v4.1.2. Prior to further analyses, the adequacy of genomic coverage was confirmed by evaluating rarefaction curves on species richness, calculated using ggrare from the ranacapa package [28]. Following rarefaction of the samples implemented through the phyloseq package [29], an alpha-diversity analysis was performed. To visually represent the microbiological differences between the two treatments, a Principal Coordinates Analysis (PCoA) was conducted using Bray–Curtis dissimilarity matrices, constructed using the vegan package [30] assessing the presence of a significant treatment effect with a PERmutational Multivariate Analysis Of Variance (PERMANOVA) based on 4999 permutations [31].

2.4. Yield, Must Chemical Composition, and Radical System Study

All the vines were individually handpicked, and the total number of clusters per vine, yield, and average cluster weight were recorded. The number of shoots was counted for each vine, and all the collected leaves were arranged on a 1.00 m × 1.00 m panel and photographed using a digital camera. The image analysis was performed using ImageJ Version 1.54i (NIH, Bethesda, MD, USA), recording parameters such as the number of leaves, leaf size, and total leaf area per vine [32]. At harvest time, the total sugar content was measured using a refractometer (Atago PR32, Fischer scientific, Hampton, USA) at 23 °C. The total acidity (expressed as $g \cdot L^{-1}$ of tartaric acid) and pH were determined using an automatic titrator (Crison Micro TT 2022) through titration with 0.1 N NaOH solution. The concentration of L-malic acid was determined spectrophotometrically using the enzymatic kit L-malic acid (Megazyme, Bray, Ireland), following the manufacturer's

instructions. The root number was determined using the profile wall method as suggested by Boehm (1979) [33,34]. Thick roots were counted for each vine using a 0.1 m high and 0.1 m wide grid system positioned against the profile wall, with the centre grapevine trunk as the reference point. Thick roots were selected due to their high metabolic activity and role as carbohydrate storage sinks [35]. The data are presented as total vine roots per square meter.

3. Results

3.1. Soil and Plant Growth Differences

The measured salinity difference highlighted the effectiveness of the treatment, showing significantly higher conductivity values ($p < 0.001$) in the treated samples, with mean values of 1.458 ± 0.103 dS/m compared to those of 0.559 ± 0.099 dS/m in the control.

Analysing the growth and yield parameters of the plant (Table 1), significant differences induced by the higher salinity of the treatment compared to the control were observed. Assessing the characteristics of grapes, a significantly higher sugar content ($p < 0.01$) in grapes from control plants (control: 23.80 ± 1.24 °Brix; treatment: 21.83 ± 1.08 °Brix) was found. Conversely, a significantly higher concentration of total acidity ($p < 0.05$) (control: 5.78 ± 0.77 g·L^{-1}; treatment: 6.89 ± 0.98 g·L^{-1}) and malic acid ($p < 0.05$) (control: 2.12 ± 0.35 g·L^{-1}; treatment: 2.47 ± 0.34 g·L^{-1}), along with significantly higher pH values ($p < 0.05$), were observed in grapes from the treatment group (control: 3.57 ± 0.23; treatment: 3.28 ± 0.14).

Table 1. Yield parameters and root density of the control and treated vines. Statistical analysis (*T* test) is reported (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$).

	Mean		SD		*p*-Value
	Control	Treated	Control	Treated	
Sugar (°Brix)	23.80	21.83	1.24	1.08	**
Total acidity (g·L^{-1})	5.78	6.89	0.77	0.98	*
pH	3.57	3.28	0.23	0.14	*
Malic acid (g·L^{-1})	2.12	2.47	0.35	0.34	*
Shoots/vine	29.00	25.90	3.13	3.84	
Leaves number/vine	610.00	588.80	27.08	30.95	
Total leaf area/vine (m^2)	31.39	29.90	6.27	5.14	
Leaf size (cm^2)	1.92	1.76	0.42	0.32	
Yield/vine (kg)	3.70	3.10	0.56	0.94	
Clusters/vine	11.90	9.00	2.96	4.03	
Cluster weight (g)	308.00	341.00	11.00	20.00	***
Root density (number of thick roots/10 cm^2 profile wall)	13.20	10.09	2.00	3.00	*

Although the vegetative growth parameters seemed to favour the control plants, no significant differences were identified in the number of shoots/vine (control: 29.00 ± 3.13; treatment: 25.90 ± 3.84), the number of leaves per vine (control: 610.00 ± 27.08; treatment: 588.80 ± 30.95), total leaf area/vine (control: 31.39 ± 6.27 m^2; treatment: 29.90 ± 5.14 m^2), and leaf size area (control: 1.92 ± 0.42 m^2; treatment: 1.76 ± 0.32 m^2). Regarding other agronomic parameters, a significantly higher cluster weight was found in the treated (control: 308.00 ± 11.00 g; treatment: 341.00 ± 20.00 g), and a higher root density ($p < 0.05$) was observed in the control plants (control: 13.20 ± 2.00; treatment: 10.09 ± 3.00).

3.2. Sequencing Results

A total of 6,391,916 reads were obtained from Illumina MiSeq sequencing, with sequences evenly distributed across all samples. Following the removal of contaminants, adapter trimming, quality filtering, and deletion of chimeric reads, approximately 33% of the sequences were discarded, leaving an average of around 45,000 reads per sample for subsequent analysis. None of the reads were identified as plant DNA. After filtering,

approximately 2,109,756 reads remained as singletons. All rarefaction curves reached saturation plateaus, indicating adequate sequencing depth for all samples and optimal coverage.

3.3. Alpha and Beta Diversity

Analysing the results derived from the assessment of alpha diversity using Shannon's H index, under conditions of higher salinity, a reduction in biodiversity in both the rhizosphere and the carposphere was observed. In the rhizosphere, concerning both bacterial populations (control H = 6.06 ± 0.62; treated H = 5.36 ± 0.82) (Figure 2A) and fungal populations (control H = 5.20 ± 0.33; treated H = 4.80 ± 0.50) (Figure 2B), the higher biodiversity was statistically significant ($p < 0.05$). Conversely, in the carposphere, although both bacterial populations (control H = 2.89 ± 0.45; treated H = 2.61 ± 0.12) (Figure 2C) and fungal populations (control H = 1.93 ± 0.35; treated H = 1.83 ± 0.54) (Figure 2D) exhibited higher levels of biodiversity in the control group, the difference was not statistically significant ($p > 0.05$).

