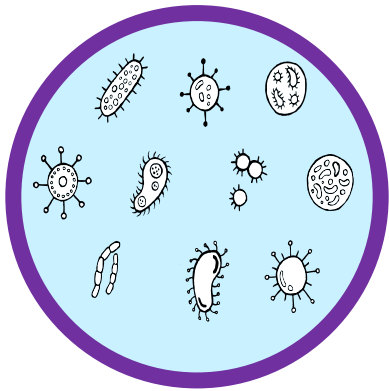


WHAT ARE ENDOPHYTIC BACTERIA?



ENDOPHYTIC
BACTERIA

- Endophytes are non-pathogenic microbial organisms that live inside plant tissues for at least part of their life cycles;
- They play an important role in plant growth, development and resistance to biotic and abiotic stresses;

<https://doi.org/10.3389/fmicb.2017.02552>

→ We can enhance phytoremediation by selecting the right isolates that can improve plant performance, such as plant growth promoters and/or degraders of contaminants, like antibiotics.

PROTOCOL



Figure 1: *Phragmites australis* exposed to a mix of antibiotics (1 PPM, each) for a period of 21 days.



ISOLATION OF CULTIVABLE ENDOPHYTIC BACTERIA

1. wash the root as a whole with water
2. slice the root into 2–3 cm pieces from several areas (ca. 30 pieces)
3. wash with miliQ-water (2x)
4. rinse with ethanol (70%) for 1 min
5. wash with NaOCl (3%) for 5 min
6. rinse with ethanol (70%) for 30 sec
7. wash with autoclaved dWater (2x)
8. pestle in a glass mortar in 2 mL of sterile water
9. dilute ten-fold (6 x) and plate into an agar plate (the same for the negative control)

BACTERIAL CHARACTERIZATION



41 isolates

**Plant growth-
promoting traits**

Ammonia production; Indole acetic acid
production; phosphate solubilisation;
siderophore production

**Morphological
identification**

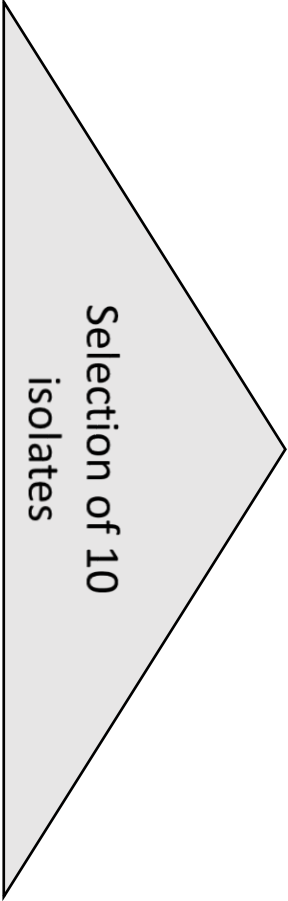
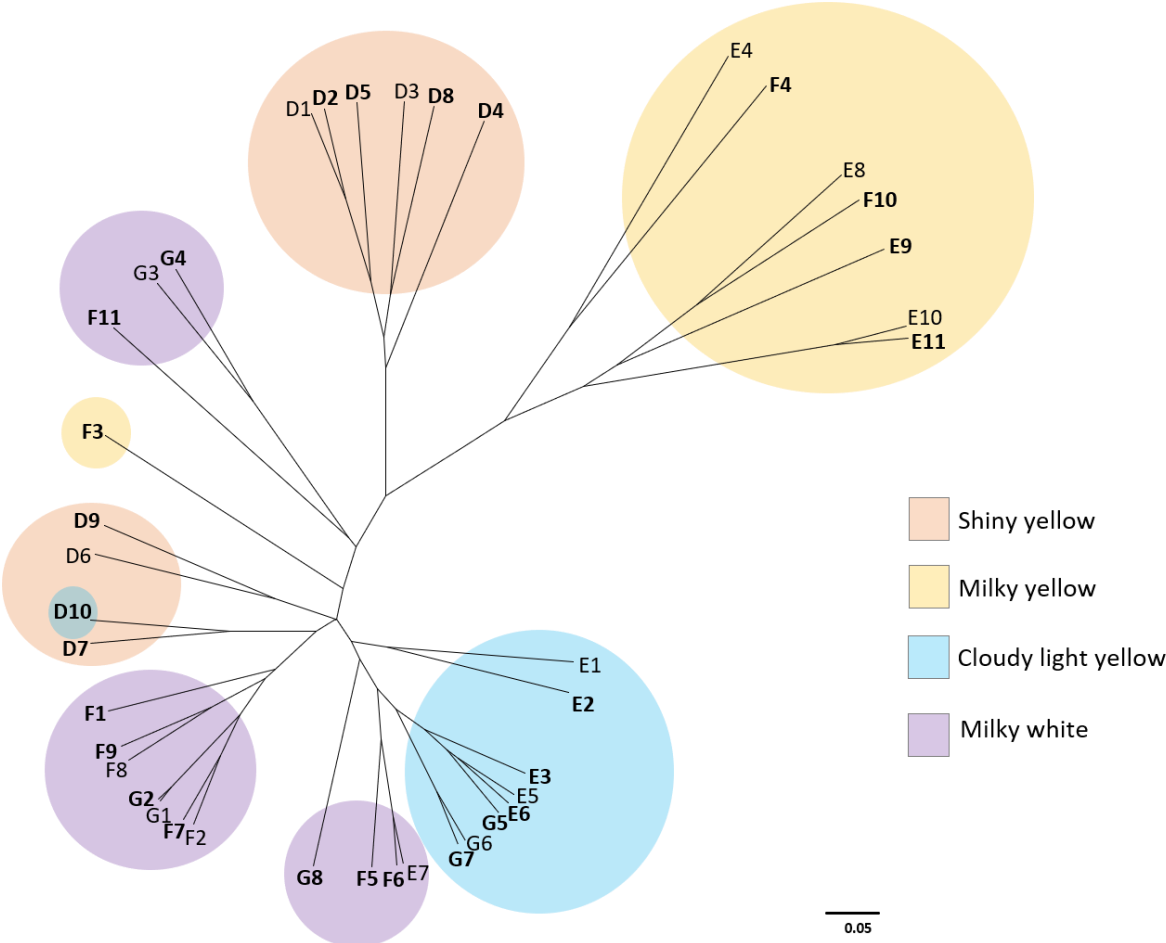
Form (circular, irregular, filamentous)
Elevation (raised, convex, flat, umbonate)
Margin (entire, undulate, filiform, curled)
<https://guides.baker.edu/c.php?g=303096&p=2022446#s-lg-box-6171053>

