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Root microbiome diversity and structure of the Sonoran desert buffelgrass (*Pennisetum ciliare* L.)

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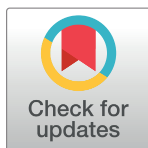

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Citation: Jara-Servin A, Silva A, Barajas H, Cruz-Ortega R, Tinoco-Ojanguren C, Alcaraz LD (2023) Root microbiome diversity and structure of the Sonoran desert buffelgrass (*Pennisetum ciliare* L.). PLoS ONE 18(5): e0285978. https://doi.org/10.1371/journal.pone.0285978

Editor: Marcela Pagano, Universidade Federal de Minas Gerais, BRAZIL

Received: February 1, 2023

Accepted: April 28, 2023

Published: May 19, 2023

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Data Availability Statement: All data used for this study can be accessed in the NCBI Sequence Read Archive (SRA) database under the Project ID PRJNA879420. All figures, tables, and datasets can be accessed in FigShare (https://doi.org/10.6084/m9.figshare.c.6605350.v2).

Funding: Consejo Nacional de Ciencia y Tecnología 765278 to Angélica Jara-Servin. Dirección General de Asuntos del Personal Académico, Universidad Nacional Autónoma de México, DGAPA-PAPIIT-UNAM IN212914 to Clara Tinoco-Ojanguren.

Abstract

Buffelgrass (*Pennisetum ciliare*) is an invasive plant introduced into Mexico's Sonoran desert for cattle grazing and has converted large areas of native thorn scrub. One of the invasion mechanisms buffelgrass uses to invade is allelopathy, which consists of the production and secretion of allelochemicals that exert adverse effects on other plants' growth. The plant microbiome also plays a vital role in establishing invasive plants and host growth and development. However, little is known about the buffelgrass root-associated bacteria and the effects of allelochemicals on the microbiome. We used 16S rRNA gene amplicon sequencing to obtain the microbiome of buffelgrass and compare it between samples treated with root exacknudates and aqueous leachates as allelochemical exposure and samples without allelopathic exposure in two different periods. The Shannon diversity values were between H' = 5.1811–5.5709, with 2,164 reported bacterial Amplicon Sequence Variants (ASVs). A total of 24 phyla were found in the buffelgrass microbiome, predominantly *Actinobacteria*, *Proteobacteria*, and *Acidobacteria*. At the genus level, 30 different genera comprised the buffelgrass core microbiome. Our results show that buffelgrass recruits microorganisms capable of thriving under allelochemical conditions and may be able to metabolize them (*e.g.*, *Planctomicrobium*, *Aurantimonas*, and *Tellurimicrobium*). We also found that the community composition of the microbiome changes depending on the developmental state of buffelgrass (p = 0.0366; ANOSIM). These findings provide new insights into the role of the microbiome in the establishment of invasive plant species and offer potential targets for developing strategies to control buffelgrass invasion.

1. Introduction

Introducing exotic species to a new environment can alter the ecosystem and decrease biological diversity. Invasive species can transform ecosystem processes over spatial and temporal

Dirección General de Asuntos del Personal Académico, Universidad Nacional Autónoma de México, DGAPA-PAPIIT-UNAM IN221420 to Luis D. Alcaraz. The funders had no role in the study design, data collection, analysis, decision to publish, or manuscript preparation.

Competing interests: I have read the journal's policy and the authors of this manuscript have the following competing interests: LDA is an Academic Editor for this journal. There are no other competing interests. This does not alter our adherence to PLOS ONE policies on sharing data and materials.

scales, with various degrees of impact [1]. In many cases, invasive species have higher values of resource-acquisition traits, larger sizes, and higher growth rates, implying larger pool sizes of nutrients that allow them to outcompete native species [1]. Buffelgrass (*Pennisetum ciliare*, *Cenchrus ciliaris*) has been North America's most noxious invasive plant since its introduction from East Africa in the 1930s [2]. The germination, establishment, and subsequent seedling growth of buffelgrass depend on soil moisture and environmental temperature conditions [3]. Buffelgrass is preferred because of its high germinability, easy establishment, high seedling production and vigor, fast growth rate, and ability to withstand fires because of its massive root system [3–6]. Buffelgrass individual tussocks and their long lifespan allow them to resprout from established tufts following a fire [5]. Fire also temporarily increases available phosphorus levels in the soil, which are rapidly exploited by buffelgrass [7].

In northwestern Mexico, large tracts of desert and thorn scrub have been converted to buffelgrass pasture to improve rangelands for cattle production [8]. Buffelgrass coverage estimates are 53% of the Sonora state and 12% of Mexico [9]. In the Sonoran Desert, buffelgrass invasion can cause a decrease by 53%–73% of the number of native plant species compared to undisturbed thorn scrub, which has a higher species diversity and is commonly dominated by Fabaceae and Cactaceae [8]. In contrast, thorny legume trees such as *Acacia cochliacantha* and *Acacia farnesiana* dominate the succession of abandoned pastures in the thorn scrub of Mexico [10], and very few native species can regenerate in the ranges [8]. The approaches taken to control buffelgrass growth include applying herbicides, manual removal, prescribed burning, and controlled animal grazing, but controlling buffelgrass worldwide is still an urgent matter [5].

One of the mechanisms proposed as a strategy for invasive species control is allelopathy [8, 11], which is based on the active synthesis and release of allelochemicals (secondary metabolites) by one plant, influencing the growth of a recipient plant, regardless of resource availability [12, 13]. The production of biochemicals that natives in the invaded range do not produce plays in favor of the invasive species since these chemicals may affect native species that lack a coevolutionary-based tolerance [14, 15]. Allelopathy plays a vital role in invasion, vegetation patterning, the exclusion of associated species, and reduced plant productivity [16]. Previous reports have shown buffelgrass allelopathy through root exudates of 3-month-old individuals, decreasing the growth and germination of various species such as *Chrysopogon aucheri*, *Hyparrhenia rufa*, *Bothriochloa pertusa*, *Panicum antidotale*, *Setaria italica*, and *Pennisetum americanum* [4]. Buffelgrass leachates and root exudates obtained from 3-month-old buffelgrass plants and used to water *Brassica campestris*, *Lactuca sativa*, and *Setaria italica* provoked a reduction in radicle growth due to allelopathy [17]. The germination rates of these three species also decreased, but only when the leachates and exudates were concentrated through evaporation [18]. The decrease in wild species could result from the amount of forage buffelgrass produced, which is 4 to 10 times greater than the production of native species, leading to possible nutrient depletion, especially of N, from the soils it grows on [3]. Such characteristics may explain, to a certain degree, the observed self-declination of pastures of *Pennisetum ciliare* [3, 18].

Various approaches have been made to describe the chemical identity of buffelgrass allelochemicals affecting the growth of many plant species. So far, the identified compounds are phenolic compounds, including *p*-OH-benzoic acid, *p*-coumaric acid, caffeic acid, vanillic acid, ferulic acid, syringic acid, and gentisic acid [17]. Although phenolic compounds have a short half-life in soils [19, 20], their reversibly sorbed fractions contribute to the pool available for allelopathic interactions [19]. Hence, compounds such as ferulic acid, *p*-coumaric acid, *p*-OH-benzoic acid, and vanillic acid could accumulate in the soil and impede the germination and growth of susceptible species [17]. However, the fate and persistence of phytotoxins in soil are unclear, as are their phytotoxicity or biodisposition by microbes [17]. The mutualistic

association formed by the invader is presumed to contribute to a competitive advantage over native species [21, 22], and plants can impact their associated microbiomes as an adaptation strategy when confronted with biotic and abiotic challenges [15, 23, 24]. Moreover, allelopathic plants release compounds that might alter the composition of the microbial community, recruiting microorganisms involved in the metabolization of the allelochemicals produced [25, 26]. Hence, understanding the rhizosphere microbiomes of invasive plants and the elements that influence the recruitment of microorganisms could be particularly valuable for understanding the factors promoting plant invasiveness and the following impacts on the ecosystems. So far, there are no reports of the buffelgrass rhizosphere microbiome composition and the effects of its allelochemicals on the microbial communities associated with its roots.

This work investigated the buffelgrass root microbiome and its compositional changes under allelochemical exposure. Microbiome changes were tested at two-time points using plants exposed to buffelgrass root exudates, watered with leachates from the aerial part of buffelgrass, and with a distilled water regime. This experimental design allowed us to describe the buffelgrass microbiome throughout buffelgrass growth, obtain a core microbiome, and determine the effect that allelochemicals may have on taxa comprising the rhizosphere microbiome.

2. Methods

2.1 Seed and soil sampling

Pennisetum ciliare seeds were collected from five plants at the end of the summer (September 2013) from induced pastures at Rancho Diamante (28˚ 41' N, 110˚ 15' W), Sonora State, Mexico, where thorn scrub is the original vegetation. Seeds were stored in a paper bag and transported to the laboratory. We also collected soil samples from the same location for further plant growth in the greenhouse. We collected samples of Buffelgrass *Cenchrus ciliaris* (*Penisetum ciliare*) from a location where we did not require special permissions, beyond the land owner. The land owner permitted us to collect the samples. It is important to note that while Buffelgrass is native to Tropical Africa and Asia, it is considered an invasive species in North America. However, it is not classified as an endangered or protected species and, therefore, does not require special permission for collection.

2.2 Plant growth under allelopathic conditions

We removed the seed covers of *P. ciliare* seeds for germination as naked caryopses have higher and more uniform germination [27]. Caryopses were germinated in Petri dishes with 2% agar in a growth chamber Biotronette (20.3L, 25˚C, 12-h photoperiod, 92 $\mu molm^{-2}s^{-1}$). Subsequently, 2-day-old seedlings were transferred to containers with peat moss, which grew for 70 days. The water regime consisted of watering with Hoagland solution every two weeks. After that, 70-day-old plants with a height of at least 10 cm were transplanted into curved PVC tubes containing sterilized silica sand and soil (at a proportion of 1:1) from Rancho Diamante. Plants in PVC tubes received a standard water regime (consisting of 25 mL of sterilized deionized water six times per week). They were divided into three different treatments: (1) root exudates, (2) aqueous leachates of the aerial plant part, and (3) control (Fig 1, https://doi.org/10.6084/m9.figshare.c.6605350.v2). The exudate treatment consisted of a second buffelgrass plant at the opposite end of the curved PVC tube, so the roots of the 70-day-old individual would be under the influence of the second buffelgrass plant, allowing to evaluate the effect of the root exudates on the rhizosphere microbiome. Physical interaction of the roots was avoided by installing a net at the middle of the tube. In the leachate treatment, one end of the PVC tube was planted with the 70-day-old buffelgrass, while the other was left unplanted. Plants under this treatment were watered with aqueous leachates extracted from buffelgrass green leaves