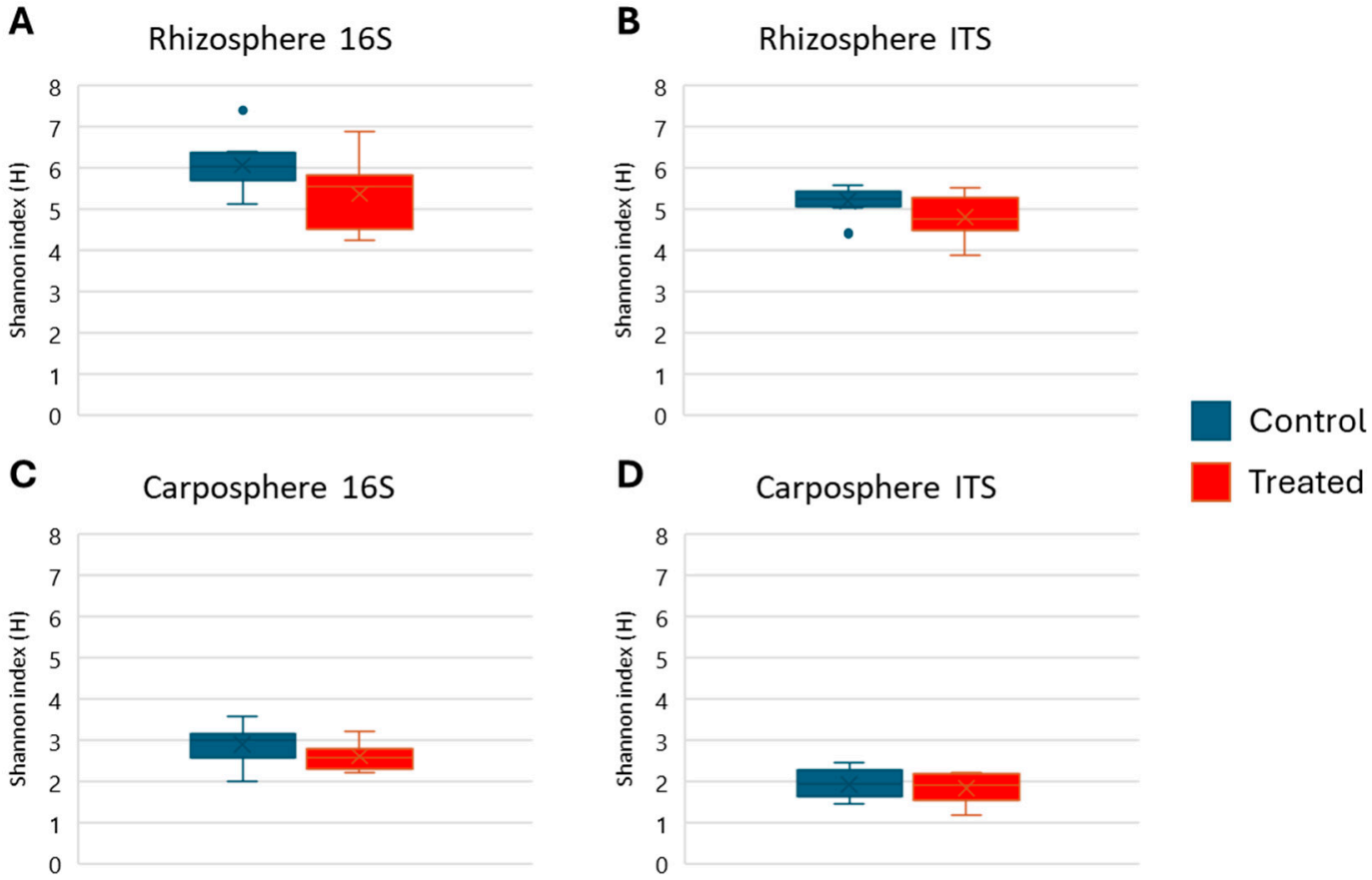

Figure 2. Rhizosphere alpha diversity considering bacterial (**A**) and fungal (**B**) kingdoms; carposphere alpha diversity considering bacterial (**C**) and fungal (**D**) kingdoms.

Considering the beta diversity, for the rhizosphere, concerning both bacterial (Figure 3A) and fungal populations (Figure 3B), no significant differences were identified when evaluating the composition of the entire communities through PCoA calculated via the Bray–Curtis dissimilarity. The same considerations can be made at the level of the carposphere, again evaluating the bacterial (Figure 4A) and fungal (Figure 4B) populations.

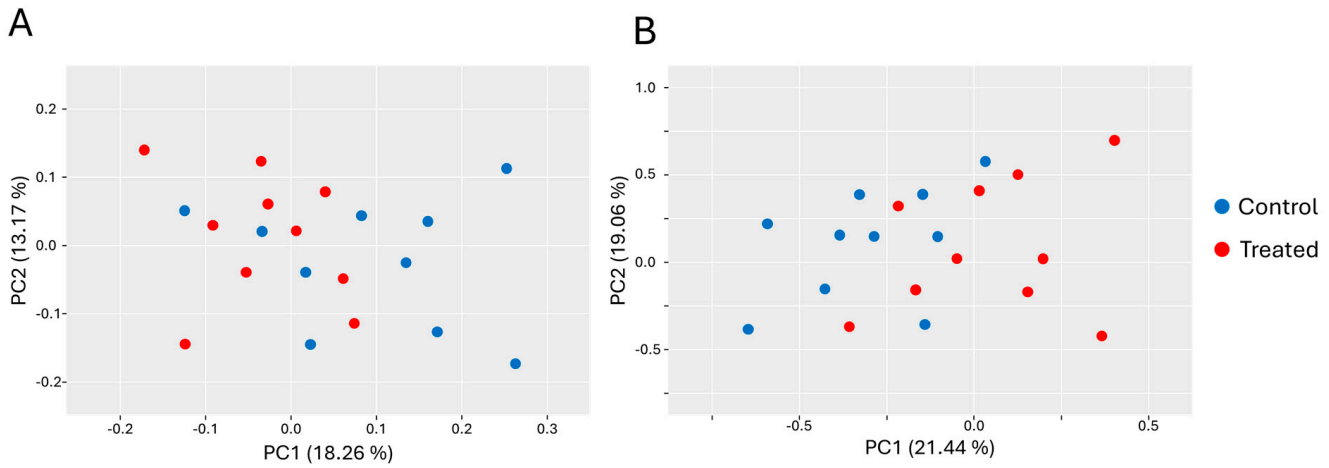

Figure 3. Rhizosphere beta diversity considering both bacterial (**A**) and fungal kingdoms (**B**).

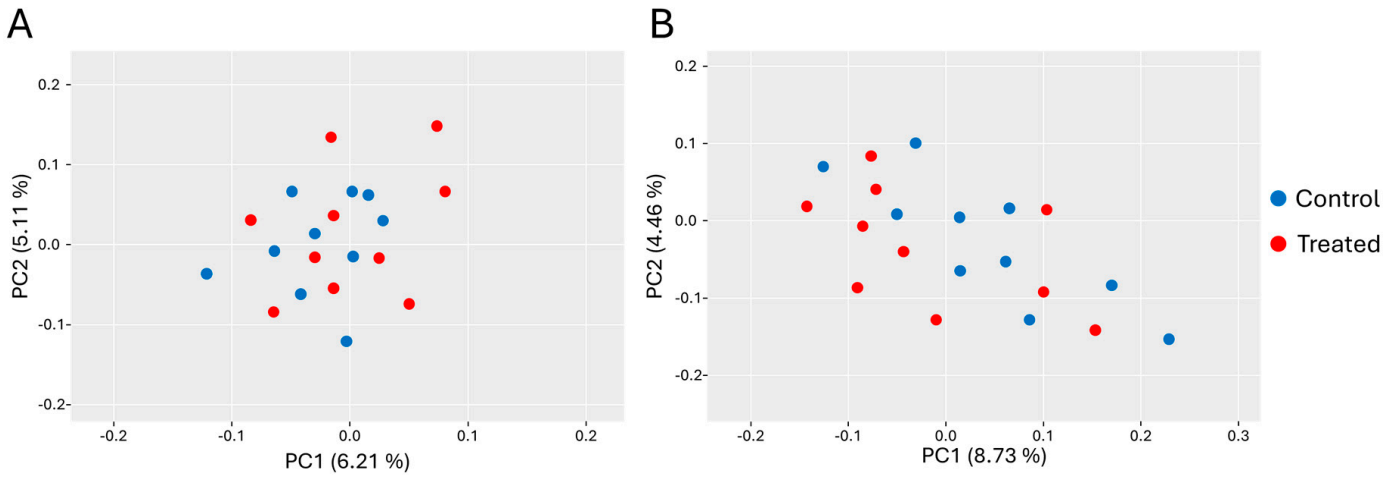

Figure 4. Carposphere beta diversity considering both bacterial (**A**) and fungal kingdoms (**B**).

3.4. Microbial Composition

By analysing the bacterial composition in the rhizosphere analyses, 12 phyla were identified with a relative percentage greater than 0.5% (Figure 5A). In both trials, the predominant phylum was Proteobacteria (control: 39.13 ± 2.77%; treated: 41.50 ± 2.78%), followed by Acidobacteria, which exhibited a significantly higher abundance ($p < 0.05$) in the treatment group (control: 11.06 ± 1.21%; treated: 13.07 ± 2.37%). Other phyla showing significant differences ($p < 0.05$) in abundance in the treated group included Chloroflexi (control: 8.76 ± 1.75%; treated: 6.53 ± 1.92%), Bacteroidetes (control: 6.27 ± 0.74%; treated: 7.37 ± 1.26%), and Nitrospirae (control: 1.71 ± 0.38%; treated: 1.38 ± 0.23%), while Firmicutes were significantly more prevalent ($p < 0.05$) in the control group (control: 5.53 ± 0.58%; treated: 4.66 ± 1.09%). Other identified phyla that did not exhibit statistically significant differences included Actinobacteria (control: 11.36 ± 2.53%; treated: 9.99 ± 3.10%), Gemmatimonadetes (control: 3.70 ± 0.73%; treated: 3.32 ± 0.84%), Patescibacteria (control: 3.71 ± 0.75%; treated: 3.54 ± 0.84%), Planctomycetes (control: 2.69 ± 0.78%; treated: 3.19 ± 0.64%), Verrucomicrobia (control: 1.44 ± 0.77%; treated: 1.86 ± 0.49%), and Cyanobacteria (control: 0.74 ± 0.29%; treated: 0.65 ± 0.17%).

