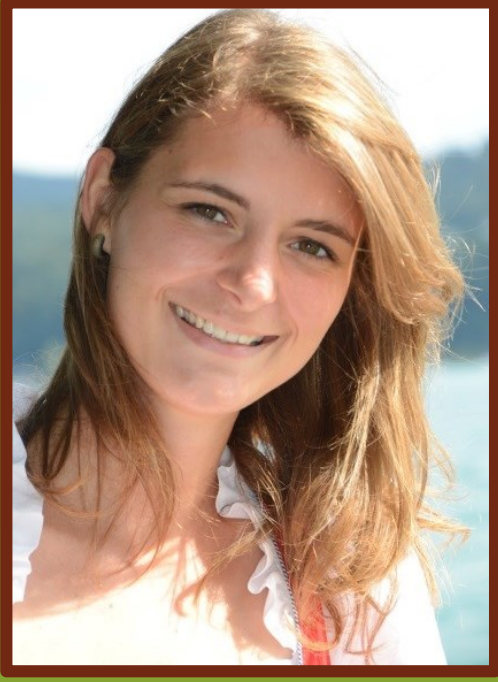


# Exploring the Microbiome of Ugandan Leafy Greens



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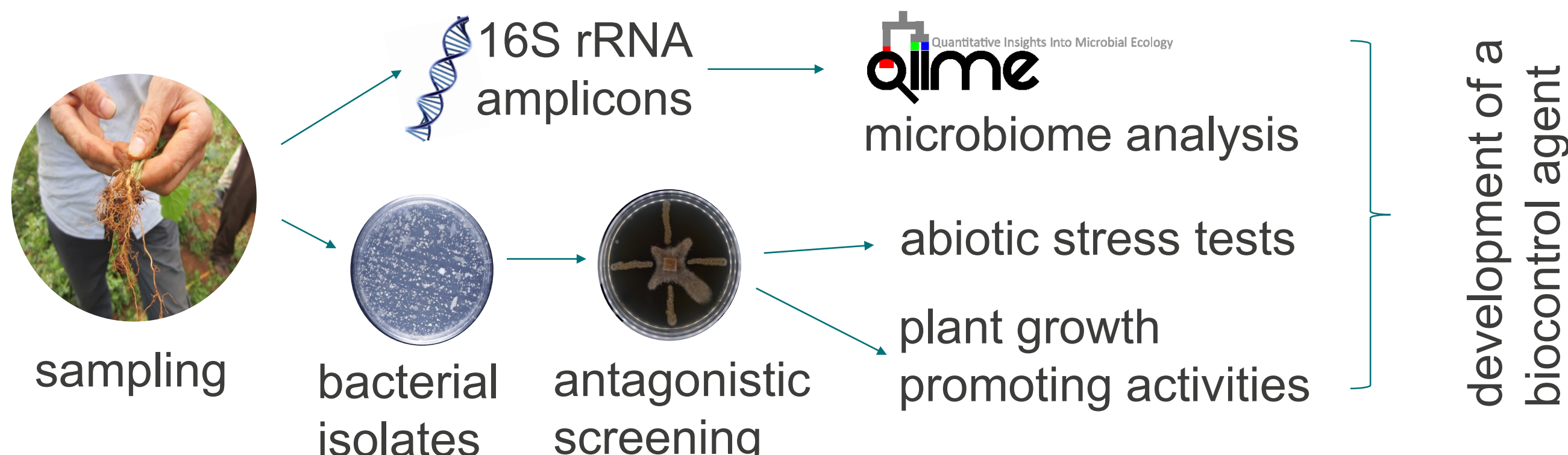
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## Background

Indigenous leafy greens, such as **okra** (*Abelmoschus esculentus*), **garden huckleberry** (*Solanum scabrum*), **spiderwisp** (*Gynandropsis gynandra*), and **black jack** (*Bidens pilosa*) struggle less from pests and are generally more robust to abiotic stresses, compared to cultivated crops [1]. The robustness of the host plant is strongly depending on its microbiome [2].