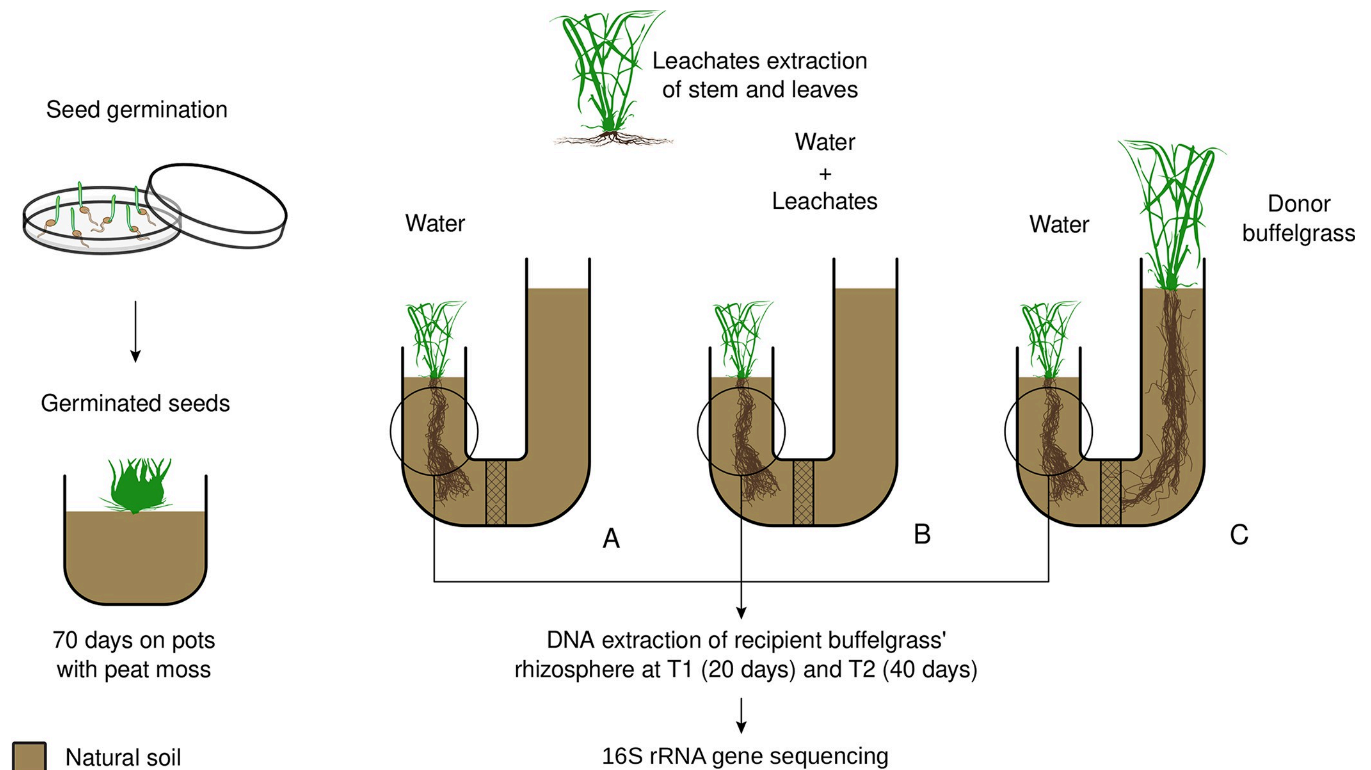

Fig 1. Experimental setup. Buffelgrass seeds germinated in a chamber and, after two days, transferred to peat moss-filled pots. After 70 days, the plants were transplanted into PVC pots containing silica and soil (1:1) from Rancho Diamante, Sonoran Desert. The plants were then divided into three groups, each with four replicates. Group (A) was watered with sterilized distilled water, Group (B) was watered with a solution of aqueous leachates from leaves and stems, and Group (C) were watered with distilled water but exposed to root exudates from another buffelgrass plant. Sampling was done at two different periods, 20 and 40 days, with two replicates from each group being analyzed at each time.

https://doi.org/10.1371/journal.pone.0285978.g001

and stems instead of water, but according to the volume and periodicity of the standard water regime. The control treatment consisted of only one end of the PVC planted with buffelgrass with the standard water regime. No Hoagland solution was added during the allelochemical treatments. The leachates were obtained as previously described [28] by collecting green leaves and stems of buffelgrass adult plants in Rancho El Diamante. Leaves were dried for 24 h at 24˚C and submerged in distilled water 1% (g/v) for 3 h, followed by filtering through Whatman #4 filtering paper to simulate rainfall. Two soil samples from Rancho Diamante were left unplanted and subjected to no treatment. These soil samples allowed us to determine the soil microbiome of the region without the influence of growing buffelgrass plants and the effect that buffelgrass has on the recruited microorganisms.

2.3 Rhizosphere sample collection

Rhizosphere samples were obtained at two different time points of growth from the PVC tubes: after 20 days (T1) and after 40 days (T2). In both cases, the roots were separated from the remaining parts of the plant to obtain the rhizosphere. Rhizosphere separation involved removing the loose soil and subsequently washing and submitting the roots to ultrasound in 1X PBS buffer (137 mM NaCl; 2.7 mM KCl; 10 mM Na_2HPO_4; 1.8 mM KH_2PO_4). All pellets obtained from buffelgrass rhizospheres were recovered through centrifugation (50-mL tubes, 1,300 g, 10 min) and kept at -80˚C until further analysis and DNA extraction.

2.4 Metagenomic DNA processing and 16S rRNA gene sequencing

The metagenomic DNA of soil samples and all the rhizosphere pellets was extracted using the Mobio PowerSoil DNA extraction kit (MoBio, Carlsbad, CA, United States) following the manufacturer's instructions, with minor modifications, heating the C6 elution solution to 60˚C before the elution step to increase the DNA yield. Two 30 μL elution processes were performed during the same spin filter.

The 16S rRNA gene amplification was performed in duplicates, following the Illumina Ⓡ MiSeq protocol for 16S metagenomic sequencing library preparation (Illumina 2013). The primer pair used for the PCR amplification was 341F/805R (targeting the V3–V4 regions), with the Illumina sequencing adaptors in 5′ overhangs (341F: 5′ -CCTACGGGNGGCWGCAG-3′; 805R: 5′ - ACTACHVGGGTATCTAATCC 3′). The PCR reaction mixture consisted of 2 μL buffer, 1.2 μL of each primer (5 μM), 3 μL enhancer, 1.6 μL dNTPs (2.5 mM), 0.6 μL Mg2S04 (1.5μM), 9.2 μL PCR-grade water, 0.16 μL Pfx polymerase (0.02 U/μL) (Invitrogen, Thermo Fisher Scientific, Waltham, MA), and 2 μL DNA template, adding up to a final volume of 20 μL.

The PCR conditions were as follows: initial 95˚C for 3 min, then 25 cycles at 94˚C for 5 s, followed by 68˚C for 30 s. The PCR products were pooled and purified with the SV Wizard PCR purification kit (Promega, Madison, WI). All samples were sequenced on the Illumina Ⓡ MiSeq platform (2 × 300 paired-end) at the University Unit of Massive Sequencing and Bioinformatics Facilities of the Biotechnology Institute, UNAM Mexico.

2.5 16S rRNA gene amplicon sequence analysis

The detailed protocol and bioinformatic methods used to process and analyze the 16S rRNA gene amplicon sequences are available on GitHub (https://github.com/genomica-fciencias-unam/buffelgrass). Briefly, all amplicon libraries were quality-checked using Dada2 [29], the first 17 bp were removed, and the sequences were trimmed to 250 bp. Only the forward reads were used since the quality profiles of the reverse reads were poor. All amplicon sequences were processed to obtain Amplicon Sequence Variants (ASV) using DADA2 (v. 1.10.1) [29] to denoise and remove chimeras. At the species level, taxonomy was assigned using the Silva database (v. 138) [30]. A phylogenetic tree was constructed using FastTreeMP [31] (S1 Dataset, https://doi.org/10.6084/m9.figshare.c.6605350.v2).

2.6 Diversity and statistical analysis

The α- and β-diversity of all samples were calculated using the phyloseq [32], ggplot2 [33], vegan [34], and R default packages [35]. We measured taxonomic α-diversity using Observed, Shannon, and Simpson diversity indices. Hierarchical clustering was performed using the hclust method on an unweighted UniFrac distance matrix [36]. The ASVs were clustered at the various taxonomic levels to perform abundance comparisons of the three treatments. Core microbiomes were analyzed using the upset function; ß-diversity was analyzed through a constrained analysis of principal coordinates (CAP) on an unweighted UniFrac distance matrix, based on the obtained ASV abundances per treatment; the clustering was evaluated through the ANOSIM statistical function [37]. Differential ASV abundances comparing treatments were calculated using DESeq2 of the R package [38]. Detailed statistical and bioinformatic methods are available on FigShare (https://doi.org/10.6084/m9.figshare.c.6605350.v2).

3. Results

We sequenced 686,078 paired-end reads, with a mean of 49,005.57 ± 10,041 sequences per sample (n = 8). We then clustered them into 2,164 Amplicon Sequence Variants (ASVs; 100%

Table 1. Alpha diversity for buffelgrass microbiomes.

Sample	Observed	Shannon	Simpson
C1_T1	308	5.350832	0.9934471
C2_T1	349	5.466695	0.9942502
C1_T2	270	5.268196	0.9935561
C2_T2	262	5.225775	0.9928567
E1_T1	291	5.298459	0.9932077
E2_T1	342	5.468742	0.9943344
E1_T2	384	5.570910	0.9947818
E2_T2	332	5.464093	0.9945279
L1_T1	126	4.433554	0.9849328
L2_T1	538	5.871980	0.9957321
L1_T2	262	5.181120	0.9923309
L2_T2	282	5.291607	0.9935689
S1	297	5.361435	0.9938688
S2	295	5.331116	0.9935597

Values calculated for Observed, Shannon, and Simpson diversity indices. C is for controls; E exudates; L leachate; S soil; T is for time.

https://doi.org/10.1371/journal.pone.0285978.t001

identity 16S rRNA gene OTUs) and described diversity by Observed ASVs, Simpson, and Shannon diversity indices (Table 1). The Shannon index (*H'*) was used to evaluate the **α**-diversity of our samples. Overall, the samples maintained a similar diversity (*H'* = 5.1811–5.5709), regardless of treatment or period, except for the outliers, corresponding to samples of the leachate treatment at the first period (*H'* = 4.4335 and *H'* = 5.8719). The diversity increased slightly with the development of buffelgrass treated with exudates (from *H'* = 5.2984 and 5.4687 in the first period to *H'* = 5.5709 and 5.4640 in the second one). On the contrary, control samples showed a diversity reduction as the experiment advanced (*H'* = 5.3508 and 5.4666 in the first period to *H'* = 5.2681 and 5.2257 in the second one). Species dominance, evaluated through the Simpson index (*D*), followed the same pattern as the one described for diversity, where the value for the exudate treatment was higher at the end of the experiment (*D* = 0.9947 and 0.9945) than at the beginning (*D* = 0.9932 and 0.9943), as opposed to the increase seen from the first to the second period in samples from the control treatment (*D* = 0.9934 and 0.9942 to *D* = 0.9935 and 0.9928). Soil samples had Shannon diversity (*H'* = 5.361425 and 5.331116) and Simpson (*D* = 0.9938688 and 0.9935597) indices not differing from the treatment values.

We detected 24 phyla in all our samples. Eleven phyla were ubiquitous and abundant: *Actinobacteria*, *Proteobacteria*, *Acidobacteria*, *Planctomycetes*, *Gemmatimonadetes*, *Chloroflexi*, *Verrucomicrobia*, *Bacteroidetes*, *Firmicutes*, *Cyanobacteria*, and *Armatimonadetes* (Fig 2, https://doi.org/10.6084/m9.figshare.c.6605350.v2). The heatmap also revealed differences in the microbiome composition among different periods. For example, *WPS-2* and *Hydrogenedentes*, whose abundances increased in exudates from T1 (relative abundance values of *WPS-2* = 0.00; *Hydrogenedentes* = 2.05E-05) to T2 (*WPS-2* = 7.29E-04; *Hydrogenedentes* = 2.64E-04). On the contrary, the abundance of *Deinococcus-Thermus* decreased from T1 (1.16E-04) to T2 (0.00) in the same treatment. The abundance of the phylum *Fibrobacteres* increased over time both in the exudate and the leachate treatments. Complete taxonomic annotation and abundances are available as Supplementary Material (S1 and S2 Tables, https://doi.org/10.6084/m9.figshare.c.6605350.v2).