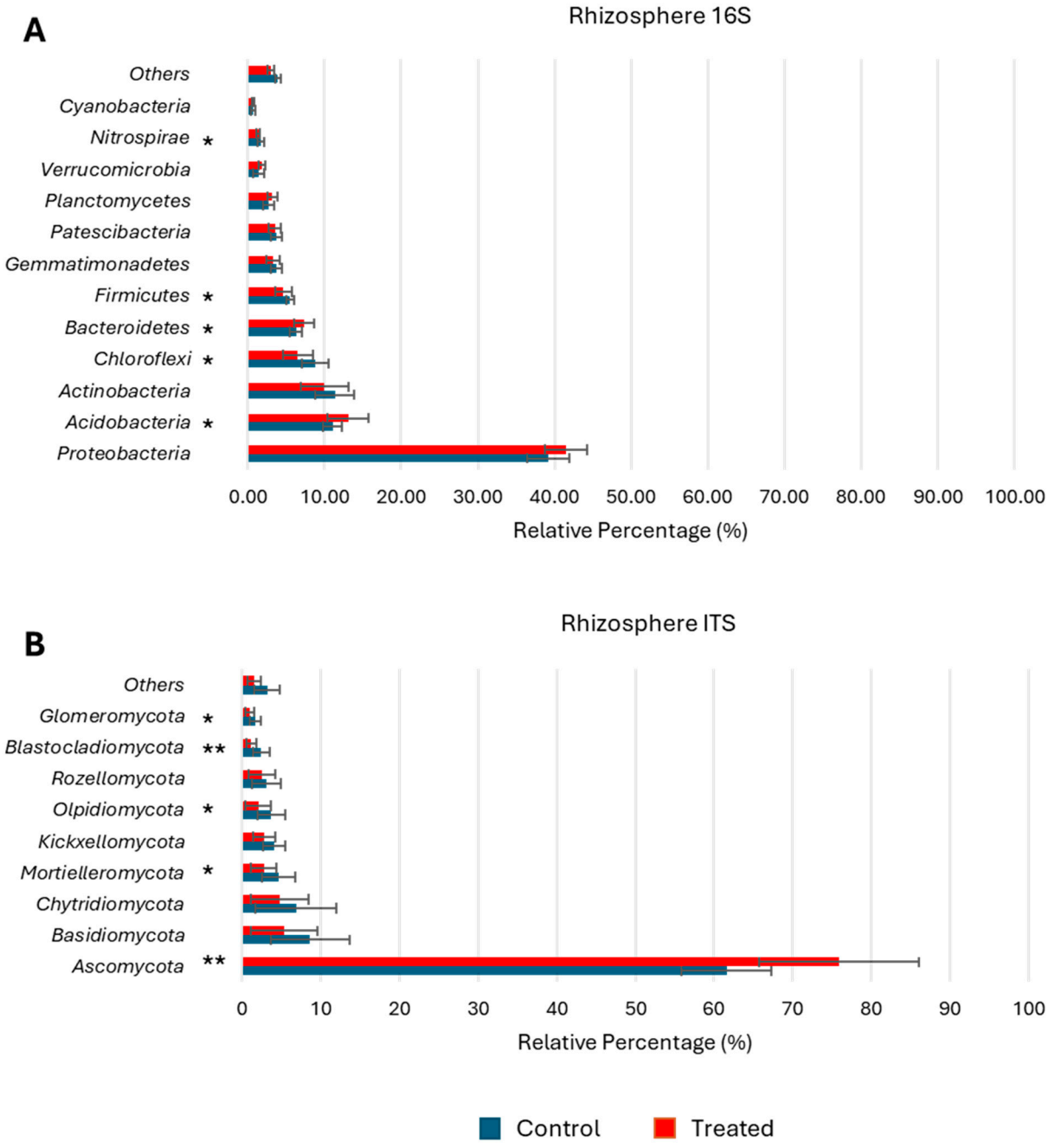

Figure 5. (**A**) Bacterial (16S) phyla above 0.5% identified in the plant rhizosphere of both control and treated plants; (**B**) fungal (ITS) phyla identified above 0.5% in the plant rhizosphere of both control and treated plants. Statistical analysis (*T* test) is reported (* $p < 0.05$, ** $p < 0.01$).

Analysing the rhizosphere-resident fungi, nine phyla accounting for more than 0.5% were identified (Figure 5B). The predominant phylum identified, comprising over 60% of the relative percentage alone, was Ascomycota. A significantly higher presence of Ascomycota was observed in the control samples ($p < 0.01$) (control: 61.56 ± 5.65%; treated: 75.89 ± 10.21%). Conversely, other phyla exhibiting a significantly greater presence in the control samples ($p < 0.05$) included Mortierellomycota (control: 4.67 ± 2.08%; treated: 2.76 ± 2.08%) and Glomeromycota (control: 1.68 ± 0.74%; treated: 1.04 ± 0.55%). The phyla for which no significant differences were identified included Basidiomycota (control: 8.65 ± 5.03%; treated: 5.37 ± 4.21%), Chytridiomycota (control: 6.89 ± 5.14%; treated: 4.83 ± 3.64%), Kickxellomycota (control: 4.04 ± 1.41%; treated: 2.79 ± 1.38%), Olpidiomycota (control: 3.71 ± 1.77%; treated: 2.06 ± 1.57%), Rozellomycota (control:

3.14 ± 1.82%; treated: 2.54 ± 1.63%), and Blastocladiomycota (control: 2.47 ± 1.06%; treated: 1.17 ± 0.60%).

Considering the analyses conducted on the carposphere, specifically focusing on bacteria and employing a class-level analysis, it was possible to identify seven classes present at a percentage greater than 0.5% (Figure 6A). The primary class identified was Proteobacteria, constituting over 80% of the identified classes alone (control: 84.37 ± 3.12%; treated: 83.91 ± 2.26%). It was followed by the phyla Sphingobacteriia (control: 5.48 ± 1.63%; treated: 5.60 ± 2.18%), Actinobacteria (control: 3.07 ± 1.39%; treated: 3.09 ± 0.75%), Bacilli (control: 1.94 ± 0.61%; treated: 1.73 ± 0.69%), Bacteroidia (control: 0.93 ± 0.38%; treated: 1.21 ± 0.43%), Clostridia (control: 0.92 ± 0.42%; treated: 1.03 ± 0.26%), and Cytophagia (control: 0.88 ± 0.20%; treated: 0.90 ± 0.24%). However, no significant differences were identified regarding the presence of these classes.