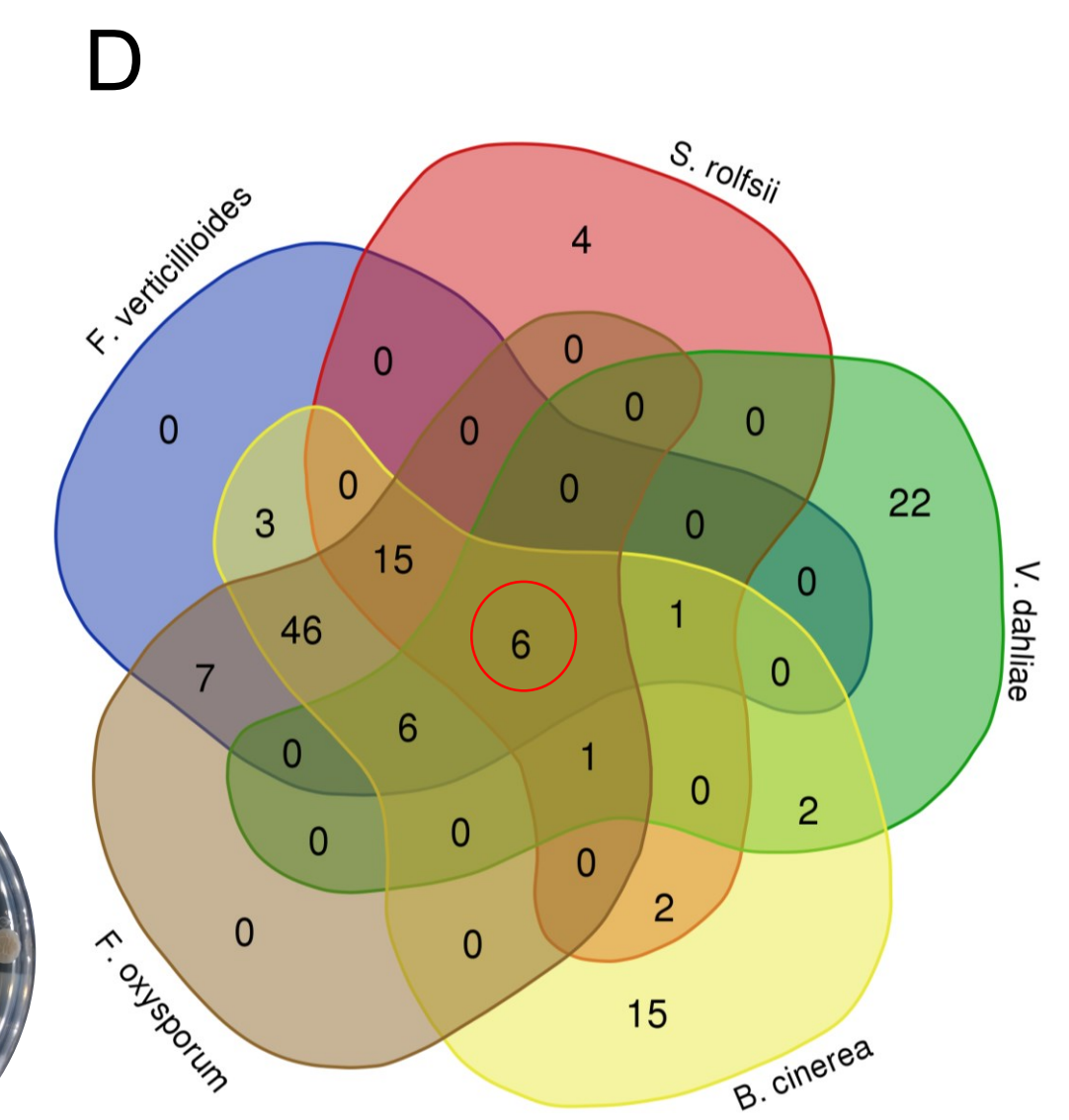
## Material and Methods



The bacterial microbiome was analyzed, based on 16S rRNA gene fragment sampling and bioinformatic analysis. Additionally, plant-associated bacterial isolates were screened against phytopathogenic fungi (*Botrytis cinerea*, *Fusarium oxysporum*, *Fusarium verticillioides*, *Sclerotium rolfsii* and *Verticillium dahliae*) and for their ability to resist abiotic stresses as well as to promote plant growth.

## Crop-associated Phytopathogen Antagonists

In total, 412 crop-associated bacterial isolates were tested against phytopathogenic fungi in **dual culture assays** (A, B, C). Out of them, 130 were highly effective against one or more of the tested fungi (D).



