Plant-microbe interactions

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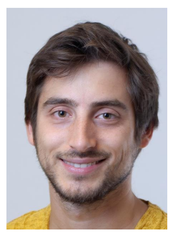
Background: Plant-microbe interactions are key to evolution and survival in diverse ecosystems. Healthy plants are colonised by a great diversity of microorganisms, referred to as the plant microbiota, which have profound effects on plant growth and fitness. Plants sense the microbes through a variety of membrane-localised receptors. Recognition at the plasma membrane level initiates a specific response in the plant host that impacts the structures and functions of the associative microbial communities. Identifying and understanding the mechanisms underlying these interactions will enable us to improve plant health and crop yields in a sustainable manner, while reducing the carbon footprint due to intensive crop growth systems based on energetically and climate expensive chemicals.

Root microbiome

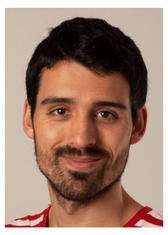
Rationale: The molecular programs that control root infection by commensals are largely unknown. We have established extensive microbial collections (SynComs) from different plant hosts grown in natural soils. These SynComs are inoculated on wild type and mutant plants to identify plant genes involved in microbiota assembly and molecular determinants of host preference.

Objectives:
- Identify a core microbiome efficient for endophytic colonisation.
- Identify plant and bacterial functions important for root microbiota assembly.

Methods:
- Plant physiology and microbiology techniques
- 16S rRNA and shotgun metagenomics
- DNA library preparation for Illumina sequencing
- Bioinformatic analyses of bacterial communities
- Programming and automation of biological analyses
- Bacterial gene enrichment at community level



Florian



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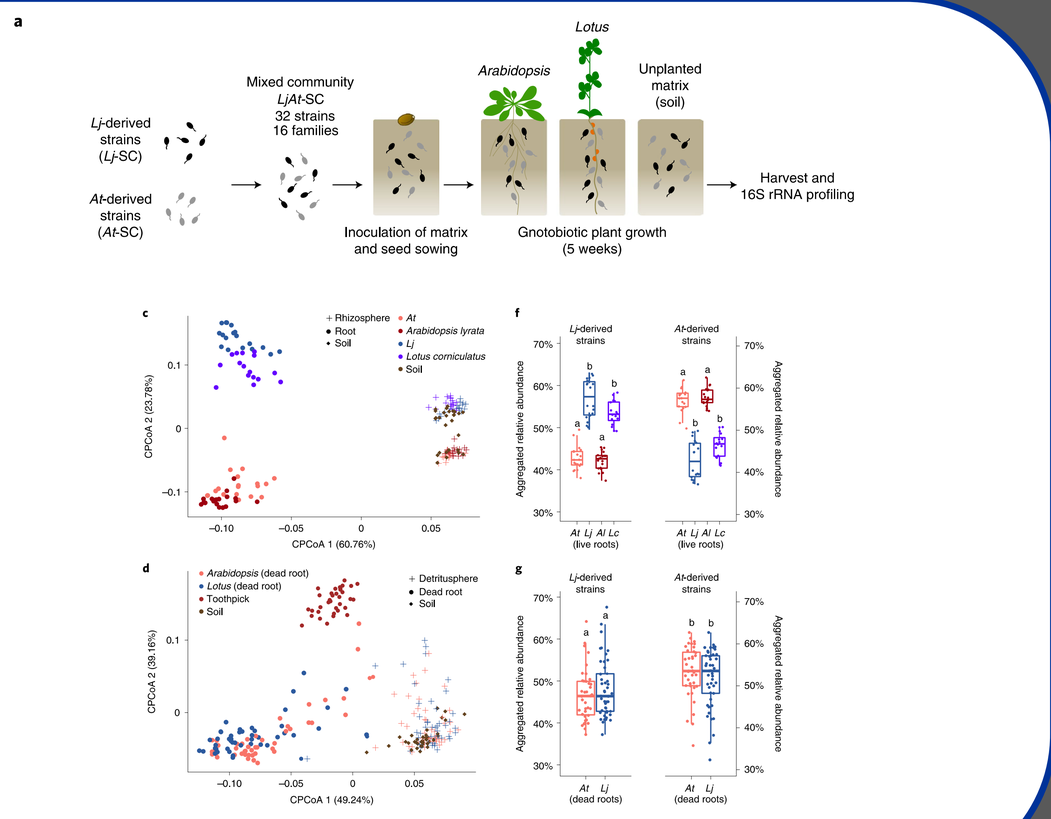


Wippel, K., Tao, K., Niu, Y. *et al.* (2021). Nature Microbiology, 6, 1150–1162.