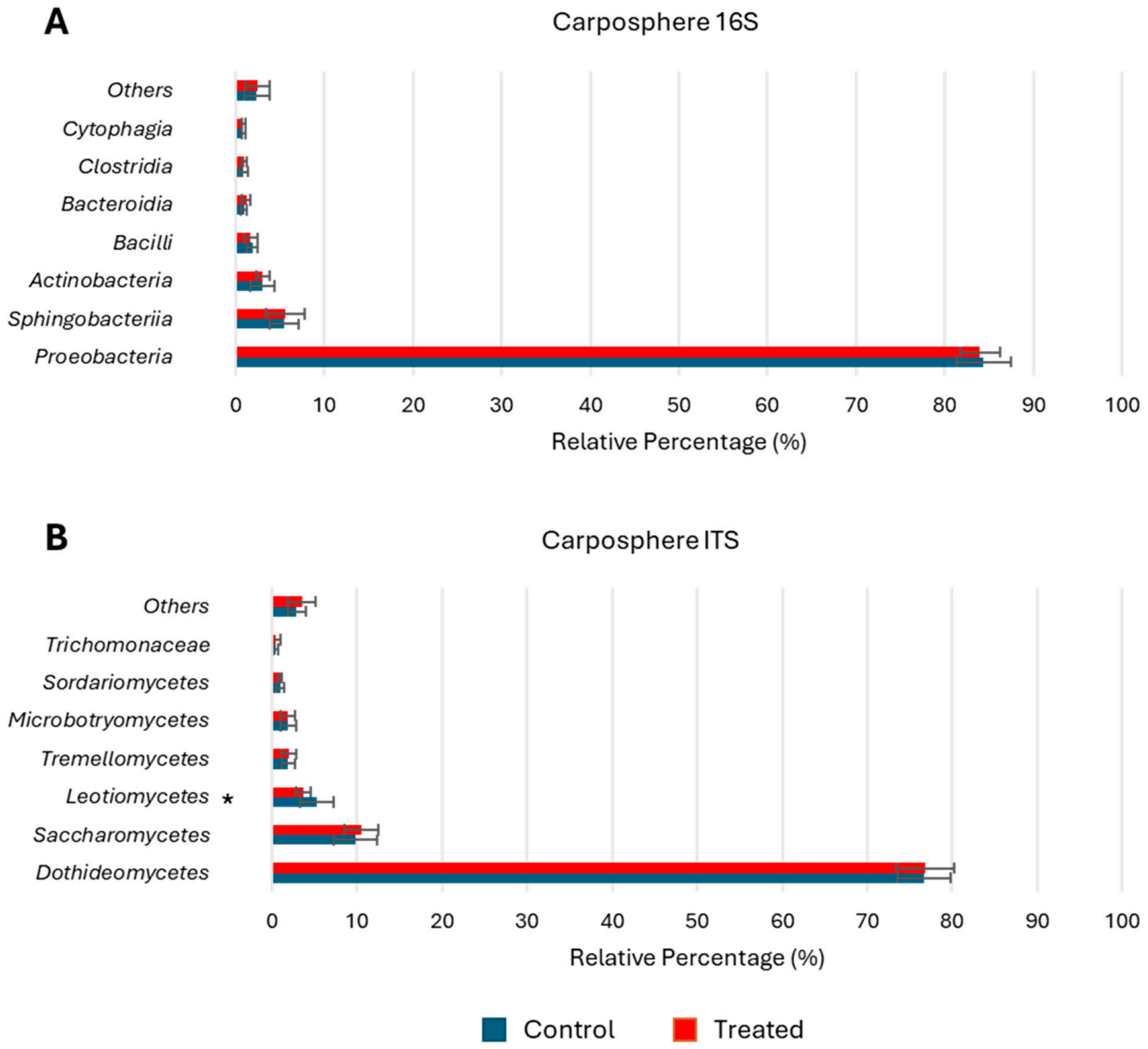

Figure 6. (**A**) Bacterial (16S) classes identified above 0.5% in the plant carposphere of both control and treated plants; (**B**) fungal (ITS) classes identified above 0.5% in the plant carposphere of both control and treated plants. Statistical analysis (*T* test) is reported (* $p < 0.05$, ** $p < 0.01$).

Considering the fungal classes present on the carposphere, once again, seven classes were identified at a percentage greater than 0.5% (Figure 6B). Dothideomycetes were found to constitute over 76% of the classes alone (control: 76.70 ± 3.08%; treated: 76.86 ± 3.38%), followed by Saccharomycetes (control: 9.85 ± 2.53%; treated: 10.54 ± 2.04%), Leotiomycetes (control: 5.25 ± 1.99%; treated: 3.73 ± 0.82%), Tremellomycetes (control: 1.93 ± 0.75%; treated: 2.02 ± 0.86%), Microbotryomycetes (control: 1.89 ± 0.93%; treated: 1.82 ± 0.84%), Sordariomycetes (control: 0.97 ± 0.49%; treated: 1.00 ± 0.19%), and Trichomonaceae (control: 0.49 ± 0.30%; treated: 0.51 ± 0.47%). In this case, only the Leotiomycetes were significantly more abundant ($p < 0.05$) in the control group.

To identify potential differences in detail, a more in-depth analysis was conducted considering the 15 principal genera identified from the analyses performed on the 16S and ITS of the carposphere. In the case of bacteria (Figure 7A), the 15 principal genera were found to be *Cupriavidus* (control: 43.99 ± 4.43%; treated: 43.98 ± 7.58%), which emerged as the primary genus, followed by *Undibacterium* (control: 26.66 ± 3.53%; treated: 28.10 ± 4.03%), *Pedobacter* (control: 15.53 ± 3.26%; treated: 17.19 ± 2.71%), *Burkholderia* (control: 7.07 ± 3.42%; treated: 5.81 ± 1.93%), *Sphingomonas* (control: 4.20 ± 2.46%; treated: 2.78 ± 2.16%), *Hymenobacter* (control: 0.95 ± 0.61%; treated: 0.61 ± 0.46%), *Massilia* (control: 0.45 ± 0.14%; treated: 0.37 ± 0.17%), *Methylobacterium* (control: 0.22 ± 0.13%; treated: 0.20 ± 0.09%), *Meiothermus* (control: 0.24 ± 0.18%; treated: 0.25 ± 0.17%), *Gluconobacter* (control: 0.18 ± 0.07%; treated: 0.19 ± 0.07%), *Pseudomonas* (control: 0.11 ± 0.03%; treated: 0.17 ± 0.06%), *Bacillus* (control: 0.11 ± 0.03%; treated: 0.14 ± 0.02%), *Lactobacillus* (control: 0.10 ± 0.05%; treated: 0.11 ± 0.07%), *Flexibacter* (control: 0.10 ± 0.01%; treated: 0.10 ± 0.01%), and *Staphylococcus* (control: 0.10 ± 0.02%; treated: 0.08 ± 0.05%). In this case, the genra *Pseudomonas* ($p < 0.01$) and *Bacillus* ($p < 0.05$) were significantly more abundant in the treatment group compared to the control. Conducting the same analysis for fungal genera (Figure 7B), the primary genus was *Cladosporium* (control: 23.23 ± 4.02%; treated: 24.64 ± 7.35%), followed by *Alternaria* (control: 19.84 ± 4.84%; treated: 17.39 ± 4.87%), *Hanseniaspora* (control: 19.38 ± 0.87%; treated: 17.91 ± 1.98%), *Aureobasidium* (control: 8.99 ± 2.53%; treated: 8.47 ± 2.87%), *Metschnikowia* (control: 7.66 ± 1.01%; treated: 8.56 ± 0.75%), *Filobasidium* (control: 5.24 ± 0.32%; treated: 5.84 ± 0.92%), *Botrytis* (control: 4.26 ± 1.11%; treated: 5.28 ± 0.93%), *Glomerella* (control: 2.48 ± 0.39%; treated: 2.74 ± 0.42%), *Davidiella* (control: 2.19 ± 0.17%; treated: 1.94 ± 0.68%), *Aspergillus* (control: 1.58 ± 0.24%; treated: 1.78 ± 0.58%), *Sporobolomyces* (control: 1.52 ± 0.13%; treated: 1.72 ± 0.41%), *Epicoccum* (control: 1.38 ± 0.05%; treated: 1.38 ± 0.28%), *Rhodotorula* (control: 0.94 ± 0.47%; treated: 1.00 ± 0.38%), *Lewia* (control: 0.73 ± 0.16%; treated: 0.75 ± 0.09%), and *Cystofilobasidium* (control: 0.60 ± 0.10%; treated: 0.62 ± 0.10%). In this case, the genus *Hanseniaspora* was significantly more abundant in the control ($p < 0.05$), while *Metschnikowia* and *Botrytis* were significantly more abundant in the treatment group ($p < 0.05$)