Six strains showed strong antagonism towards all five tested phytopathogens (D). Based on 16S rRNA sequences, they were assigned to **Bacillus sp.** and **Sphingomonas sp.**

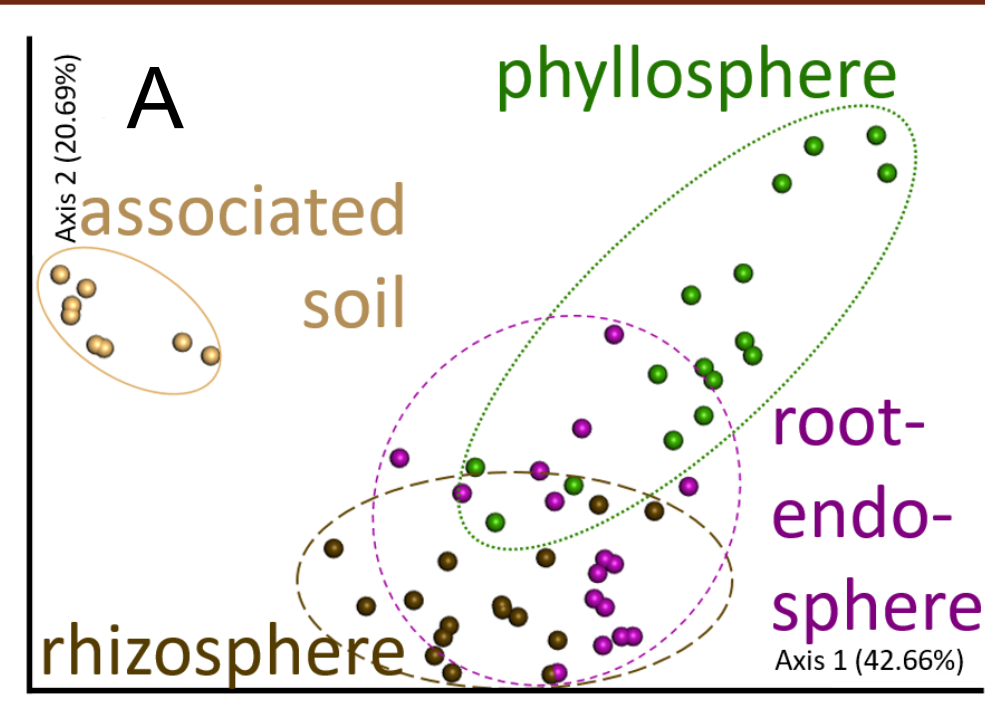
## Strain Characteristics

The suggested future biocontrol agents **promote plant growth** and are able to **stand abiotic stress**.