Bacterial genetics

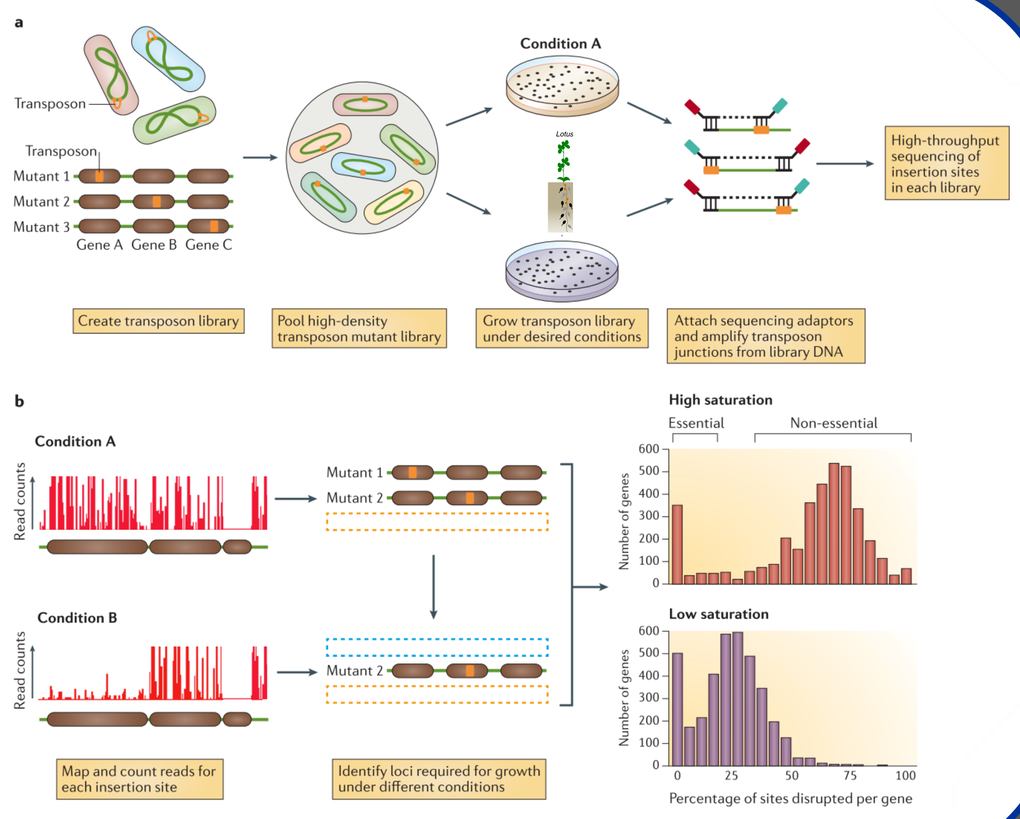
Rationale: The underlying bacterial components that contribute to host preference or community interactions within the root microbiota remain unknown. Investigation of identified strains from the *Lotus japonicus* SynCom enables comprehensive exploration into the genetic factors of plant-associated microorganisms that contribute to colonisation and survival in different plant compartments.

Objectives:
- Identify microbial genes important for plant colonisation.
- Identify microbial genes that control the metabolic and signalling networks.