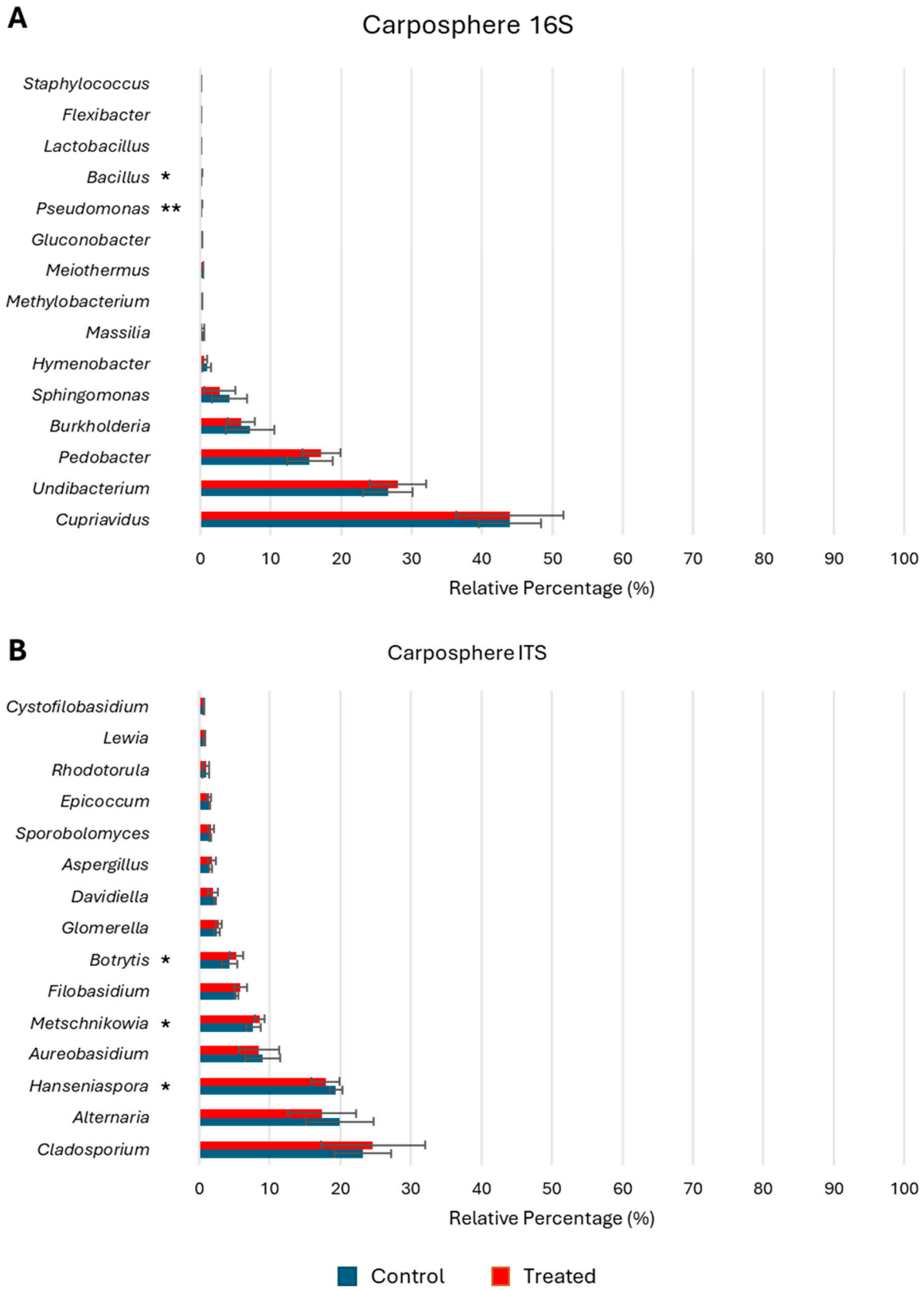

Figure 7. (**A**) Fifteen mains bacterial (16S) genera identified in the plant rhizosphere of both control and treated plants; (**B**) fifteen main fungal (ITS) genera identified in the plant rhizosphere of both control and treated plants. Statistical analysis (*T* test) is reported (* $p < 0.05$, ** $p < 0.01$).

4. Discussion

From the observed results, it has been discerned that the use of irrigation water with added NaCl succeeded in eliciting significant differences in the soil compared to the control plots irrigated with rainwater. Specifically, both the pH and salinity values in the control soil plots closely resembled the actual soil characteristics identified throughout most of the Veneto region, the primary producer of the Glera grape variety. The control exhibited pH values around neutrality and non-saline soils, consistent with the predominant soil conditions. In contrast, the treated soil reflected salinity and alkalinity levels found in certain areas of the Po River delta [36]. These disparities resulted in significant differences in the yield-related parameters and the root system. The sugar content demonstrated a higher level in the control in comparison to the treated group; however, although the total yield was higher in the control, it was not statistically significant. In contrast, acidity levels, encompassing both total and malic acid, reached their peak in the treated group, subsequently influencing the pH. This observed pattern aligns with established findings in the literature, where salt stress typically leads to a reduced sugar content, resulting in increased acidity and decreased pH [37].

Furthermore, the trend suggested a better growth state of the control plants, with a greater number of shoots, leaves, total leaf area, and leaf area index, however with statistically non-significant differences. This suggests a mildly stressful impact on the general growth conditions of the plant caused by the induced salinity conditions. This is crucial to compare the microbiota of the two treatments solely in relation to the differences induced by varying salinity levels. If the plant were in a state of decline, it would not be possible to make a proper comparison. This observation supports the higher sugar content in the control group, emphasizing the importance of canopy characteristics in facilitating photosynthesis, metabolite accumulation, and overall grapevine growth [38].

The development of thick roots was reduced by the induction of salt stress (treated vines). This observation holds significance as vines with a decreased presence of thick roots tend to exhibit a reduced exploration of the soil surrounding the trunk. Consequently, this limited exploration can pose challenges for nutrient and water uptake by the vines. These factors are pertinent considerations for the long-term perspective and overall resilience of the grapevines. The observed alteration in root development under salt stress underscores potential challenges in nutrient and water availability, as well as the storage of key metabolites, emphasizing the need to account for these aspects in the holistic assessment of grapevine health and performance over time [39].

In addition to the modification of grapevine growth, as observed in other studies [40,41], the results obtained for the induced salt stress conditions revealed significant differences induced in the root-associated microbiota due to salinity. Indeed, salinity stress conditions induce elevated osmotic pressures that exert a selective pressure on microorganisms unable to adapt [42], simultaneously favouring the development of salt-tolerant and obligate microorganisms, leading to their dominance [43]. This often results in a depletion of microbial biodiversity [44]. In this regard, this study aimed to assess these differences, given the importance of these microbial populations concerning their influence on the normal nutrient cycling, disease resistance, yield, and aromatic profile of wines [45]. The evaluation focused on identifying the microorganisms that have most effectively adapted to saline conditions and understanding their contribution to soil diversity and overall plant health [46]. This knowledge is not only crucial for comprehending the vineyard ecosystem but also holds practical implications for optimizing soil health, enhancing grapevine resilience, and potentially shaping the terroir of wines.

By evaluating the effects induced through the analysis of alpha diversity, it was found that the Shannon diversity values in saline soils were significantly lower than those in low-salinity soils as previously reported in the literature [47]. Saline conditions can be challenging for many organisms, and some may thrive while others struggle or perish. A decrease in Shannon diversity indicates a shift in the composition of the microbial

community, with certain species dominating and others being less represented, as it was in our study.

With regard to the analysis of beta diversity, the fungal and bacterial populations did not differ significantly based on treatment, both at the carposphere or rhizosphere levels. This suggests a limited overall impact on the biodiversity of these microbial populations. On the contrary, it was possible to observe differences in individual species.

By analysing in detail the differences induced on the rhizosphere, evaluating the taxonomic differences at the phyla level, considering the fungi kingdom, *Ascomycota* were present at higher percentages in the treated vines. This finding is supported by the literature, in which higher abundances of *Ascomycota* were observed in extremely saline soils. In fact, numerous studies have shown that fungal communities are influenced by soil salinity [48,49], where the relative abundance of the fungal phylum *Ascomycota* is increased [45,50,51]. Also, *Mortierellomycota* and *Glomeromycota* increased in the treated vines, in accordance with previous works [52,53]. In particular, *Glomeromycota* was identified as the keystone taxa in predicting the soil salinity soil fertility, and plant growth trait [54].

