

DATA NOTE

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Amplicon-based metagenomic survey of microbes associated with the organic and inorganic rhizosphere soil of *Glycine max* L.

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Abstract

Objectives The metagenomic dataset of 16S rRNA and ITS gene amplicons of DNA were obtained from the cultivated soybean rhizosphere of organic and inorganic treatments. The organic treatments consisted of poultry waste, and cow dung treatments while the inorganic consisted of samples from untreated soybean plots and the bulk. Amplicon sequencing was performed on the Illumina platform, and the raw sequence data were processed and analyzed using Quantitative Insights Into Microbial Ecology (QIIME 2 version 2019.1.).

Data description The analysis revealed a metagenomic library from soybean rhizospheric soils, providing insights into diversity and distribution of the bacterial and fungal community diversities. The most predominant bacteria phylum taxa across the treatments were Proteobacteria, Firmicutes, Actinobacteriota and Bacteriodota, while those for fungi were Ascomycota, Basidiomycota and Glomeromycota. The dataset provides insights into how different organic fertilization sources affect the structure, composition, and diversity of the microbiome in the soybean rhizosphere. The sequences have been deposited in the Sequence Read Archive (SRA) of the National Center for Biotechnology Information (NCBI) with assigned bioproject accession numbers; 16S rRNA (SRP540791) and ITS (SRP541849).

Keywords 16S rRNA gene, Amplicon sequencing, Cow dung, ITS gene, Metagenome, Microbial communities, PCR amplification, Poultry waste, QIIME

Objective

Soybean (*Glycine max* L.) is one of the important legumes across the globe, including South Africa. Its production using organic treatment sources has been encouraged due to their ecofriendly nature [1]. The dataset provides insight into the influence of poultry waste and cow dung as organic soil treatments in soybean cultivation in comparison to the untreated soybean fields. The resulting effects on the community structure and diversity of microbial communities within the soybean rhizosphere of each treatment were evaluated through 16S ribosomal RNA (16S rRNA gene) and Internal transcribe

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Table 1 Overview of data files/data sets

Label	Name of data file/data set	File types (file extension)	Data repository and identifier (DOI or accession number)
Data file 1	Sequence information and Microbial community structure	PDF file (.pdf)	Figshare (https://doi.org/10.6084/m9.figshare.28760330) [10]
Data set 1	Bacterial profiling and functions in soybean rhizosphere under organic fertilization	fastQ files	NCBI SRA SRP540791 https://identifiers.org/ncbi/insdc.sra:SRP540791 [11]
Data set 2	Fungal profiling and functions in soybean rhizosphere under organic fertilization	fastQ files	NCBI SRA SRP541849 https://identifiers.org/ncbi/insdc.sra:SRP541849 [12]

spacer (ITS) amplicon metagenomic sequencing. This dataset explores the diversity, composition, and prevalence of rhizospheric bacteria under different organic and inorganic applications, and the bulk. Raw sequence data available in fastq.gz format have been deposited in the National Center for Biotechnology Information (NCBI) Sequence Read Archive (SRA) database with accession number SRP540791 for 16S rRNA and SRP541849 for ITS sequences. This dataset holds value for identifying novel microbial genes and bioactive compounds that promote plant growth, which could be leveraged to enhance soybean yield, resilience, and sustainable agricultural practices. Additionally, understanding microbial dynamics under different fertilization strategies can aid in developing soil management approaches that support agroecosystem health and productivity [2].

Data description

Rhizospheric soil samples tightly bound to the root surface were collected from the cultivated soybean (*Glycine Max* L.) farm sites at the North-West University farm, Molelwane, Mahikeng (25° 48' 11.577"S, 25° 35' 53.762"E), which has an average rainfall of 540 mm per year. The samples were carried out in the soybean farms under different organic treatments consisting of poultry litter waste, cow dung, untreated soybean plot (control), and the bulk soil. These soil samples were collected in three replicates each, at a 5 cm diameter, and 10-cm depth, and transported in cooler boxes on ice to the laboratory. Samples were weighed while extraction of the whole community Deoxyribonucleic Acid (DNA) was carried out using the NucleoSpin Soil kit (Macherey-Nagel, Germany) by adhering to the manufacture's instruction. The sequence of the bacterial 16S ribosomal RNA gene was amplified using the universal primer pair regions 16S V4 and 16S V4-V5 with the primers 515F (5'-GTGCCAGCMGCCGCGGTAA-3') and 907R (5'-CCGTCAATTCCTTTGAGTTT-3'), while the internal transcribed spacer regions (ITS1 and ITS2) of the fungal ribosomal DNA were amplified using the primers pair ITS1F (5'-CTTGGTCA TTTAGAGGAAGTAA-3') and ITS2 (5'-GCTGCGTTC TTCATCGATGC-3'). Amplicon metagenomic sequencing was carried out using NovaSeq 6000 system (Novogene, Singapore). Quantified libraries were pooled and sequenced on Illumina platforms [3], following standard

protocol. Data obtained from this analysis were used to determine the structural diversity and functional profiles of bacteria and fungi in the soybean rhizosphere, across the treatments. The raw sequence dataset in FASTQ format was denoised and chimeras removed using DADA2 plugin within QIIME 2 version 2019.1 [4]. Amplicon sequence variants (ASVs) were classified taxonomically using Silva version 138.1 reference database for 16S rRNA gene [5], while a pre-trained Naïve Bayes classifier using the Unite_INSDC version 8.0 (2018) for ITS [6]. PICRUST2 [7] and FUNGuild [8] were employed for the functional annotation of the 16S rRNA and ITS genes respectively. Downstream analyses included diversity metrics (alpha and beta diversity), visualization of community composition, and phylogenetic tree construction using the SATé-Enabled Phylogenetic Placement (SEPP) [9] plugin for accurate placement of Amplicon Sequence Variants (ASVs) within the phylogenetic framework. The pipeline facilitated robust and reproducible insights into microbial community structure and function.