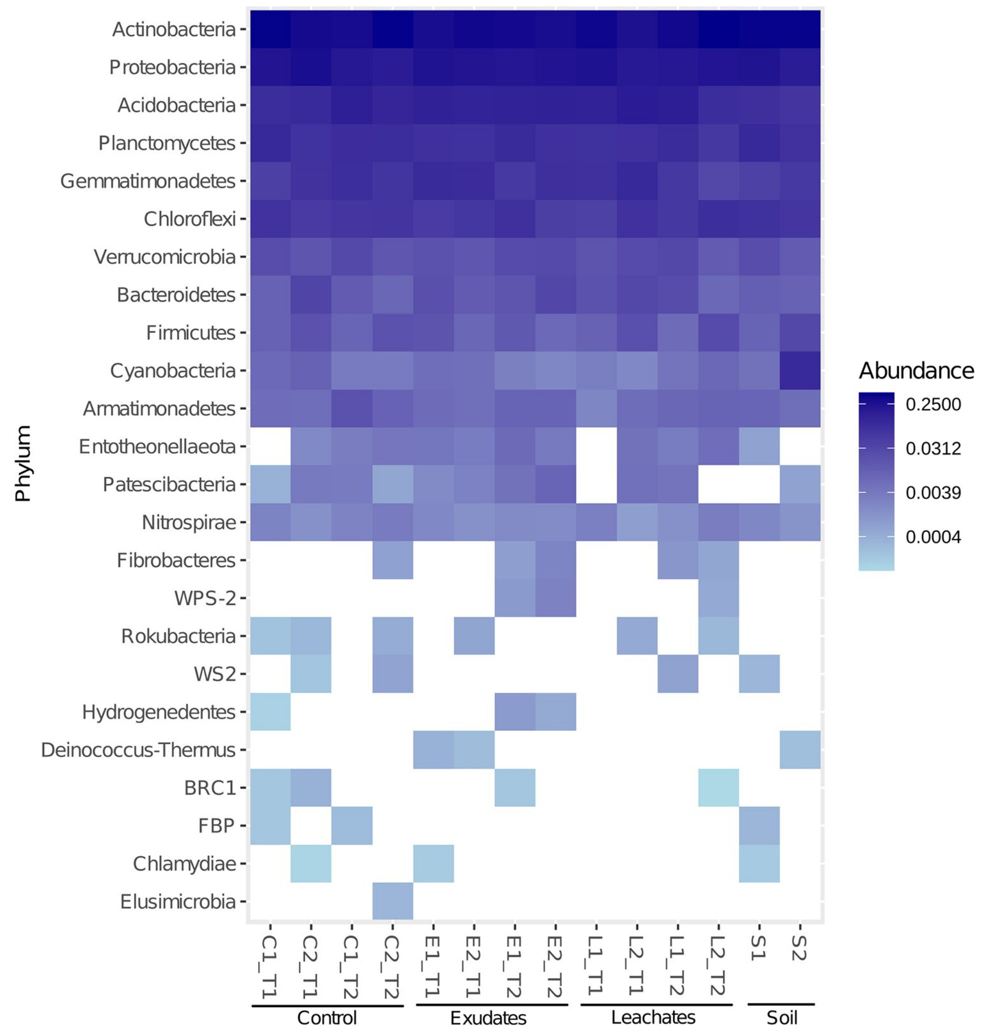

Fig 2. Buffelgrass microbiome at the phylum level. Each sample is labeled indicating control (C), exudates (E), leachates (L), soil (S), and sampling time (T1 and T2).

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The UniFrac dendrogram shows most communities clustered together according to time (Fig 3A, https://doi.org/10.6084/m9.figshare.c.6605350.v2). T2 showed larger dispersion in the dendrogram, suggesting community diversification over time. We further evaluated ß-diversity through a constrained analysis of principal coordinates (CAP) based on an unweighted UniFrac distance matrix (Fig 3B). The CAP analysis explained 20.9% of the observed variance. The x-axis described most of the ordination variance (10.8%) and separated the natural soil samples from the buffelgrass samples on the bottom right side of the ordination (Fig 3B). Control samples (C) are closer to the natural soils than the samples from allelochemical treatments (E and L). The CAP ordination clusters were evaluated with ANOSIM to test the treatment differences; no significant differences were found (R: 0.06579; p = 0.2902; 9,999 permutations). All T1 samples clustered at the upper half, as opposed to T2, located in the lower half of the ordination (Fig 3B). The ANOSIM test showed that time-based clustering was statistically significant (R: 0.2258; p = 0.0423, 9,999 permutations), meaning that the samples differed depending on time.

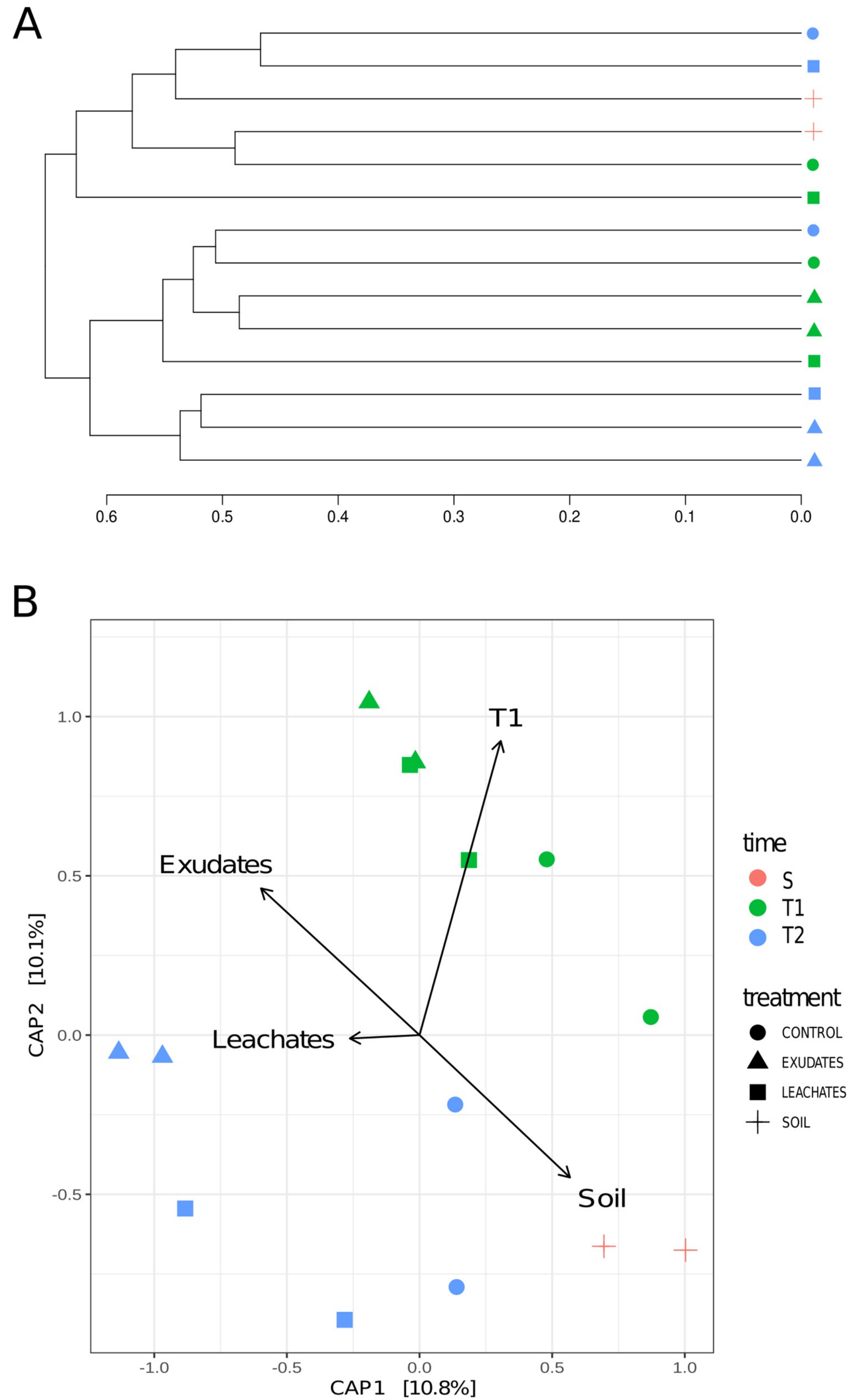

Fig 3. Beta diversity of the buffelgrass microbiome. (A) Dendrogram of microbiome relatedness based on an unweighted UniFrac distance matrix (A). (B) Beta diversity of the buffelgrass microbiome is represented as a constrained analysis of principal coordinates (CAP) based on an unweighted UniFrac distance matrix for all

treatments and natural soils (B). Vectors display the experiment's different treatments and time points. Statistical significance was evaluated using the ANOSIM test for treatments ($p = 0.092$) and time ($p = 0.0366$).

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We used a DESeq2 analysis to compare and identify abundant differential genera at each time point, irrespective of the treatment used (Fig 4A, S3 Table, https://doi.org/10.6084/m9.figshare.c.6605350.v2). In T1, we identified the following genera: *Nodosilinea_PCC-7104*, *Oceanibaculum*, *Pedobacter*, and *Flavitalea*, whereas *Ohtaekwangia*, *IS-44*, *Phytohabitans*, and *Saccharothrix* were enriched in T2. Subsequently, we compared the microbiome diversity at the genus level and found 14 genera representing 16 ASVs shared in all samples, including natural soils (S2 Fig, https://doi.org/10.6084/m9.figshare.c.6605350.v2). However, these genera represented only 0.78% of the whole ASVs dataset. Most ASVs were shared between all samples, with only 13 ASVs being unique to soil samples.

Actinobacteria were highly dominant across treatments and time. We found 30 genera in the treatment and control samples, comprising the buffelgrass core microbiome (Fig 5; S1 Fig, https://doi.org/10.6084/m9.figshare.c.6605350.v2). The core genera were *Geodermatophilus*, *RB41*, *Krasilnikovia*, *Angustibacter*, *Microvirga*, *Kribbella*, *Bradyrhizobium*, *Rubrobacter*, *Modestobacter*, *Pseudonocardia*, *Gemmatirosa*, *Bryobacter*, *Nitrospira*, *Nitrolancea*, *Solirubrobacter*, *Gaiella*, Candidatus_*Udaeobacter*, *Dactylosporangium*, *Altererythrobacter*, *Micromonospora*, *Roseomonas*, *Gemmata*, *Skermanella*, *Chthoniobacter*, *Gemmatimonas*, *Sphingomonas*, *JCM_18997*, Candidatus_*Alysiosphaera*, *Amycolatopsis*, and *Crossiella* (Fig 5). Most of them belong to the phylum *Actinobacteria*, except for *RB41* (*Acidobacteria*), *Gemmatirosa* and *Gemmatimonas* (*Gemmatimonadetes*), Candidatus_*Udaeobacter* and *Chthoniobacter* (*Verrucomicrobia*), *Gemmata* (*Planctomycetes*), and *Sphingomonas*, Candidatus_*Alysiosphaera*, *Roseomonas*, *Skermanella*, *Altererythrobacter*, and *Microvirga* (*Proteobacteria*). A complete list of the core microbiome species and a summary of where they had been isolated is available in S4 Table (https://doi.org/10.6084/m9.figshare.c.6605350.v2).

We identified enriched genera in the leachates compared to the controls, namely *Mle1-7*, *Caulobacter*, and *Phytohabitans* (Fig 4D, S3 Table). Compared to the controls, the genera enriched in the exudates were *Planctomicrobium*, *Aurantimonas*, *Cryptosporangium*, *Bauldia*, *Tellurimicrobium*, *Oxalicibacterium*, *Dokdonella*, *Nodosilinea-PCC-7104*, *Saccharothrix*, and *Caulobacter* (Fig 4B, S3 Table). The following genera were higher in the exudates than the leachates: *Saccharothrix*, *Jatrophihabitans*, *Tellurimicrobium*, *Bauldia*, *Aurantimonas*, *Oxalicibacterium*, *Planctomicrobium*, *Reyranella*, and *Singulisphaera* (Fig 4C, S3 Table). The genera enriched in the leachates were *Phytohabitans*, *Isoptericola*, *Pedobacter*, *Kouleothrix*, *Oceanibaculum*, *Yonghaparkia*, and *Pseudarthrobacter* (Fig 4C, S3 Table).