At the same time, analysing the bacterial phyla associated with the grapevine rhizosphere, in agreement with what was observed in previous studies [55], *Acidobacteria* and *Bacteroidetes* were higher in the saline soils, as in our trial. Changes in these phyla are interesting as some of the bacteria belonging to them have the potential to establish associations with other key taxa and also govern carbon dynamics in saline soils [56]. Also, *Chloroflexi*, *Firmicutes*, and *Nitrospirae* increased their presence in the treated conditions. These results are in accordance with the previous literature, and in particular, differences in *Firmicutes'* presence are of high importance, as this phyla is associated with the development of soil bioremediation for sustainable agriculture, a matter to be taken into account when talking about climate change and soil salinization [57,58]. In particular, among the phyla *Firmicutes*, the *Bacillus* genus was reported as tolerant to salt stress conditions [59], and its application proved useful for plant growth, producing industrially important enzymes and participating in the bioremediation of toxic chemicals [60].

Shifting to the berry microbiota and with regard to the fungi kingdom, the *Leotiomycetes* class showed a higher presence in the control than in the treated vines, as also observed by other authors [59]. More in detail, concerning the fungi carposphere genra, *Hanseniaspora* was more marked in the control. This is a key taxa, since apiculate yeasts of the genus *Hanseniaspora* are the main species present on mature grapes and play a significant role at the beginning of fermentation, producing enzymes and aroma compounds that expand the diversity of wine colour and flavour [61]. This is of great interest in a worldwide new scenario of soil salinization and oenological performance of wines. *Metschnikowia* spp. and *Botrytis* spp. were also found in the highest presence in the treated wines. Particularly noteworthy are *Metschnikowia* spp., which have been identified as prevalent in saline environments [61]. This genus stands out among non-*Saccharomyces* wine yeasts due to its extensive investigation, owing to its widespread presence and significant impact on winemaking. *Metschnikowia* spp. have been detected in various habitats such as grapevine phyllospheres, fruit flies, grapes, and wine fermentations, contributing to the resident microbiota of wineries and winemaking equipment. Its versatility lies in its capacity to coexist with other yeast species, notably *S. cerevisiae*, during initial wine fermentation stages, thereby influencing the synthesis of secondary metabolites to enhance the wine's sensory characteristics. *Metschnikowia* spp. exhibit moderate fermentation abilities, intriguing enzymatic activities related to aromatic and colour precursors and potential antimicrobial properties against spoilage yeasts and fungi. Consequently, this yeast has garnered attention as a valuable tool for enhancing wine quality [62].

Concerning the bacteria in the berry carposphere, at class level, no differences arose. Indeed, at genus level, both *Pseudomonas* and *Bacillus* were more abundant in the treated wines compared to the control. In this sense, *Pseudomonas* was reported to grow in saline environments, and its biochemical traits are associated with plant growth promotion [63,64]. *Pseudomonas* also dominated the bacterial community in canker-free grapevine tissues [65];

thus, it could be considered as evidence of the grapevine's health status. Concerning the *Bacillus* genus, due to their great metabolic/genomic background and spore formation, they have shown high resilience to salt stress enhancing the tolerance of plants to saline environments [66]. *Bacillus* is important in the grapevine sector being a biological controller of anthracnose [67], which is one of the economically important diseases in grapes, and other diseases, thus, correlating with the health status. Also, this genus plays a role in the microbiota of grape berries, influencing the fermentation process during winemaking. The presence of *Bacillus* in the grape microbiota in fact was reported to contribute to the distinctive microbial fingerprint of a vineyard, adding to the complexity and uniqueness of the resulting wine [37]. For instance, *Bacillus* could inhibit the malolactic bacterium, *Oenococcus oeni*, responsible for malolactic fermentation [68].

5. Conclusions and Remarks

The impact of salt stress on the grapevine rhizosphere and carposphere microbiota was evident, affecting various components. Specifically, indigenous yeasts exhibited increased presence under salt stress. This observation is critical, as the greater presence and biodiversity of these microorganisms can pose challenges for the proper control of the initial fermentation phases, yet they also represent a potential bioresource for identifying microorganisms suitable for indigenous fermentations in the future. Additionally, certain bacteria demonstrated higher abundance under salt stress, suggesting a potential protective role. These collective findings underscore the intricate dynamics of salt stress on both the microbial and physiological aspects of grapevine health, highlighting the importance of understanding and managing these factors in viticulture practices.

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References

1. Corwin, D.L. Climate change impacts on soil salinity in agricultural areas. *Eur. J. Soil Sci.* **2021**, *72*, 842–862. [CrossRef]
2. Ullah, A.; Bano, A.; Khan, N. Climate Change and Salinity Effects on Crops and Chemical Communication Between Plants and Plant Growth-Promoting Microorganisms Under Stress. *Front. Sustain. Food Syst.* **2021**, *5*, 618092. [CrossRef]
3. Shani, U.; Ben-Gal, A. Long-term response of grapevines to salinity: Osmotic effects and ion toxicity. *Am. J. Enol. Vitic.* **2005**, *56*, 148–154. [CrossRef]
4. Al-Taey, D.K.A.; Al-Ameer, A. A Effect of Salinity on the Growth and Yield of Grapes: A review. *IOP Conf. Ser. Earth Environ. Sci.* **2023**, *1262*, 042038. [CrossRef]
5. Elnashar, W.; Elyamany, A. Managing Risks of Climate Change on Irrigation Water in Arid Regions. *Water Resour. Manag.* **2023**, *37*, 2429–2446. [CrossRef]
6. Lee, J.L.; Huang, W.C. Impact of climate change on the irrigation water requirement in Northern Taiwan. *Water* **2014**, *6*, 3339–3361. [CrossRef]
7. Incoom, A.B.M.; Adjei, K.A.; Odai, S.N.; Akpoti, K.; Siabi, E.K. Impacts of climate change on crop and irrigation water requirement in the Savannah regions of Ghana. *J. Water Clim. Chang.* **2022**, *13*, 3338–3356. [CrossRef]
8. Austin, J.; Zhang, L.; Jones, R.N.; Durack, P.; Dawes, W.; Hairsine, P. Climate change impact on water and salt balances: An assessment of the impact of climate change on catchment salt and water balances in the Murray-Darling Basin, Australia. *Clim. Chang.* **2010**, *100*, 607–631. [CrossRef]
9. Colombani, N.; Osti, A.; Volta, G.; Mastrocicco, M. Impact of Climate Change on Salinization of Coastal Water Resources. *Water Resour. Manag.* **2016**, *30*, 2483–2496. [CrossRef]