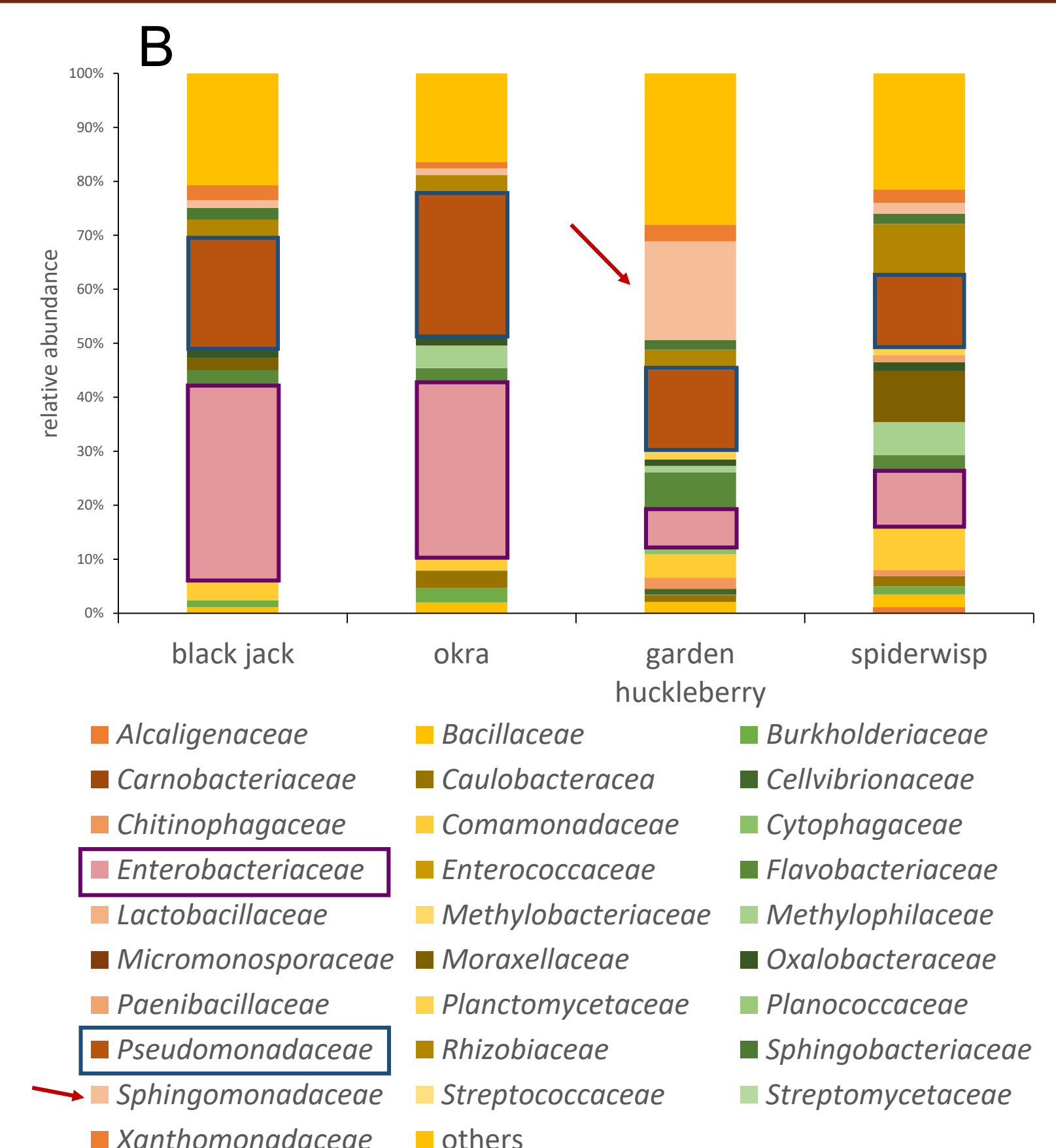
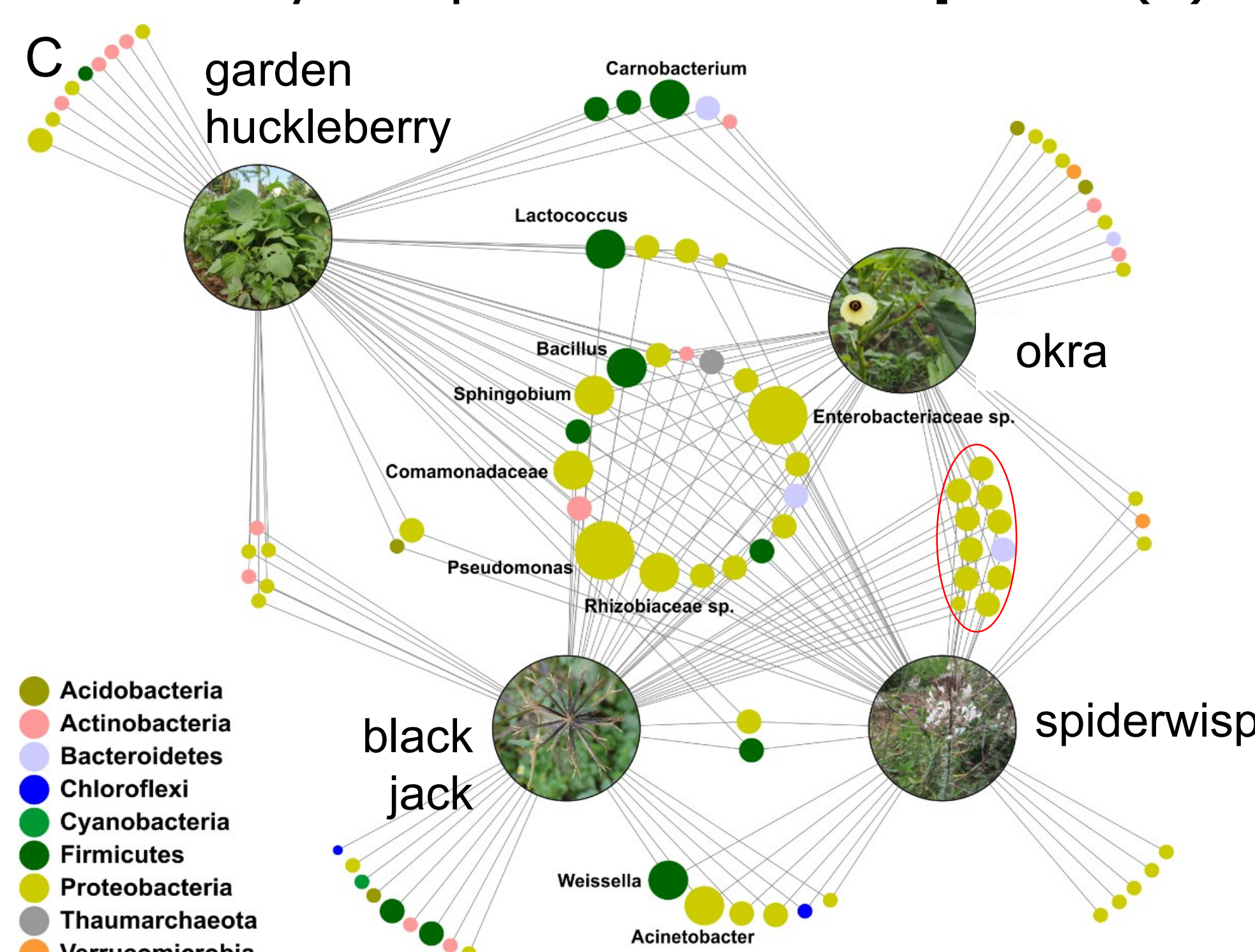
strain name	PGP (roots) [%]	PGP (shoots) [%]	drought [months]	rO [mmol H <sub>2</sub> O <sub>2</sub> ]	salinity [%NaCl]
<i>Bacillus sp.</i> soil-I-11	83.9±3	60.8±5	> 3	0	6
<i>Bacillus sp.</i> soil-I-14	82.3±2	58.1±3	> 3	2	7
<i>Sphingomonas sp.</i> soil-I-45	-11.3±3	10.8±2	> 3	0	11
<i>Sphingomonas sp.</i> ORE-30	50.0±7	14.3±7	> 3	0.1	10
<i>Sphingomonas sp.</i> ORE-44	21.4±7	-9.5±5	> 3	0	11
<i>Bacillus sp.</i> NSRE-37	85.7±0	42.9±1	> 3	0.9	0