4. Discussion

4.1 The buffelgrass microbiome, clues for success as an invasive species?

Exploring the microbiomes of invasive species may shed light on the establishment and propagation of those species and their impacts on ecosystems, hence providing a turning point for developing new plant control strategies. Even though a high number of ASVs are shared between all treatment and soil samples, the microbiome composition of the rhizosphere of buffelgrass is distinguishable from that of the soil microbiome (Fig 3B, S1 Fig). Our results suggest that the overall bacterial communities of *Pennisetum ciliare* are structured by different factors. Overall, the Shannon values obtained for the samples in this study were within the range of Shannon values ($H' = 5.1811–5.5709$, Table 1) reported for microbiomes in desert soils [39]. Regarding the exudate treatment, previous reports have shown that the rhizosphere microbial

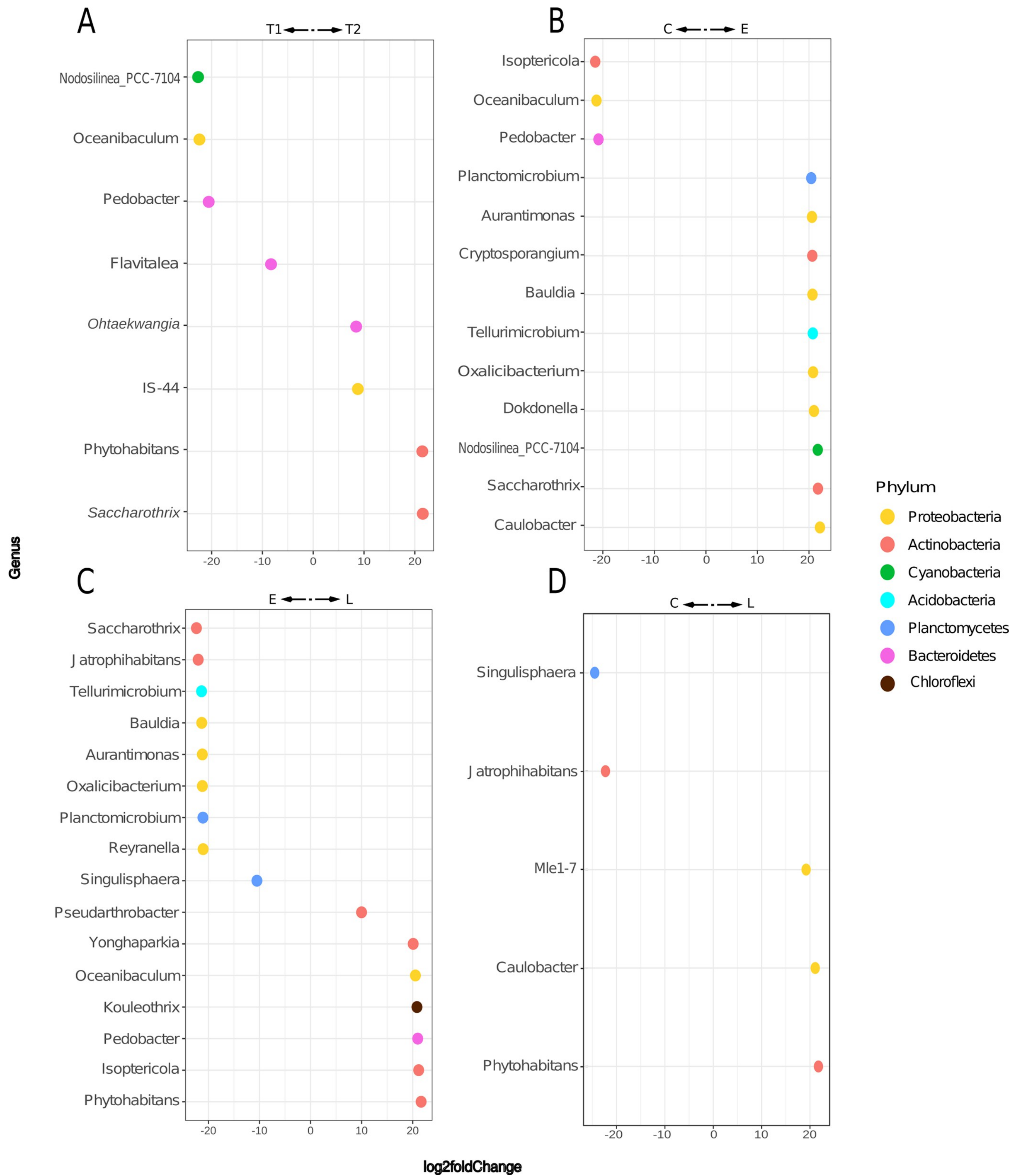

Fig 4. Enriched bacterial genera in allelochemical treatments. Shown are (A) the log2fold ratio between T1 and T2 (A), (B) exudates and leachates (B), (C) exudates and controls (C), and (D) leachates and controls (D). DESeq2 was used to get the significantly enriched genera in each condition using an **α** = 0.01.

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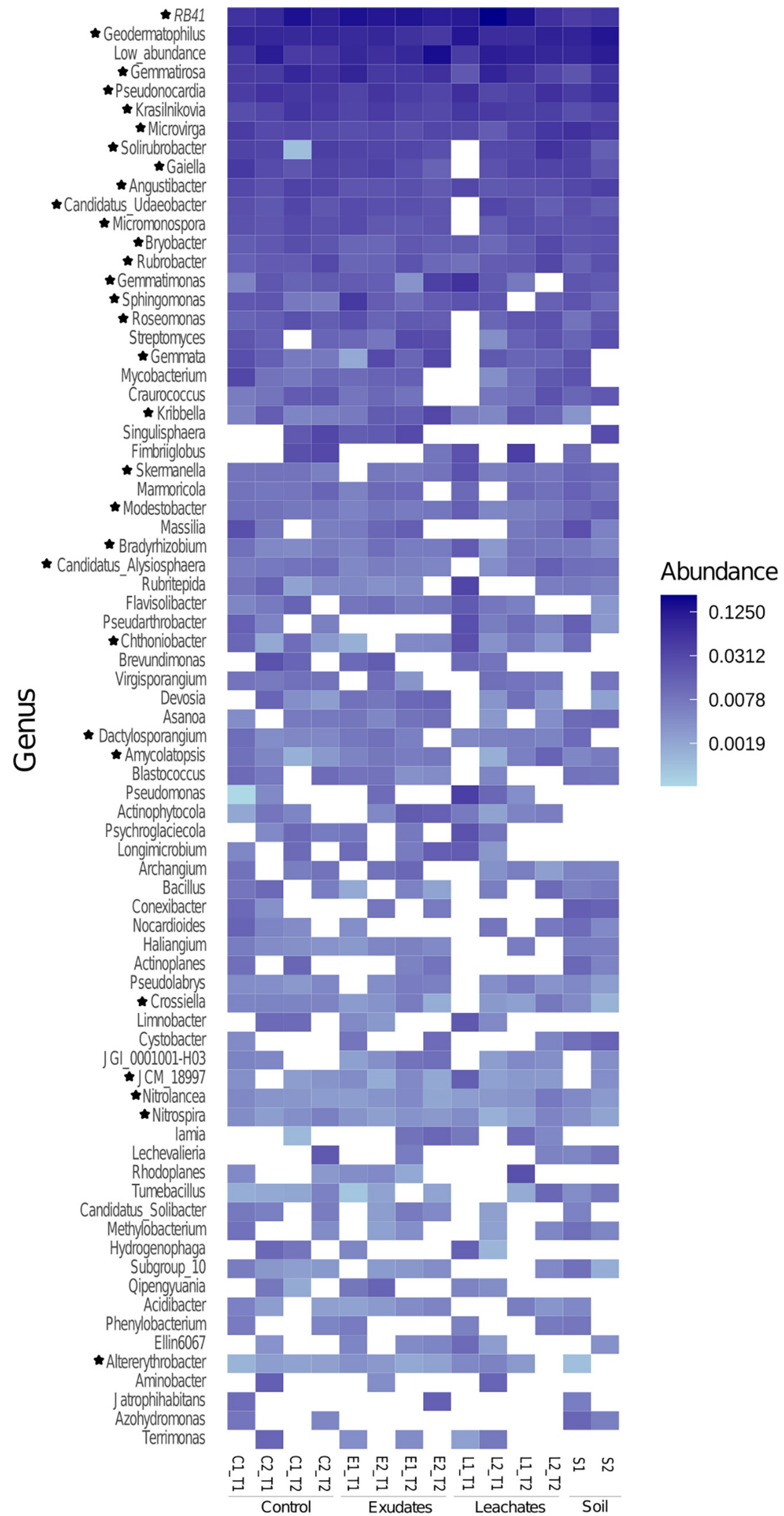
Genus
RB41
Geodermatophilus
Low_abundance
Gemmatirosa
Pseudonocardia
Krasilnikovia
Microvirga
Solirubrobacter
Gaiella
Angustibacter
Candidatus_Udaeobacter
Micromonospora
Bryobacter
Rubrobacter
Gemmatimonas
Sphingomonas
Roseomonas
Streptomyces
Gemmata
Mycobacterium
Craurococcus
Kribbella
Singulisphaera
Fimbriiglobus
Skermanella
Marmoricola
Modestobacter
Massilia
Bradyrhizobium
Candidatus_Alysiosphaera
Rubritepida
Flavisolibacter
Pseudarthrobacter
Chthoniobacter
Brevundimonas
Virgisporangium
Devosia
Asanoa
Dactylosporangium
Amycolatopsis
Blastococcus
Pseudomonas
Actinophytocola
Psychroglaciecola
Longimicrobium
Archangium
Bacillus
Conexibacter
Nocardioides
Haliangium
Actinoplanes
Pseudolabrys
Crossiella
Limnobacter
Cystobacter
JGI_0001001-H03
JCM_18997
Nitrolancea
Nitrospira
Iamia
Lechevalieria
Rhodoplanes
Tumebacillus
Candidatus_Solibacter
Methylobacterium
Hydrogenophaga
Subgroup_10
Qipengyuania
Acidibacter
Phenylobacterium
Ellin6067
Altererythrobacter
Aminobacter
Jatrophihabitans
Azohydromonas
Terrimonas
C1_T1
C2_T1
C1_T2
C2_T2
E1_T1
E2_T1
E1_T2
E2_T2
L1_T1
L2_T1
L1_T2
L2_T2
S1
S2
Control
Exudates
Leachates
Soil
Abundance
0.1250
0.0312
0.0078
0.0019

Fig 5. Buffelgrass core microbiome at the genus level. In the heatmap, genera marked with stars comprise the buffelgrass core microbiome. We collapsed genera with low abundances (< 0.03%).

https://doi.org/10.1371/journal.pone.0285978.g005

diversity tends to be higher when treated with root exudates, specifically when exposed to phenolic compounds [40]. These diversity observations align with the Shannon values obtained for exudates (*H'* = 5.51) and control treatments in T2 (*H'* = 5.24). The diversity for buffelgrass exposed to root exudates at the end of the experiment (*H'* = 5.51) was higher than the value from the bulk soil (*H'* = 5.34), a pattern already reported for ruderal plants, whose diversity in the rhizosphere tends to be higher than in bulk soil [41–43]. Our data showed that the rhizosphere microbiome of buffelgrass is dominated by the phyla *Actinobacteria*, *Proteobacteria*, *Acidobacteria*, *Planctomycetes*, *Gemmatimonadetes*, and *Chloroflexi* (Fig 2), in agreement with a previous report [2]. Likewise, the surrounding soil of the allelopathic desert shrub *Artemisia sieberi* shows similar phyla diversity [11]. The buffelgrass microbiome composition is also dominated by *Actinobacteria*, as reported in multiple arid areas [44, 45]. The ability of actinobacterial spores to germinate in environments with low water availability enables their adaptation to drought conditions [46] and shrub root zones of deserts [47].