10. Nielsen, D.L.; Brock, M.A. Modified water regime and salinity as a consequence of climate change: Prospects for wetlands of Southern Australia. *Clim. Chang.* **2009**, *95*, 523–533. [CrossRef]
11. Zarraonaindia, I.; Owens, S.M.; Weisenhorn, P.; West, K.; Hampton-Marcell, J.; Lax, S.; Bokulich, N.A.; Mills, D.A.; Martin, G.; Taghavi, S.; et al. The soil microbiome influences grapevine-associated microbiota. *mBio* **2015**, *6*, e02527-14. [CrossRef] [PubMed]
12. Griggs, R.G.; Steenwerth, K.L.; Mills, D.A.; Cantu, D.; Bokulich, N.A. Sources and Assembly of Microbial Communities in Vineyards as a Functional Component of Winegrowing. *Front. Microbiol.* **2021**, *12*, 673810. [CrossRef] [PubMed]
13. Xia, F.; Hao, H.; Qi, Y.; Bai, H.; Li, H.; Shi, Z.; Shi, L. Effect of Salt Stress on Microbiome Structure and Diversity in Chamomile (*Matricaria chamomilla* L.) Rhizosphere Soil. *Agronomy* **2023**, *13*, 1444. [CrossRef]
14. Mukhtar, S.; Mirza, B.S.; Mehnaz, S.; Mirza, M.S.; Mclean, J.; Malik, K.A. Impact of soil salinity on the microbial structure of halophyte rhizosphere microbiome. *World J. Microbiol. Biotechnol.* **2018**, *34*, 136. [CrossRef]
15. Miransari, M. *Arbuscular Mycorrhizal Fungi and Soil Salinity. Mycorrhizal Mediation of Soil: Fertility, Structure, and Carbon Storage*; Elsevier Inc.: Amsterdam, The Netherlands, 2017. [CrossRef]
16. Wang, B.; Wang, X.; Wang, Z.; Zhu, K.; Wu, W. Comparative metagenomic analysis reveals rhizosphere microbial community composition and functions help protect grapevines against salt stress. *Front. Microbiol.* **2023**, *14*, 1102547. [CrossRef]
17. Bettenfeld, P.; Cadena i Canals, J.; Jacquens, L.; Fernandez, O.; Fontaine, F.; van Schaik, E.; Courty, P.-E.; Trouvelot, S. The microbiota of the grapevine holobiont: A key component of plant health. *J. Adv. Res.* **2022**, *40*, 1–15. [CrossRef]
18. Garde-Cerdán, T.; Ancín-Azpilicueta, C. Contribution of wild yeasts to the formation of volatile compounds in inoculated wine fermentations. *Eur. Food Res. Technol.* **2006**, *222*, 15–25. [CrossRef]
19. Liu, D.; Zhang, P.; Chen, D.; Howell, K. From the Vineyard to the Winery: How Microbial Ecology Drives Regional Distinctiveness of Wine. *Front. Microbiol.* **2019**, *10*, 2679. [CrossRef] [PubMed]
20. Mian, G.; Cipriani, G.; Firrao, G.; Martini, M.; Ermacora, P. Genetic diversity of Actinidia spp. shapes the oomycete pattern associated with Kiwifruit Vine Decline Syndrome (KVDS). *Sci. Rep.* **2023**, *13*, 16449. [CrossRef]
21. Colautti, A.; Golinelli, F.; Iacumin, L.; Tomasi, D.; Cantone, P.; Mian, G. Triacontanol (long-chain alcohol) positively enhances the microbial ecology of berry peel in Vitis vinifera cv. 'Glera' yet promotes the must total soluble sugars content. *OENO One* **2023**, *57*, 477–488. [CrossRef]
22. Bolyen, E.; Rideout, J.R.; Dillon, M.R.; Bokulich, N.A.; Abnet, C.C.; Al-Ghalith, G.A.; Alexander, H.; Alm, E.J.; Arumugam, M.; Asnicar, F.; et al. Reproducible, interactive, scalable and extensible microbiome data science using QIIME 2. *Nat. Biotechnol.* **2019**, *37*, 852–857. [CrossRef] [PubMed]
23. Martin, M. Cutadapt removes adapter sequences from high-throughput sequencing reads. *EMBnet J.* **2011**, *17*, 10. [CrossRef]
24. Callahan, B.J.; McMurdie, P.J.; Rosen, M.J.; Han, A.W.; Johnson, A.J.A.; Holmes, S.P. DADA2: High-resolution sample inference from Illumina amplicon data. *Nat. Methods* **2016**, *13*, 581–583. [CrossRef] [PubMed]
25. Bokulich, N.A.; Kaehler, B.D.; Rideout, J.R.; Dillon, M.; Bolyen, E.; Knight, R.; Huttley, G.A.; Gregory Caporaso, J. Optimizing taxonomic classification of marker-gene amplicon sequences with QIIME 2's q2-feature-classifier plugin. *Microbiome* **2018**, *6*, 90. [CrossRef] [PubMed]
26. Quast, C.; Pruesse, E.; Yilmaz, P.; Gerken, J.; Schweer, T.; Yarza, P.; Peplies, J.; Glöckner, F.O. The SILVA ribosomal RNA gene database project: Improved data processing and web-based tools. *Nucleic Acids Res.* **2013**, *41*, 590–596. [CrossRef]
27. Nilsson, R.H.; Larsson, K.H.; Taylor, A.F.S.; Bengtsson-Palme, J.; Jeppesen, T.S.; Schigel, D.; Kennedy, P.; Picard, K.; Glöckner, F.O.; Tedersoo, L.; et al. The UNITE database for molecular identification of fungi: Handling dark taxa and parallel taxonomic classifications. *Nucleic Acids Res.* **2019**, *47*, D259–D264. [CrossRef]
28. Kandlikar, G.S.; Gold, Z.J.; Cowen, M.C.; Meyer, R.S.; Freise, A.C.; Kraft, N.J.B.; Moberg-Parker, J.; Sprague, J.; Kushner, D.J.; Curd, E.E. Ranacapa: An R package and shiny web app to explore environmental DNA data with exploratory statistics and interactive visualizations [version 1; referees: 1 approved, 2 approved with reservations]. *F1000Research* **2018**, *7*, 1734. [CrossRef]
29. McMurdie, P.J.; Holmes, S. Phyloseq: A bioconductor package for handling and analysis of high-throughput phylogenetic sequence data. *Pacific. Symp. Biocomput.* **2012**, *2011*, 235–246. [CrossRef]
30. Dixon, P. VEGAN, a package of R functions for community ecology. *J. Veg. Sci.* **2003**, *14*, 927–930. [CrossRef]
31. Colautti, A.; Civilini, M.; Contin, M.; Celotti, E.; Iacumin, L. Organic vs. conventional: Impact of cultivation treatments on the soil microbiota in the vineyard. *Front. Microbiol.* **2023**, *14*, 1242267. [CrossRef]
32. Falginella, L.; Gaiotti, F.; Belfiore, N.; Mian, G.; Lovat, L.; Tomasi, D. Effect of early cane pruning on yield components, grape composition, carbohydrates storage and phenology in Vitis vinifera L. cv. Merlot. *OENO One* **2022**, *56*, 19–28. [CrossRef]
33. Mian, G.; Celotti, E.; Falginella, L.; de Oliveria Cantão, F.R.; Belfiore, N. Effect of manure application timing on roots, canopy and must quality in Vitis vinifera "Merlot": A case study in Italy, North-East. *VITIS J. Grapevine Res.* **2022**, *61*, 87–92. [CrossRef]
34. Mian, G.; Colautti, A.; Belfiore, N.; Marcuzzo, P.; Tomasi, D.; Bell, L.; Celotti, E. Fertigation affects photosynthesis, modulation of secondary metabolism and sensory profiles of Vitis vinifera cv. "Schioppettino" withered grapes and wines. *Sci. Hortic.* **2024**, *328*, 112954. [CrossRef]
35. Mian, G.; Cipriani, G.; Saro, S.; Martini, M.; Ermacora, P. Potential of Different Actinidia Genotypes as Resistant Rootstocks for Preventing Kiwifruit Vine Decline Syndrome. *Horticulturae* **2022**, *8*, 627. [CrossRef]
36. ARPAV. La Carta della Reazione(pH) e della Salinità della Regione Veneto. 2020. Available online: https://www.arpa.veneto.it/temi-ambientali/suolo/file-e-allegati/documenti (accessed on 21 December 2023).