**Molecular
identification**

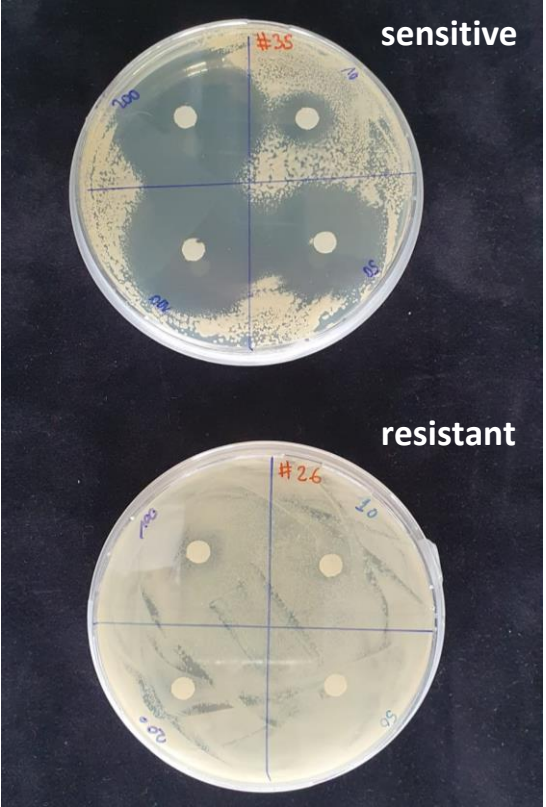
DNA fingerprint
16S sanger sequence

ENDOPHYTIC BACTERIA (SELECTION)

DENDROGRAM WITH THE 41 ISOLATES + MORPHOLOGICAL CHARACTERISTICS



DIFFUSION DISC TEST:
Selection of antibiotic resistant bacteria (i.e. antibiotic degraders)



LIST WITH THE ISOLATED ENDOPHYTIC BACTERIA

Name	% identify	GenBank	Description	Risk group	Antibiotic resistant?
<i>Chitinophaga pinensis</i>	99.49	KU305719.1		1	no
<i>Herbaspirillum huttiense</i>	100.00	MH283846.1	Gram-negative	1	yes
<i>Beijerinckia fluminensis</i>	99.92	MK872339.1	free-living nitrogen-fixing aerobic	1	yes
<i>Pseudomonas nitritireducens</i>	99.44	MN889029.1	nitrite reduction bacterium isolated from wheat soil	1	yes
<i>Bosea sp.</i>	99.45	MN067645.1		-	no
<i>Sphingopyxis chilensis</i>	98.68	LT673851.1	chlorophenol-degrading bacterium that accumulates polyhydroxyalkanoate	1	no
<i>Sphingobium sp.</i>	98.58	DQ376584.1		-	no
<i>Stenotrophomonas rhizophila</i>	99.85	CP050062.1	plant-associated bacterium with antifungal properties	na	yes
<i>Achromobacter denitrificans</i>	97.67	CP053987.1	Gram-negative; isolated from soil;	2	yes

DO YOU HAVE QUESTIONS?
IDEAS?
COMMENTS?



Don't hesitate to contact us!



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