Regarding the genus diversity, *RB41*, *Geodermatophilus*, *Gemmatirosa*, *Pseudonocardia*, *Krasilnikovia*, *Microvirga*, *Solirubrobacter*, *Gaiella*, *Angustibacter*, *Candidatus_Udaeobacter*, *Micromonospora*, *Bryobacter*, *Gemmatimonas*, *Sphingomonas*, and *Roseomonas* were highly abundant in buffelgrass, irrespective of the treatment or sampling time (Fig 5). Overall, 2,164 ASVs corresponded to 235 different genera of bacteria detected in our samples. Those 235 genera constitute the extended microbiome of buffelgrass roots under all tested conditions (S1 Fig). Those bacteria comprise what is referred to as the buffelgrass core microbiome. A core microbiome is a set of microorganisms forming cores of interactions that can be used to optimize microbial functions at the individual plant and ecosystem levels [23]. Diversity manipulation might be a key battleground where hosts and various hubs cooperate or compete, making them potential targets for plant biocontrol studies [48]. Rather than just promoting host plant growth, the core microbiome is vital in organizing the community assemblies mediating and organizing plant-microbe and microbe-microbe interactions by recruiting indigenous microorganisms with diverse functions and even suppressing high pathogen loads in the field [23]. Hence, the detailed study of core microbiomes should enable microbial species identification and functions on plant-microbe interactions that impact plant adaptation to arid environments and their featured plant-associated lifestyles [49, 50].

Among the core microbiome of buffelgrass (Fig 5, S2 Fig), some genera have been related to allelopathic conditions, such as *RB41*, *Bryobacter*, *Nitrospira*, *Gaiella*, and *Microvirga*, whose abundances changed depending on the amount of vanillic acid available [20, 51, 52]. Additionally, the genus *Nitrospira* has been reported in the soil surrounding buffelgrass and can oxidize ammonia to nitrate (commamox) [2]; commamox activity is frequent in oligotrophic habitats [53]. Hence, the presence of this genus in the core microbiome could help buffelgrass obtain nitrogen in deserts, where this element tends to be scarce. *Bradyrhizobium* is a common species in close association with plant roots, including known invasive species such as *Acacia dealbata*, and can enhance the competition ability of the tree [21, 54]. The potential for producing soluble vitamins, antimicrobials, and antibiotics detected in rhizosphere metagenomes and genomes of *Bradyrhizobium*, *Geodermatophilus*, *Pseudonocardia*, *Micromonospora*, *Crosiella*, *Amycolatopsis*, and *Kribbela*, may also be relevant in the context of plant invasions [55–60]. The genus *Sphingomonas* produces molecules that promote plant growth [61], whereas *Kribbella* isolates from allelopathic shrubs and other plant species show antifungal activity and contain genes related to the production of secondary metabolites [62, 63].

Gemmatimonas belong to the phylum *Gemmatimonadetes*, including taxa adapted to arid and oligotrophic conditions, and are among the most abundant bacteria in soils [64, 65]. The ability to thrive in replanted soils and to accumulate polyphosphates may be beneficial for the development of buffelgrass, which creates an allelopathic autotoxic environment and depends on phosphorus compounds to develop [5, 17, 66–68].

Solirubrobacter (*Actinobacteria*) is ubiquitous in buffelgrass. Only a few cultivated bacteria represent *Solirubrobacter*, with indirect evidence that showed them as a diverse group in nature [69]. *Solirubrobacter* can thrive in phenolic environments, using chlorogenic acid to grow [69], and can develop in the rhizosphere of cucumber treated with vanillin [20]. The relationship of *Solirubrobacter* with the plant host phenotype is ambiguous, with positive and negative correlations in multiple hosts [67, 70, 71]. Considering that several studies propose that certain fungi and bacteria contribute to the invasion success of plants introduced to novel habitats [25, 54], it is fair to suggest that the microorganisms recruited in the rhizosphere of buffelgrass contribute to a certain extent to the establishment and quick development of this grass in the habitats it invades.

4.2 Buffelgrass recruits taxa capable of thriving in an allelopathic environment

The allelochemicals exogenously amended during the experiment favored differential taxa abundance, according to our DESeq2 analysis (Fig 4). This effect of allelochemicals on microbiome structuring was observed in the CAP analysis, where the samples from the exudate and leachate treatments from T2 are further apart than they were in the first period (Fig 3B). Moreover, microbiomes from allelochemical treatments clustered apart from both soil and the controls. Even though the microbiomes under leachate treatment cluster closer to the controls, the distance is noticeable (Fig 3B). This strongly suggests that the presence of buffelgrass influences the root microbiome diversity.

Plants in desert ecosystems have developed ecophysiological adaptations, including salt and chemical compound excretion beneath the plant canopy, creating an allelopathic environment that affects organic matter and soil moisture levels [11]. Previously reported buffelgrass roots exudates comprised chlorogenic, ferulic, caffeic, *p*-OH-benzoic, *p*-coumaric, vanillic, syringic, and gentisic acids, all phenolic acids, and proven allelopathic agents [17]. Phenolic compounds recruit bacterial taxa more precisely than other rhizodeposits [72], and some compounds, such as coumaric acid, affect growth and microbial community composition in a concentration-dependent manner [19, 73]. Moreover, this concentration may increase through soil microbial production and using vanillic acid and *p*-OH-benzoic acid from ferulic acid and *p*-coumaric acid, respectively [19, 74, 75]. Since the buffelgrass microbiome changes across time and development, the root-recruited taxa are likely able to metabolize the phenolic compounds secreted by the roots at different growth stages [25].

We found 17 abundant differential bacterial genera in the treatments. As expected, some ASVs enriched in the exudates or leachate treatment are known for their capabilities to metabolize and produce phenolic compounds (S3 Table). Enriched taxa in allelochemical treatments were: *Caulobacter*, *Phytohabitans*, *Mle1-7*, *Planctomicrobium*, *Aurantimonas*, *Cryptosporangium*, *Bauldia*, *Tellurimicrobium*, *Oxalicibacterium*, *Dokdonella*, *Nodosilinea-PCC-7104*, and *Saccharothrix* (Fig 4B and 4D, S3 Table). *Caulobacter* (*Alphaproteobacteria*) was the only one enriched in both allelochemical treatments. *Caulobacter* acts as a microbial community hub on the leaf microbiome of *Arabidopsis* [48]; also as a plant growth-promoting bacterium (PGPB) through the production of plant hormones [76, 77]. Different species of *Caulobacter* can thrive in rhizosphere environments containing the same phenolic acids as those produced

by buffelgrass [18, 78–80]. *Cryptosporangium* and *Bauldia* are found in the roots of different allelopathic plants, such as *Eucalyptus* and *Andrographis* [81, 82].

Buffelgrass is a plant species preferred for grazing because of its high nutrient content. However, it can accumulate oxalate to potentially toxic concentrations for cattle [5, 17, 83]. *Oxalicibacterium* (*Burkholderiaceae)* comprises species isolated from soils and litter from oxalate-forming plants and collects oxalic acid or calcium oxalate as metabolic products, although oxalate can also be produced through the degradation of other compounds such as citrate [84–88]. Bacterial oxidation of calcium oxalate, which can be carried out by species such as *Oxalicibacterium* and *Bradyrhizobium*, entails soil alkalinization, which may act as a soil acidification buffer [88]. Similarly, *Tellurimicrobium* can grow on oxaloacetate [89]. The relevance of the ability to metabolize oxalic compounds is rooted in the importance of oxalate regarding plant physiology. Oxalate solubilizes insoluble phosphorus (in the form of aluminum phosphate) in the rhizosphere, is involved in pH regulation and calcium storage, and acts as a ligand for aluminum [85, 90]. Furthermore, oxalate degradation by some taxa, such as *Burkholderia*, lowers oxalate levels on plant surfaces and decreases the infection potential of pathogenic microorganisms attracted by oxalate [90].

Allelochemicals are compounds synthesized not only by plants but also by fungi and bacteria. The actinobacterium *Saccharothrix* was also enriched in the root exudate treatment compared to the controls. This genus can biosynthesize caffeic acid and cinnamic acid, the latter being a substrate for the biosynthesis of other phenolic compounds [91, 92]. The biosynthesis of phenolic compounds, such as vanillic acid from ferulic acid, through mechanisms involved with ß-oxidation, has been described in many bacteria [75]. The accumulation of allelochemicals, such as phenolic compounds, may have both plant and bacterial origins. Buffelgrass is a plant with a C4 metabolism, which enables it to thrive under drought conditions and high temperatures [6, 93]. Under those conditions, plants draw upon different strategies, such as regulating stomatal aperture [94]. Albeit in a concentration-dependent manner, phenolic acids influence the aperture and closure of stomata in various plant species [95–97] and may even alter photosynthetic performance [98]. Hence allelochemicals could act as signals in a cross-kingdom talk, and it is possible that the presence of phenolic compounds, either supplied by the plant or by the microorganisms, in the buffelgrass rhizosphere influences buffelgrass physiology through the regulation of stomatal aperture and closure, which in turn could diminish the growth of the plant while promoting its development in an arid environment. Further analysis using a metagenomic perspective may confirm the interpretations made in this study.

4.3 Time as a microbiome community-structuring factor

Rhizosphere microbiomes are composed of bacteria filtered by the same plant from the bulk soil, creating microbial communities in the rhizosphere, distinguishable from the soil communities [99–101]. The soil-to-rhizosphere effect was shown in the ordination (10.8% of the variance, Fig 3B), where the samples from bulk soil (S) cluster apart from all the treatment samples derived from buffelgrass plants. Interestingly, the Unifrac dendrogram shows a closer phylogenetic relationship between communities belonging to the same period (Fig 3A). Similarly, CAP analysis revealed that time influences the microbiome structure, shown through differences in the microbiome between T1 and T2 (Fig 3B). Studies have reported plant developmental effects on plant-associated microbiomes [102–104]. Even though the period evaluated is relatively short (20 days difference), specific phyla changed in their abundances concerning T1 and T2 (Fig 2). The *WPS-2* and *Hydrogenedentes* showed increased quantities from T1 to T2 in the root exudates treatment. In contrast, *Fibrobacteres* rose in the root exudate and leachate treatment. *Hydrogenedentes* have been reported in contaminated zones with

compounds such as phenanthrene [105] and associated with multiple plants [40,106]. Phylum *WPS-2* has been registered as dominant in the invasive tree *Eucalyptus*, with a declining abundance after the first year [107]. *Deinococcus-Thermus* abundance was reduced from T2 to T1 in the root exudate treatment; in a previous study, the absence of phenolic allelochemicals correlated with a decreased *Deinococcus-Thermus* abundance [40].

Additionally, plant age is the main factor shaping root microbiomes in maize and *Eucalyptus*, stabilizing over time [107, 108]. Regarding buffelgrass, the microbiome changes probably reflect plant aging. The plant's different development stages, including the allelochemicals exuded by the roots, cause changes in the compounds secreted by the roots [78]. Time differences were highlighted by differential taxa in buffelgrass (Fig 4A, S3 Table). During the first period (T1), the significantly enriched bacteria belonged to the phyla *Cyanobacteria* (*Nodosilinea PCC-7104*), *Proteobacteria* (*Oceanibaculum*), and *Bacteroidetes* (*Pedobacter* and *Flavitalea*). *Pedobacter* and *Oceanibaculum* have increased abundances during the intermediate plant growth stages [109]. Bacteria in the rhizosphere of younger plants tend to use simple amino acids instead of complex carbohydrates, as do older plants [110]. This young plant hypothesis is backed by previous studies, where the amendment of soils with a mixture of long-chain fatty acids and amino acids led to an enrichment of *Flavitalea* [111].

We observed that *Bacteroidetes* (*Ohtaekwangia*), *Proteobacteria* (*IS-44*), and *Actinobacteria* (*Phytohabitans* and *Saccharothrix*) were enriched in T2. This is consistent with previous findings that *Ohtaekwangia* dominates in the middle-growth stages of other plant microbiomes and decreases over time [104, 109]. Our results also agree with this pattern, as T2 buffelgrass plants were not at a young growth stage, and *Ohtaekwangia* was enriched in 112-day-old buffelgrass plants.

The changes in root exudates of a plant throughout its development can significantly impact the microbial communities in the surrounding soil, as highlighted by recent studies [78, 112]. This highlights the importance of considering the plant's phenology and physiological and chemical traits when examining the effects of invasive plant species on ecosystems [1, 73]. Additionally, the impact of phenolic compounds on the soil microbiome is known to be concentration-dependent [19, 73], and changes in root exudate patterns during the development of buffelgrass could provide new insights into the formation of rhizosphere microbiomes. Our experimental design, which included the use of PVC pots, may have also played a role in the accumulation of phenolics and leachates, leading to the selection of microorganisms that are capable of thriving in environments with high concentrations of phenolic compounds or can degrade these compounds as part of their metabolism. These microorganisms are probably selected based on the buffelgrass's growth stage and allelochemicals' fate and concentration. This interaction could allow the plant to benefit from the production of antimicrobial compounds and the degradation of contaminants, allowing the plant to expand its invasive range.