37. Tomasi, D.; Marcuzzo, P.; Nardi, T.; Lonardi, A.; Lovat, L.; Flamini, R.; Mian, G. Influence of Soil Chemical Features on Aromatic Profile of, *V. vinifera* cv. Corvina Grapes and Wines: A Study-Case in Valpolicella Area (Italy) in a Calcareous and Non-Calcareous Soil. *Agriculture* **2022**, *12*, 1980. [CrossRef]
38. Mian, G.; Nassivera, F.; Sillani, S.; Iseppi, L. Grapevine Resistant Cultivars: A Story Review and the Importance on the Related Wine Consumption Inclination. *Sustainability* **2022**, *15*, 390. [CrossRef]
39. Pereyra, G.; Pellegrino, A.; Ferrer, M.; Gaudin, R. How soil and climate variability within a vineyard can affect the heterogeneity of grapevine vigour and production. *OENO One* **2023**, *57*, 297–313. [CrossRef]
40. Chowdhury, N.; Marschner, P.; Burns, R. Response of microbial activity and community structure to decreasing soil osmotic and matric potential. *Plant Soil.* **2011**, *344*, 241–254. [CrossRef]
41. Elmajdoub, B.; Marschner, P. Responses of soil microbial activity and biomass to salinity after repeated additions of plant residues. *Pedosphere* **2015**, *25*, 177–185. [CrossRef]
42. Polonenko, D.R.; Mayfield, C.I.; Dumbroff, E.B. Microbial responses to salt-induced osmotic stress. *Plant Soil* **1986**, *92*, 417–425. [CrossRef]
43. Rath, K.M.; Rousk, J. Salt effects on the soil microbial decomposer community and their role in organic carbon cycling: A review. *Soil Biol. Biochem.* **2015**, *81*, 108–123. [CrossRef]
44. Zhang, L.; Tang, C.; Yang, J.; Yao, R.; Wang, X.; Xie, W.; Ge, A.-H. Salinity-dependent potential soil fungal decomposers under straw amendment. *Sci. Total Environ.* **2023**, *891*, 164569. [CrossRef] [PubMed]
45. Coller, E.; Cestaro, A.; Zanzotti, R.; Bertoldi, D.; Pindo, M.; Larger, S.; Albanese, D.; Mescalchin, E.; Donati, C. Microbiome of vineyard soils is shaped by geography and management. *Microbiome* **2019**, *7*, 140. [CrossRef] [PubMed]
46. Qin, Y.; Druzhinina, I.S.; Pan, X.; Yuan, Z. Microbially Mediated Plant Salt Tolerance and Microbiome-based Solutions for Saline Agriculture. *Biotechnol. Adv.* **2016**, *34*, 1245–1259. [CrossRef] [PubMed]
47. Yang, C.; Sun, J. Soil Salinity Drives the Distribution Patterns and Ecological Functions of Fungi in Saline-Alkali Land in the Yellow River Delta, China. *Front. Microbiol.* **2020**, *11*, 594284. [CrossRef] [PubMed]
48. Mohamed, D.J.; Martiny, J.B.H. Patterns of fungal diversity and composition along a salinity gradient. *ISME J.* **2011**, *5*, 379–388. [CrossRef] [PubMed]
49. Krishnamoorthy, R.; Kim, K.; Kim, C.; Sa, T. Changes of arbuscular mycorrhizal traits and community structure with respect to soil salinity in a coastal reclamation land. *Soil Biol. Biochem.* **2014**, *72*, 1–10. [CrossRef]
50. Kim, K.; Samaddar, S.; Chatterjee, P.; Krishnamoorthy, R.; Jeon, S.; Sa, T. Structural and functional responses of microbial community with respect to salinity levels in a coastal reclamation land. *Appl. Soil Ecol.* **2019**, *137*, 96–105. [CrossRef]
51. Yang, C.; Wang, X.; Miao, F.; Li, Z.; Tang, W.; Sun, J. Assessing the effect of soil salinization on soil microbial respiration and diversities under incubation conditions. *Appl. Soil Ecol.* **2020**, *155*, 103671. [CrossRef]
52. Valentin, D.N.; Voyron, S.; Soteras, F.; Iriarte, H.J.; Giovannini, A.; Lumini, E.; Lugo, M.A. Modeling geographic distribution of arbuscular mycorrhizal fungi from molecular evidence in soils of Argentinean Puna using a maximum entropy approach. *PeerJ* **2023**, *11*, e14651. [CrossRef]
53. Guo, H.N.; Huang, Z.J.; Li, M.Q.; Min, W. Response of soil fungal community structure and diversity to saline water irrigation in alluvial grey desert soils. *Appl. Ecol. Environ. Res.* **2020**, *18*, 4969–4985. [CrossRef]
54. Zhang, L.; Ge, A.H.; Tóth, T.; Yang, F.; Wang, Z.; An, F. Enrichment of keystone fungal taxa after flue gas desulphurization gypsum application drives reclamation of the saline-sodic soil. *Land Degrad. Dev.* **2023**, *34*, 2276–2287. [CrossRef]
55. Zhao, S.; Liu, J.J.; Banerjee, S.; Zhou, N.; Zhao, Z.Y.; Zhang, K.; Hu, M.; Tian, C. Biogeographical distribution of bacterial communities in saline agricultural soil. *Geoderma* **2020**, *361*, 114095. [CrossRef]
56. Banerjee, S.; Kirkby, C.A.; Schmutter, D.; Bissett, A.; Kirkegaard, J.A.; Richardson, A.E. Network analysis reveals functional redundancy and keystone taxa amongst bacterial and fungal communities during organic matter decomposition in an arable soil. *Soil Biol. Biochem.* **2016**, *97*, 188–198. [CrossRef]
57. Hou, Y.; Zeng, W.; Hou, M.; Wang, Z.; Luo, Y.; Lei, G.; Zhou, B.; Huang, J. Responses of the soil microbial community to salinity stress in maize fields. *Biology* **2021**, *10*, 1114. [CrossRef] [PubMed]
58. Hashmi, I.; Bindschedler, S.; Junier, P. Firmicutes. In *Beneficial Microbes in Agro-Ecology*; Academic Press: Cambridge, MA, USA, 2020. [CrossRef]
59. Mukhtar, S.; Mehnaz, S.; Mirza, M.S.; Mirza, B.S.; Malik, K.A. Diversity of Bacillus-like bacterial community in the rhizospheric and non-rhizospheric soil of halophytes (Salsola stocksii and Atriplex amnicola), and characterization of osmoregulatory genes in halophilic Bacilli. *Can. J. Microbiol.* **2018**, *64*, 567–579. [CrossRef] [PubMed]
60. Tsotetsi, T.; Nephali, L.; Malebe, M.; Tugizimana, F. Bacillus for Plant Growth Promotion and Stress Resilience: What Have We Learned? *Plants* **2022**, *11*, 2482. [CrossRef] [PubMed]
61. Martin, V.; Jose Valera, M.; Medina, K.; Boido, E.; Carrau, F. Oenological impact of the Hanseniaspora/Kloeckera yeast genus on wines—A review. *Fermentation* **2018**, *4*, 76. [CrossRef]
62. Vicente, J.; Ruiz, J.; Belda, I.; Benito-Vázquez, I.; Marquina, D.; Calderón, F.; Santos, A.; Benito, S. The genus metschnikowia in enology. *Microorganisms* **2020**, *8*, 1038. [CrossRef]
63. Lami, M.J.; Adler, C.; Caram-Di Santo, M.C.; Zenoff, A.M.; de Cristóbal, R.E.; Espinosa-Urgel, M.; Vincent, P.A. Pseudomonas stutzeri MJL19, a rhizosphere-colonizing bacterium that promotes plant growth under saline stress. *J. Appl. Microbiol.* **2020**, *129*, 1321–1336. [CrossRef]

64. Elabed, H.; González-Tortuero, E.; Ibacache-Quiroga, C.; Bakhrouf, A.; Johnston, P.; Gaddour, K.; Blázquez, J.; Rodríguez-Rojas, A. Seawater salt-trapped Pseudomonas aeruginosa survives for years and gets primed for salinity tolerance. *BMC Microbiol.* **2019**, *19*, 142. [CrossRef] [PubMed]
65. Niem, J.M.; Billones-Baaijens, R.; Stodart, B.; Savocchia, S. Diversity Profiling of Grapevine Microbial Endosphere and Antagonistic Potential of Endophytic Pseudomonas Against Grapevine Trunk Diseases. *Front. Microbiol.* **2020**, *11*, 477. [CrossRef] [PubMed]
66. Ibarra-Villarreal, A.L.; Gándara-Ledezma, A.; Godoy-Flores, A.D.; Herrera-Sepúlveda, A.; Díaz-Rodríguez, A.M.; Parra-Cota, F.I.; Santos-Villalobos, S.d.L. Salt-tolerant Bacillus species as a promising strategy to mitigate the salinity stress in wheat (Triticum turgidum subsp. durum). *J. Arid Environ.* **2021**, *186*, 104399. [CrossRef]
67. Sawant, I.S.; Wadkar, P.N.; Rajguru, Y.R.; Mhaske, N.H.; Salunkhe, V.P.; Sawant, S.D.; Upadhyay, A. Biocontrol potential of two novel grapevine associated Bacillus strains for management of anthracnose disease caused by Colletotrichum gloeosporioides. *Biocontrol Sci. Technol.* **2016**, *26*, 964–979. [CrossRef]
68. Bae, S.; Fleet, G.H.; Heard, G.M. Occurrence and significance of Bacillus thuringiensis on wine grapes. *Int. J. Food Microbiol.* **2004**, *94*, 301–312. [CrossRef]