**PGP**: plant growth promotion of 15 d old seed-primed tomato plants (% increase in fresh weight relative to control); **drought**: culture showing >10<sup>5</sup> CFUs/ml after desiccation; **rO**: highest tolerable concentration of reactive oxygen (H<sub>2</sub>O<sub>2</sub>) in growth medium (LB); **salinity**: maximum tolerable concentration of NaCl (w/v).

## Microbiome Composition



PCoA plot (weighted UniFrac) and ANOSIM revealed microbial communities rather to be **microhabitat-specific** ( $p \leq 0.001$ ) than plant-specific ( $p = 0.064$ ) (A). All microhabitats were dominated by *Pseudomonadaceae* and *Enterobacteriaceae*, with highest differences in bacterial community composition in **rhizosphere** (B).



**Garden huckleberry** showed significant differences in both alpha (Shannon H = 6.2 ± 0.1 vs. 5.2-5.9) and beta diversity indices. Bacterial network of core taxa showed eleven families that are shared between all leafy greens except garden huckleberry (C).

## Conclusion

The core microbiome of **garden huckleberry** revealed to be significantly **different** to the three other leafy greens. We hypothesize this effect to be attributable to the high amount of biologically active secondary metabolites found within the family *Solanaceae* [3]. **Microhabitat** was found to be the driving force for **microbial diversity**. Plant-specificity of the bacterial community could only be found within microhabitats and differences were most significant within rhizosphere. We isolated six bacterial strains, belonging to ***Sphingomonas sp.*** and ***Bacillus sp.***, showing antagonism towards phytopathogens, tolerance towards **abiotic stresses** and **plant growth promotion**. We suggest their application as **native biocontrol agents in East African agriculture**.

For an overview of the whole project, visit Poster 35

[1] Venkateswarlu, B., K. Shanker Arun, Shanker Chitra, and M. Maheswari. *Crop Stress and Its Management: Perspectives and Strategies*. Springer Science & Business Media, 2011.

[2] Schläppli, Klaus, and Davide Bulgarelli. 'The Plant Microbiome at Work'. *Molecular Plant-Microbe Interactions* 28, no. 3 (16 December 2014): 212–17. <https://doi.org/10.1094/MPMI-10-14-0334-FI>.

[3] Castillo-Sánchez, Luis Enrique, Juan José Jiménez-Osorio, and María América Delgado-Herrera. 'Secondary Metabolites of the Annonaceae, Solanaceae and Meliaceae Families Used as Biological Control of Insects'. *Tropical and Subtropical Agroecosystems* 12, no. 3 (2010): 445–62.