Methods:
- General microbiology techniques
- Bacterial genome analyses, bioinformatics, and *in silico* construct design
- PCR, plasmid cloning, Sanger sequencing analysis
- Directed mutagenesis of bacteria (gene KOs)
- Transposon insertion sequencing (Tn-seq & Barseq)

Shaun

Eber



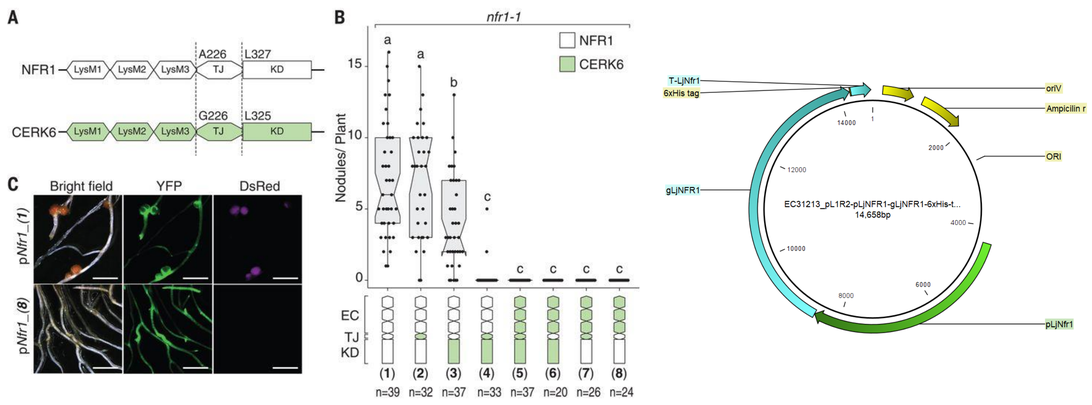
Modified from Chao, M. C., Abel, S., Davis, B. M., & Waldor, M. K. (2016). Nature Reviews Microbiology, 14(2), 119-128.

Plant receptors and signalling

Background: Legumes recognise and respond specifically to symbionts and pathogens that produce chitin-based ligands. LysM receptor kinases recognise different microbial carbohydrates and initiate immune or symbiotic pathways. In *Lotus japonicus* **NFR1/5** perceive rhizobial **Nod factor** and initiate symbiosis, while **CERK6** perceives **chitin** and activates defence mechanisms.

Objectives:
- Identify distinct motifs in LysM receptors controlling symbiosis or immune signalling.
- Understand how dynamics of signalling from LysM receptors lead to symbiosis or immunity.

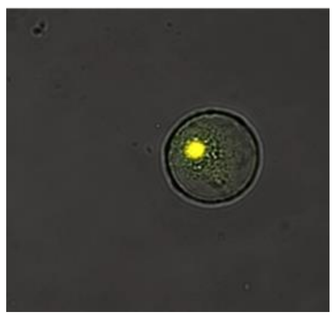
Methods:
- Golden Gate design and cloning in *E. coli*
- PCR, Sanger sequencing analysis
- Transformation of model plants, phenotypical analysis
- Microscopy (bright field, fluorescent, confocal)
- Signalling in single root cells (protoplasts)

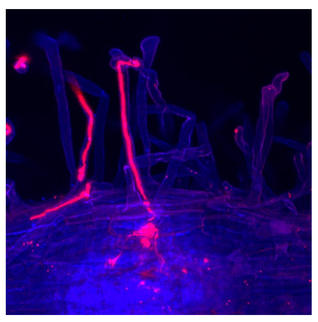


Bozsoki, Z., Gysel, K., Hansen, S.B. *et al.* (2020). Science, 369, 663-670.

Maria

Magda





Publications relevant for the different projects:
1 - Wippel, K., Tao, K., Niu, Y. *et al.* (2021). Nature Microbiology, 6, 1150–1162.
2 - Trivedi, P., Leach, J.E., Tringe, S.G. *et al.* (2020). Nature Reviews Microbiology, 18, 607-621.
3 - Cain, A.K., Barquist, L., Goodman, A.L. *et al.* Nat Rev Genet 21, 526–540 (2020).
4 - Bozsoki, Z., Gysel, K., Hansen, S.B. *et al.* (2020). Science, 369, 663-670.
5 - Bozsoki, Z., Cheng, J., Feng, F. *et al.* (2017). PNAS, 114, E8118-E8127.

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