5. Conclusions

Actinobacteria dominated the microbiome of buffelgrass, resembling the microbiomes of other plants in deserts, arid zones, and other desert allelopathic shrubs. Our study found that rhizosphere microbiomes treated with allelochemicals had a higher abundance of specific taxa, highlighting the impact of buffelgrass on soil bacteria. Time was also found to be a significant factor in shaping the buffelgrass microbiome, and further research should focus on the effects of allelochemicals on the root exudate pattern throughout the plant's development. Additionally, we identified a core microbiome comprising microorganisms known for their antimicrobial or vitamin production capabilities, which likely played a role in shaping the remaining

microbial community associated with the roots of buffelgrass. Our findings suggest that certain recruited bacteria can metabolize allelochemical compounds excreted by the roots, potentially influencing buffelgrass physiology.

Supporting information

S1 Fig. Buffelgrass microbiome composition heatmap at the genus level.
(EPS)

S2 Fig. Shared genera between samples visualized in an UpSet plot. Shared taxa at the genus level between allelochemical and control treatments. The histogram shows the number of shared elements for each intersection set, ordered in a decreasing manner. Genera marked with stars comprise the buffelgrass core microbiome, considering taxa present in all treatment and control samples but allowing absence in one of the samples.
(TIFF)

S1 Table. Taxonomy table indicating the taxonomic assignment of each ASV.
(XLSX)

S2 Table. Counts table indicating the number of appearances of each ASV per sample.
(XLSX)

S3 Table. Genera significantly enriched in paired comparisons. The table shows significantly differentially abundant bacteria calculated by DESeq2 analysis. Information about the microbe genera, sources of isolation, and associated publications are also provided.
(XLSX)

S4 Table. Genera comprising the buffelgrass core microbiome. Bibliographical information about microbial sources of isolation and associated publications.
(XLSX)

S1 Dataset. Phylogenetic tree. Newick file for the phylogenetic tree constructed with FastTreeMP using 16S rRNA gene sequences obtained from buffelgrass microbiome samples.
(NWK)

Acknowledgments

We thank field work assistance by José F. Martínez.

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References

1. Ehrenfeld JG. Ecosystem Consequences of Biological Invasions. Annu Rev Ecol Evol Syst. 2010; 41: 59–80. https://doi.org/10.1146/annurev-ecolsys-102209-144650
2. Gornish ES, Franklin K, Rowe J, Barberán A. Buffelgrass invasion and glyphosate effects on desert soil microbiome communities. Biol Invasions. 2020; 22: 2587–2597. https://doi.org/10.1007/s10530-020-02268-8
3. Hanselka CW, Hussey MA FI. Buffelgrass. Warm-Season (C4) Grasses. 2015. pp. 477–502. https://doi.org/10.2134/agronmonogr45.c13
4. Hussain F, Naqvi HH, Ilahi I. Interference Exhibited by *Cenchrus ciliaris* L. and *Bothriochloa pertusa* (L.) A. Camus. Bull Torrey Bot Club. 1982; 109: 513. https://doi.org/10.2307/2996492
5. Marshall VM, Lewis MM, Ostendorf B. Buffel grass (*Cenchrus ciliaris*) as an invader and threat to biodiversity in arid environments: A review. J Arid Environ. 2012; 78: 1–12. https://doi.org/10.1016/j.jaridenv.2011.11.005
6. Lyons KG, Maldonado-Leal BG, Owen G. Community and Ecosystem Effects of Buffelgrass (*Pennisetum ciliare*) and Nitrogen Deposition in the Sonoran Desert. Invasive Plant Sci Manag. 2013; 6: 65–78. https://doi.org/10.1614/IPSM-D-11-00071.1
7. Miller G, Friedel M, Adam P, Chewings V. Ecological impacts of buffel grass (*Cenchrus ciliaris* L.) invasion in central Australia—does field evidence support a fire-invasion feedback? Rangel J. 2010; 32: 353. https://doi.org/10.1071/RJ09076
8. Tinoco-Ojanguren C, Díaz A, Martínez J, Molina-Freaner F. Species diversity and regeneration of native species in *Pennisetum ciliare* (buffelgrass) pastures from the thornscrub of Sonora, México. J Arid Environ. 2013; 97: 26–37. https://doi.org/10.1016/j.jaridenv.2013.05.009
9. Arriaga L, Castellanos V. AE, Moreno E, Alarcón J. Potential Ecological Distribution of Alien Invasive Species and Risk Assessment: a Case Study of Buffel Grass in Arid Regions of Mexico. Conserv Biol. 2004; 18: 1504–1514. https://doi.org/10.1111/j.1523-1739.2004.00166.x
10. Burgos A, Maass JM. Vegetation change associated with land-use in tropical dry forest areas of Western Mexico. Agric Ecosyst Environ. 2004; 104: 475–481. https://doi.org/10.1016/j.agee.2004.01.038
11. Saul-Tcherkas V, Unc A, Steinberger Y. Soil Microbial Diversity in the Vicinity of Desert Shrubs. Microb Ecol. 2013; 65: 689–699. https://doi.org/10.1007/s00248-012-0141-8 PMID: 23192699
12. Ridenour WM, Callaway RM. The relative importance of allelopathy in interference: the effects of an invasive weed on a native bunchgrass. Oecologia. 2001; 126: 444–450. https://doi.org/10.1007/s004420000533 PMID: 28547460
13. Schandry N, Becker C. Allelopathic Plants: Models for Studying Plant–Interkingdom Interactions. Trends Plant Sci. 2020; 25: 176–185. https://doi.org/10.1016/j.tplants.2019.11.004 PMID: 31837955
14. Kato-Noguchi H. Allelopathy and Allelochemicals of *Leucaena leucocephala* as an Invasive Plant Species. Plants. 2022; 11: 1672. https://doi.org/10.3390/plants11131672 PMID: 35807624
15. Thorpe AS, Thelen GC, Diaconu A, Callaway RM. Root exudate is allelopathic in invaded community but not in native community: field evidence for the novel weapons hypothesis. J Ecol. 2009; 97: 641–645. https://doi.org/10.1111/j.1365-2745.2009.01520.x
16. Rice E. L. Allelopathy. 2nd ed. Academic Press, Orlando, Florida, USA. 1984.
17. Hussain F, Ilahi I, Malik SA, Dasti AA, Ahmad B. Allelopathic effects of rain leachates and root exudates of *Cenchrus ciliaris* L. And *Bothriochloa pertusa* (L.) A. Camus. Pakistan J Bot. 2011; 43: 341–350.
18. Hussain MI, Reigosa MJ. Allelochemical stress inhibits growth, leaf water relations, PSII photochemistry, non-photochemical fluorescence quenching, and heat energy dissipation in three C3 perennial species. J Exp Bot. 2011; 62: 4533–4545. https://doi.org/10.1093/jxb/err161 PMID: 21659663
19. Blum U. Effects of microbial utilization of phenolic acids and their phenolic acid breakdown products on allelopathic interactions. J Chem Ecol. 1998; 24: 685–708. https://doi.org/10.1023/A:1022394203540
20. Song J, Qiu S, Zhao J, Han C, Wang Y, Sun X, et al. *Pseudonocardia tritici* sp. nov., a novel actinomycete isolated from rhizosphere soil of wheat (*Triticum aestivum* L.). Kleunen M, editor. Antonie Van Leeuwenhoek. 2019; 112: 765–773. https://doi.org/10.1007/s10482-018-01210-2 PMID: 30536164
21. Klock MM, Barrett LG, Thrall PH, Harms KE. Host promiscuity in symbiont associations can influence exotic legume establishment and colonization of novel ranges. Kleunen M, editor. Divers Distrib. 2015; 21: 1193–1203. https://doi.org/10.1111/ddi.12363
22. Fonseca E da S, Peixoto RS, Rosado AS, Balieiro F de C, Tiedje JM, Rachid CTC da C. The Microbiome of Eucalyptus Roots under Different Management Conditions and Its Potential for Biological

Nitrogen Fixation. Microb Ecol. 2018; 75: 183–191. https://doi.org/10.1007/s00248-017-1014-y PMID: 28634640

23. Toju H, Peay KG, Yamamichi M, Narisawa K, Hiruma K, Naito K, et al. Core microbiomes for sustainable agroecosystems. Nat Plants. 2018; 4: 247–257. https://doi.org/10.1038/s41477-018-0139-4 PMID: 29725101

24. Ravanbakhsh M, Kowalchuk GA, Jousset A. Root-associated microorganisms reprogram plant life history along the growth–stress resistance tradeoff. ISME J. 2019; 13: 3093–3101. https://doi.org/10.1038/s41396-019-0501-1 PMID: 31511619

25. Trognitz F, Hackl E, Widhalm S, Sessitsch A. The role of plant–microbiome interactions in weed establishment and control. Muyzer G, editor. FEMS Microbiol Ecol. 2016;92: fiw138. https://doi.org/10.1093/femsec/fiw138 PMID: 27387910

26. Kamutando CN, Vikram S, Kamgan-Nkuekam G, Makhalanyane TP, Greve M, Roux JJ Le, et al. Soil nutritional status and biogeography influence rhizosphere microbial communities associated with the invasive tree *Acacia dealbata*. Sci Rep. 2017; 7: 6472. https://doi.org/10.1038/s41598-017-07018-w PMID: 28747705

27. Tinoco-Ojanguren C, Reyes-Ortega I, Sánchez-Coronado ME, Molina-Freaner F, Orozco-Segovia A. Germination of an invasive *Cenchrus ciliaris* L. (buffel grass) population of the Sonoran Desert under various environmental conditions. South African J Bot. 2016; 104: 112–117. https://doi.org/10.1016/j.sajb.2015.10.009

28. Cruz-Ortega R, Ayala-Cordero G, Anaya AL. Allelochemical stress produced by the aqueous leachate of *Callicarpa acuminata*: effects on roots of bean, maize, and tomato. Physiol Plant. 2002; 116: 20–27. https://doi.org/10.1034/j.1399-3054.2002.1160103.x PMID: 12207658

29. Callahan BJ, McMurdie PJ, Rosen MJ, Han AW, Johnson AJA, Holmes SP. DADA2: High-resolution sample inference from Illumina amplicon data. Nat Methods. 2016; 13: 581–583. https://doi.org/10.1038/nmeth.3869 PMID: 27214047

30. Yilmaz P, Parfrey LW, Yarza P, Gerken J, Pruesse E, Quast C, et al. The SILVA and "All-species Living Tree Project (LTP)" taxonomic frameworks. Nucleic Acids Res. 2014; 42: D643–D648. https://doi.org/10.1093/nar/gkt1209 PMID: 24293649

31. Price MN, Dehal PS, Arkin AP. FastTree: Computing Large Minimum Evolution Trees with Profiles instead of a Distance Matrix. Mol Biol Evol. 2009; 26: 1641–1650. https://doi.org/10.1093/molbev/msp077 PMID: 19377059

32. McMurdie PJ, Holmes S. phyloseq: An R Package for Reproducible Interactive Analysis and Graphics of Microbiome Census Data. Watson M, editor. PLoS One. 2013; 8: e61217. https://doi.org/10.1371/journal.pone.0061217 PMID: 23630581

33. Wickham H, Chang W. Package 'ggplot2'. R Foundation for Statistical Computing. 2016; Vienna, Austria. Available from https://cran.r-project.org/web/packages/ggplot2/ggplot2.pdf.