The dataset is made up of raw sequence data available in fastq.gz obtained through Amplicon sequencing of soybean rhizosphere metagenome. The sequence information on dataset and an overview on the microbial community structure are represented in Data file 1. All the obtained datasets in fastq.gz file was deposited at the NCBI-SRA database with accession number SRP540791 and SRP541849 for bacteria (Data set 1) and fungi (Data set 2) respectively in Table 1.

Limitations

Not applicable.

Abbreviations

DNA	Deoxyribonucleic Acid
NCBI	National Center for Biotechnology Information
SRA	Sequence Read Archive
DADA2	Divisive Amplicon Denoising Algorithm 2
QIIME	Quantitative Insights into Microbial Ecology
ITS	Internal Transcribed Spacer
16S rRNA gene	16S ribosomal RNA
SEPP	SATé-enabled Phylogenetic Placement
ASVs	Amplicon Sequence Variants

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Authors' contributions

OOB: Conceptualization, Supervision, Resources, Investigation, Writing– review & editing, Funding acquisition. IEO: Methodology, Visualization, Investigation, Software, Formal analysis, Writing– original draft, Writing– review & editing. AOA: Methodology, Investigation, Software, Formal analysis, Data curation, Visualization, Writing– review & editing.

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Data availability

The data described in this Data note can be freely and openly accessed on NCBI SRA under <https://www.ncbi.nlm.nih.gov/sra/?term=SRP540791> and <https://www.ncbi.nlm.nih.gov/sra/?term=SRP541849>. Please see Table 1 and references [7 and 8] for details and links to the data.

Declarations

Ethics approval and consent to participate

The current work follows the ethical requirements for publication. It does not involve human subjects, animal experiments, or any data collected from social media platforms.

Consent for publication

All authors approved the manuscript for publication.

Competing interests

The authors declare no competing interests.

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References

1. Ajiboye TT, Ayangbenro AS, Babalola OO. Functional diversity of microbial communities in the soybean (*Glycine max* L.) rhizosphere from free State, South Africa. *Int J Mol Sci*. 2022;23(16):9422.
2. Akanmu AO, Olowe OM, Phiri AT, Nirere D, Odeboode AJ, Karemera Umuhoza NJ, et al. Bioresources in organic farming: implications for sustainable agricultural systems. *Horticulturae*. 2023;9(6):659.
3. Bokulich NA, Mills DA. Improved selection of internal transcribed spacer-specific primers enables quantitative, ultra-high-throughput profiling of fungal communities. *Appl Environ Microbiol*. 2013;79(8):2519–26.
4. Bolyen E, Rideout JR, Dillon MR, Bokulich NA, Abnet CC, Al-Ghalith GA, et al. Reproducible, interactive, scalable and extensible Microbiome data science using QIIME 2. *Nat Biotechnol*. 2019;37(8):852–7.
5. Callahan BJ, Grinevich D, Thakur S, Balamotis MA, Yehezkel TB. Ultra-accurate microbial amplicon sequencing with synthetic long reads. *Microbiome*. 2021;9(1):130.
6. Bokulich NA, Kaehler BD, Rideout JR, Dillon M, Bolyen E, Knight R, et al. Optimizing taxonomic classification of marker-gene amplicon sequences with QIIME 2's q2-feature-classifier plugin. *Microbiome*. 2018;6:1–17.
7. Yan Z, Wang H, Wang L, Liu X, Chen X, Liu D, et al. Functional responses of giant panda gut microbiota to high-fiber diets. *Ursus*. 2024;2024(35e5):1–9.
8. Tanunchai B, Ji L, Schroeter SA, Wahdan SFM, Hossen S, Delelegn Y, et al. FungalTraits vs. FUNGuild: comparison of ecological functional assignments of leaf-and needle-associated fungi across 12 temperate tree species. *Microb Ecol*. 2023;85(2):411–28.
9. Tsang CTT, Hui TKL, Chung NM, Yuen WT, Tsang LM. Comparative analysis of gut Microbiome of Mangrove brachyuran crabs revealed patterns of phyllosymbiosis and codiversification. *Mol Ecol*. 2024;33(12):e17377.
10. Osuji IE, Akanmu AO, Babalola OO. Sequence information and microbial community structure. 2025. Figshare. <https://doi.org/10.6084/m9.figshare.28760330>.
11. NCBI. Sequence Read Archive. 2025. <https://identifiers.org/ncbi/insdc.sra:SRP540791>.
12. NCBI. Sequence Read Archive. 2025. <https://identifiers.org/ncbi/insdc.sra:SRP541849>.

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