34. Oksanen J. Vegan: ecological diversity. In: R Package Version 2.4–4 [Internet]. 2017 p. 11. Available: https://cran.r-project.org/package=vegan=vegan

35. R Core Team. R: A Language and Environment for Statistical Computing [Internet]. 2013 Vienna, Austria: R Foundation for Statistical Computing. Available: http://www.r-project.org/

36. Lozupone C, Knight R. UniFrac: a New Phylogenetic Method for Comparing Microbial Communities. Appl Environ Microbiol. 2005; 71: 8228–8235. https://doi.org/10.1128/AEM.71.12.8228-8235.2005 PMID: 16332807

37. Clarke KR. Non-parametric multivariate analyses of changes in community structure. Austral Ecol. 1993; 18: 117–143. https://doi.org/10.1111/j.1442-9993.1993.tb00438.x

38. Love MI, Huber W, Anders S. Moderated estimation of fold change and dispersion for RNA-seq data with DESeq2. Genome Biol. 2014; 15: 550. https://doi.org/10.1186/s13059-014-0550-8 PMID: 25516281

39. Vásquez-Dean J, Maza F, Morel I, Pulgar R, González M. Microbial communities from arid environments on a global scale. A systematic review. Biol Res. 2020; 53: 29. https://doi.org/10.1186/s40659-020-00296-1 PMID: 32631429

40. Fang C, Zhuang Y, Xu T, Li Y, Li Y, Lin W. Changes in Rice Allelopathy and Rhizosphere Microflora by Inhibiting Rice Phenylalanine Ammonia-lyase Gene Expression. J Chem Ecol. 2013; 39: 204–212. https://doi.org/10.1007/s10886-013-0249-4 PMID: 23385369

41. Thompson LR, Sanders JG, McDonald D, Amir A, Ladau J, Locey KJ, et al. A communal catalogue reveals Earth's multiscale microbial diversity. Nature. 2017; 551: 457–463. https://doi.org/10.1038/nature24621 PMID: 29088705

42. Marasco R, Mosqueira MJ, Fusi M, Ramond J-B, Merlino G, Booth JM, et al. Rhizosheath microbial community assembly of sympatric desert speargrasses is independent of the plant host. Microbiome. 2018; 6: 215. https://doi.org/10.1186/s40168-018-0597-y PMID: 30514367

43. Barajas HR, Martínez-Sánchez S, Romero MF, Álvarez CH, Servín-González L, Peimbert M, et al. Testing the Two-Step Model of Plant Root Microbiome Acquisition Under Multiple Plant Species and Soil Sources. Front Microbiol. 2020; 11: 1–17. https://doi.org/10.3389/fmicb.2020.542742 PMID: 33162946

44. Crits-Christoph A, Robinson CK, Barnum T, Fricke WF, Davila AF, Jedynak B, et al. Colonization patterns of soil microbial communities in the Atacama Desert. Microbiome. 2013; 1: 28. https://doi.org/10.1186/2049-2618-1-28 PMID: 24451153

45. Naylor D, Coleman-Derr D. Drought Stress and Root-Associated Bacterial Communities. Front Plant Sci. 2018; 8: 1–16. https://doi.org/10.3389/fpls.2017.02223 PMID: 29375600

46. Mohammadipanah F, Wink J. Actinobacteria from Arid and Desert Habitats: Diversity and Biological Activity. Front Microbiol. 2016; 6: 1–10. https://doi.org/10.3389/fmicb.2015.01541 PMID: 26858692

47. Steven B, Gallegos-Graves LV, Starkenburg SR, Chain PS, Kuske CR. Targeted and shotgun metagenomic approaches provide different descriptions of dryland soil microbial communities in a manipulated field study. Environ Microbiol Rep. 2012; 4: 248–256. https://doi.org/10.1111/j.1758-2229.2012.00328.x PMID: 23757280

48. Agler MT, Ruhe J, Kroll S, Morhenn C, Kim S-T, Weigel D, et al. Microbial Hub Taxa Link Host and Abiotic Factors to Plant Microbiome Variation. Waldor MK, editor. PLOS Biol. 2016; 14: e1002352. https://doi.org/10.1371/journal.pbio.1002352 PMID: 26788878

49. Busby PE, Soman C, Wagner MR, Friesen ML, Kremer J, Bennett A, et al. Research priorities for harnessing plant microbiomes in sustainable agriculture. PLOS Biol. 2017; 15: e2001793. https://doi.org/10.1371/journal.pbio.2001793 PMID: 28350798

50. Alcaraz LD, Peimbert M, Barajas HR, Dorantes-Acosta AE, Bowman JL, Arteaga-Vázquez MA. *Marchantia liverworts* as a proxy to plants' basal microbiomes. Sci Rep. 2018; 8: 12712. https://doi.org/10.1038/s41598-018-31168-0 PMID: 30140076

51. Zhou XG, Jia HT, Pan DD, Ge X, Qiao PL, Wu FZ. Effects of vanillin on cucumber rhizosphere bacterial community. Allelopath J. 2018; 44: 191–200. https://doi.org/10.26651/allelo.j./2018-44-2-1164

52. Zhou X, Wu F. Vanillic acid changed cucumber (*Cucumis sativus* L.) seedling rhizosphere total bacterial, *Pseudomonas* and *Bacillus* spp. communities. Sci Rep. 2018; 8: 4929. https://doi.org/10.1038/s41598-018-23406-2 PMID: 29563548

53. Podlesnaya G, Krasnopeev A, Potapov S, Tikhonova I, Shtykova Y, Suslova M, et al. Diversity of nitrifying bacteria in microbial communities from water and epilithic biofilms of the Lake Baikal littoral zone. Limnol Freshw Biol. 2020; 2020: 1008–1010. https://doi.org/10.31951/2658-3518-2020-A-4-1008

54. Kamutando CN, Vikram S, Kamgan-Nkuekam G, Makhalanyane TP, Greve M, Le Roux JJ, et al. The Functional Potential of the Rhizospheric Microbiome of an Invasive Tree Species, *Acacia dealbata*. Microb Ecol. 2019; 77: 191–200. https://doi.org/10.1007/s00248-018-1214-0 PMID: 29948018

55. Wardle DA, Nicholson KS, Ahmed M, Rahman A. Interference effects of the invasive plant Carduus nutans L. against the nitrogen fixation ability of *Trifolium repens* L. Plant Soil. 1994; 163: 287–297. https://doi.org/10.1007/BF00007978

56. Gao R, Liu C, Zhao J, Jia F, Yu C, Yang L, et al. *Micromonospora jinlongensis* sp nov, isolated from muddy soil in China and emended description of the genus *Micromonospora*. Antonie van Leeuwenhoek, Int J Gen Mol Microbiol. 2014; 105: 307–315. https://doi.org/10.1007/s10482-013-0074-3 PMID: 24257967

57. Boumehira AZ, El-Enshasy HA, Hacène H, Elsayed EA, Aziz R, Park EY. Recent progress on the development of antibiotics from the genus *Micromonospora*. Biotechnology and Bioprocess Engineering. Korean Society for Biotechnology and Bioengineering; 2016. pp. 199–223. https://doi.org/10.1007/s12257-015-0574-2

58. Tanvir R, Sajid I, Hasnain S, Kulik A, Grond S. Rare actinomycetes *Nocardia caishijiensis* and *Pseudonocardia carboxydivorans* as endophytes, their bioactivity and metabolites evaluation. Microbiol Res. 2016; 185: 22–35. https://doi.org/10.1016/j.micres.2016.01.003 PMID: 26946375

59. Gonzalez-Pimentel JL, Dominguez-Moñino I, Jurado V, Laiz L, Caldeira AT, Saiz-Jimenez C. The Rare *Actinobacterium Crossiella* sp. Is a Potential Source of New Bioactive Compounds with Activity against Bacteria and Fungi. Microorganisms. 2022; 10: 1575. https://doi.org/10.3390/microorganisms10081575 PMID: 36013993

60. Song Z, Xu T, Wang J, Hou Y, Liu C, Liu S, et al. Secondary Metabolites of the Genus *Amycolatopsis*: Structures, Bioactivities and Biosynthesis. Molecules. 2021; 26: 1884. https://doi.org/10.3390/molecules26071884 PMID: 33810439

61. Khan AL, Waqas M, Kang SM, Al-Harrasi A, Hussain J, Al-Rawahi A, et al. Bacterial endophyte *Sphingomonas* sp. LK11 produces gibberellins and IAA and promotes tomato plant growth. J Microbiol. 2014; 52: 689–695. https://doi.org/10.1007/s12275-014-4002-7 PMID: 24994010

62. Zhou Z, Gu J, Li Y-Q, Wang Y. Genome plasticity and systems evolution in *Streptomyces*. BMC Bioinformatics. 2012; 13: S8. https://doi.org/10.1186/1471-2105-13-S10-S8 PMID: 22759432

63. Gong X, Shi J, Zhou X, Yuan T, Gao D, Wu F. Crop Rotation With Cress Increases Cucumber Yields by Regulating the Composition of the Rhizosphere Soil Microbial Community. Front Microbiol. 2021; 12. https://doi.org/10.3389/fmicb.2021.631882 PMID: 33776961

64. DeBruyn JM, Nixon LT, Fawaz MN, Johnson AM, Radosevich M. Global Biogeography and Quantitative Seasonal Dynamics of *Gemmatimonadetes* in Soil. Appl Environ Microbiol. 2011; 77: 6295–6300. https://doi.org/10.1128/AEM.05005-11 PMID: 21764958

65. Zeng Y, Nupur Wu N, Madsen AM, Chen X, Gardiner AT, et al. *Gemmatimonas groenlandica* sp. nov. Is an Aerobic Anoxygenic Phototroph in the Phylum *Gemmatimonadetes*. Front Microbiol. 2021; 11: 1–18. https://doi.org/10.3389/fmicb.2020.606612 PMID: 33519753

66. Zhang Y, Cong J, Lu H, Yang C, Yang Y, Zhou J, et al. An Integrated Study to Analyze Soil Microbial Community Structure and Metabolic Potential in Two Forest Types. Ibekwe AM, editor. PLoS One. 2014; 9: e93773. https://doi.org/10.1371/journal.pone.0093773 PMID: 24743581

67. Franke-Whittle IH, Manici LM, Insam H, Stres B. Rhizosphere bacteria and fungi associated with plant growth in soils of three replanted apple orchards. Plant Soil. 2015; 395: 317–333. https://doi.org/10.1007/s11104-015-2562-x

68. Liu M-J, Jin C-Z, Ersiman A, Park D-J, Kim C-J. *Flavitalea flava* sp. nov., a bacterium isolated from a soil sample, and emended description of the genus *Flavitalea*. Antonie Van Leeuwenhoek. 2019; 112: 275–281. https://doi.org/10.1007/s10482-018-1156-z PMID: 30171433

69. Whitman WB. *Solirubrobacter*. Bergey's Man Syst Archaea Bact. 2015. https://doi.org/10.1002/9781118960608.gbm00227

70. Yang J, Ruegger PM, McKenry M V., Becker JO, Borneman J. Correlations between Root-Associated Microorganisms and Peach Replant Disease Symptoms in a California Soil. Yang C-H, editor. PLoS One. 2012; 7: e46420. https://doi.org/10.1371/journal.pone.0046420 PMID: 23071565

71. Hernández-Álvarez C, García-Oliva F, Cruz-Ortega R, Romero MF, Barajas HR, Piñero D, et al. Squash root microbiome transplants and metagenomic inspection for in situ arid adaptations. Sci Total Environ. 2022; 805: 150136. https://doi.org/10.1016/j.scitotenv.2021.150136 PMID: 34818799

72. Badri D V., Chaparro JM, Zhang R, Shen Q, Vivanco JM. Application of Natural Blends of Phytochemicals Derived from the Root Exudates of *Arabidopsis* to the Soil Reveal That Phenolic-related Compounds Predominantly Modulate the Soil Microbiome. J Biol Chem. 2013; 288: 4502–4512. https://doi.org/10.1074/jbc.M112.433300 PMID: 23293028

73. Zhou X, Wu F. p-Coumaric Acid Influenced Cucumber Rhizosphere Soil Microbial Communities and the Growth of *Fusarium oxysporum* f.sp. *cucumerinum* Owen. Gilbert JA, editor. PLoS One. 2012; 7: e48288. https://doi.org/10.1371/journal.pone.0048288 PMID: 23118972

74. Smith-Becker J, Marois E, Huguet EJ, Midland SL, Sims JJ, Keen NT. Accumulation of Salicylic Acid and 4-Hydroxybenzoic Acid in Phloem Fluids of Cucumber during Systemic Acquired Resistance Is Preceded by a Transient Increase in Phenylalanine Ammonia-Lyase Activity in Petioles and Stems1. Plant Physiol. 1998; 116: 231–238. https://doi.org/10.1104/pp.116.1.231 PMID: 9449843

75. Świzdor A, Panek A, Milecka-Tronina N, Kołek T. Biotransformations Utilizing β-Oxidation Cycle Reactions in the Synthesis of Natural Compounds and Medicines. Int J Mol Sci. 2012; 13: 16514–16543. https://doi.org/10.3390/ijms131216514 PMID: 23443116

76. Luo D, Langendries S, Mendez SG, De Ryck J, Liu D, Beirinckx S, et al. Plant Growth Promotion Driven by a Novel *Caulobacter Strain*. Mol Plant-Microbe Interact. 2019; 32: 1162–1174. https://doi.org/10.1094/MPMI-12-18-0347-R PMID: 30933667

77. Berrios L, Ely B. Plant growth enhancement is not a conserved feature in the *Caulobacter* genus. Plant Soil. 2020; 449: 81–95. https://doi.org/10.1007/s11104-020-04472-w

78. Hao ZP, Wang Q, Christie P, Li XL. Allelopathic potential of watermelon tissues and root exudates. Sci Hortic (Amsterdam). 2007; 112: 315–320. https://doi.org/10.1016/j.scienta.2006.12.030

79. Thanbichler M, Iniesta AA, Shapiro L. A comprehensive set of plasmids for vanillate- and xylose-inducible gene expression in *Caulobacter crescentus*. Nucleic Acids Res. 2007; 35: e137–e137. https://doi.org/10.1093/nar/gkm818 PMID: 17959646

80. Sun L-N, Yang E-D, Hou X-T, Wei J-C, Yuan Z-X, Wang W-Y. *Caulobacter rhizosphaerae* sp. nov., a stalked bacterium isolated from rhizosphere soil. Int J Syst Evol Microbiol. 2017; 67: 1771–1776. https://doi.org/10.1099/ijsem.0.001860 PMID: 28598303

81. Himaman W, Suksaard P, Mingma R, Matsumoto A, Duangmal K. *Cryptosporangium eucalypti* sp. nov., an actinomycete isolated from *Eucalyptus camaldulensis* roots. Int J Syst Evol Microbiol. 2017; 67: 3077–3082. https://doi.org/10.1099/ijsem.0.002086 PMID: 28857736

82. Li J, Chen X, Li S, Zuo Z, Zhan R, He R. Variations of rhizospheric soil microbial communities in response to continuous *Andrographis paniculata* cropping practices. Bot Stud. 2020; 61: 18. https://doi.org/10.1186/s40529-020-00295-1 PMID: 32542518

83. Rahman MM, Abdullah RB, Wan Khadijah WE. A review of oxalate poisoning in domestic animals: tolerance and performance aspects. J Anim Physiol Anim Nutr (Berl). 2013; 97: 605–614. https://doi.org/10.1111/j.1439-0396.2012.01309.x PMID: 22548678

84. Tamer AÜ, Aragno M, Şahin N. Isolation and Characterization of a New Type of Aerobic, Oxalic Acid Utilizing Bacteria, and Proposal of *Oxalicibacterium flavum* gen. nov., sp. nov. Syst Appl Microbiol. 2002; 25: 513–519. https://doi.org/10.1078/07232020260517643 PMID: 12583711

85. Watanabe T, Osaki M. Role of organic acids in aluminum accumulation and plant growth in *Melastoma malabathricum*. Tree Physiol. 2002; 22: 785–792. https://doi.org/10.1093/treephys/22.11.785 PMID: 12184982

86. Sahin N, Portillo MC, Kato Y, Schumann P. Description of *Oxalicibacterium horti* sp. nov. and *Oxalicibacterium faecigallinarum* sp. nov., new aerobic, yellow-pigmented, oxalotrophic bacteria. FEMS Microbiol Lett. 2009; 296: 198–202. https://doi.org/10.1111/j.1574-6968.2009.01636.x PMID: 19459944

87. Rahman MM, Kawamura O. Oxalate Accumulation in Forage Plants: Some Agronomic, Climatic and Genetic Aspects. Asian-Australasian J Anim Sci. 2011; 24: 439–448. https://doi.org/10.5713/ajas.2011.10208

88. Xu Q, Li K, Ruan Y, Kong Y, Liu M, Ling N, et al. Distinct response patterns of soil bacteria to oxalate imply their role in buffering soil acidification: Evidence from red soils with long-term fertilisation regimes. L Degrad Dev. 2019; 30: 1632–1641. https://doi.org/10.1002/ldr.3353

89. Pascual J, Huber KJ, Foesel BU, Overmann J. *Tellurimicrobium*. Bergey's Manual of Systematics of Archaea and Bacteria. Wiley; 2017. pp. 1–5. https://doi.org/10.1002/9781118960608.gbm01430

90. Kumar V, Irfan M, Datta A. Manipulation of oxalate metabolism in plants for improving food quality and productivity. Phytochemistry. 2019; 158: 103–109. https://doi.org/10.1016/j.phytochem.2018.10.029 PMID: 30500595

91. Berner M, Krug D, Bihlmaier C, Vente A, Müller R, Bechthold A. Genes and Enzymes Involved in Caffeic Acid Biosynthesis in the Actinomycete *Saccharothrix espanaensis*. J Bacteriol. 2006; 188: 2666–2673. https://doi.org/10.1128/JB.188.7.2666–2673.2006

92. Li Z-H, Wang Q, Ruan X, Pan C-D, Jiang D-A. Phenolics and Plant Allelopathy. Molecules. 2010; 15: 8933–8952. https://doi.org/10.3390/molecules15128933 PMID: 21139532

93. de Albuquerque FS, Macías-Rodríguez MÁ, Búrquez A, Astudillo-Scalia Y. Climate change and the potential expansion of buffelgrass (*Cenchrus ciliaris* L., *Poaceae*) in biotic communities of Southwest United States and northern Mexico. Biol Invasions. 2019; 21: 3335–3347. https://doi.org/10.1007/s10530-019-02050-5

94. Henry C, John GP, Pan R, Bartlett MK, Fletcher LR, Scoffoni C, et al. A stomatal safety-efficiency trade-off constrains responses to leaf dehydration. Nat Commun. 2019; 10: 3398. https://doi.org/10.1038/s41467-019-11006-1 PMID: 31363097

95. Einhellig FA, Kuan L-Y. Effects of Scopoletin and Chlorogenic Acid on Stomatal Aperture in Tobacco and Sunflower. Bull Torrey Bot Club. 1971; 98: 155. https://doi.org/10.2307/2484031

96. Rai VK, Sharma SS, Sharma S. Reversal of ABA-Induced Stomatal Closure by Phenolic Compounds. J Exp Bot. 1986; 37: 129–134. https://doi.org/10.1093/jxb/37.1.129

97. Tombesi S, Nardini A, Frioni T, Soccolini M, Zadra C, Farinelli D, et al. Stomatal closure is induced by hydraulic signals and maintained by ABA in drought-stressed grapevine. Sci Rep. 2015; 5: 12449. https://doi.org/10.1038/srep12449 PMID: 26207993

98. Yang CM, Chang IF, Lin SJ, Chou CH. Effects of three allelopathic phenolics on chlorophyll accumulation of rice (*Oryza sativa*) seedlings: II. Stimulation of consumption-orientation. Bot Bull Acad Sin. 2004; 45: 119–125. https://doi.org/10.7016/BBAS.200404.0119

99. Bulgarelli D, Rott M, Schlaeppi K, Ver Loren van Themaat E, Ahmadinejad N, Assenza F, et al. Revealing structure and assembly cues for *Arabidopsis* root-inhabiting bacterial microbiota. Nature. 2012; 488: 91–95. https://doi.org/10.1038/nature11336 PMID: 22859207

100. Lundberg DS, Lebeis SL, Paredes SH, Yourstone S, Gehring J, Malfatti S, et al. Defining the core *Arabidopsis thaliana* root microbiome. Nature. 2012; 488: 86–90. https://doi.org/10.1038/nature11237 PMID: 22859206

101. Bulgarelli D, Schlaeppi K, Spaepen S, van Themaat EVL, Schulze-Lefert P. Structure and Functions of the Bacterial Microbiota of Plants. Annu Rev Plant Biol. 2013; 64: 807–838. https://doi.org/10.1146/annurev-arplant-050312-120106 PMID: 23373698

102. O'Brien FJM, Dumont MG, Webb JS, Poppy GM. Rhizosphere Bacterial Communities Differ According to Fertilizer Regimes and Cabbage (*Brassica oleracea* var. *capitata* L.) Harvest Time, but Not Aphid Herbivory. Front Microbiol. 2018; 9: 1–18. https://doi.org/10.3389/fmicb.2018.01620 PMID: 30083141

103. Tkacz A, Bestion E, Bo Z, Hortala M, Poole PS. Influence of Plant Fraction, Soil, and Plant Species on Microbiota: a Multikingdom Comparison. Bailey MJ, editor. MBio. 2020; 11: 1–17. https://doi.org/10.1128/mBio.02785-19 PMID: 32019791

104. Li X, Rui J, Mao Y, Yannarell A, Mackie R. Dynamics of the bacterial community structure in the rhizosphere of a maize cultivar. Soil Biol Biochem. 2014; 68: 392–401. https://doi.org/10.1016/j.soilbio.2013.10.017

105. Li X, Song Y, Yao S, Bian Y, Gu C, Yang X, et al. Can biochar and oxalic acid alleviate the toxicity stress caused by polycyclic aromatic hydrocarbons in soil microbial communities? Sci Total Environ. 2019; 695: 133879. https://doi.org/10.1016/j.scitotenv.2019.133879 PMID: 31425980

106. Liu J, Yu Z, Yao Q, Hu X, Zhang W, Mi G, et al. Distinct soil bacterial communities in response to the cropping system in a Mollisol of northeast China. Appl Soil Ecol. 2017; 119: 407–416. https://doi.org/10.1016/j.apsoil.2017.07.013

107. Qu F, ElOmari K, Wagner A, De Simone A, Beis K. Desolvation of the substrate-binding protein TauA dictates ligand specificity for the alkanesulfonate ABC importer TauABC. Biochem J. 2019; 476: 3649–3660. https://doi.org/10.1042/BCJ20190779 PMID: 31802112

108. Walters WA, Jin Z, Youngblut N, Wallace JG, Sutter J, Zhang W, et al. Large-scale replicated field study of maize rhizosphere identifies heritable microbes. Proc Natl Acad Sci. 2018; 115: 7368–7373. https://doi.org/10.1073/pnas.1800918115 PMID: 29941552

109. Wang D, Lin J, Lin J, Wang W, Li S. Biodegradation of Petroleum Hydrocarbons by *Bacillus subtilis* BL-27, a Strain with Weak Hydrophobicity. Molecules. 2019; 24: 3021. https://doi.org/10.3390/molecules24173021 PMID: 31438460

110. Houlden A, Timms-Wilson TM, Day MJ, Bailey MJ. Influence of plant developmental stage on microbial community structure and activity in the rhizosphere of three field crops. FEMS Microbiol Ecol. 2008; 65: 193–201. https://doi.org/10.1111/j.1574-6941.2008.00535.x PMID: 18616582

111. Wen T, Zhao M, Yuan J, Kowalchuk GA, Shen Q. Root exudates mediate plant defense against foliar pathogens by recruiting beneficial microbes. Soil Ecol Lett. 2021; 3: 42–51. https://doi.org/10.1007/s42832-020-0057-z

112. Qiao Q, Wang F, Zhang J, Chen Y, Zhang C, Liu G, et al. The Variation in the Rhizosphere Microbiome of Cotton with Soil Type, Genotype and Developmental Stage. Sci Rep. 2017; 7: 3940. https://doi.org/10.1038/s41598-017-04213-7 PMID: 28638057