

ENDOPHYTIC BACTERIA FROM AROMATIC PLANTS AND THEIR POTENTIAL FOR BIOINOCULATION ON GREEN ROOFS VEGETATION

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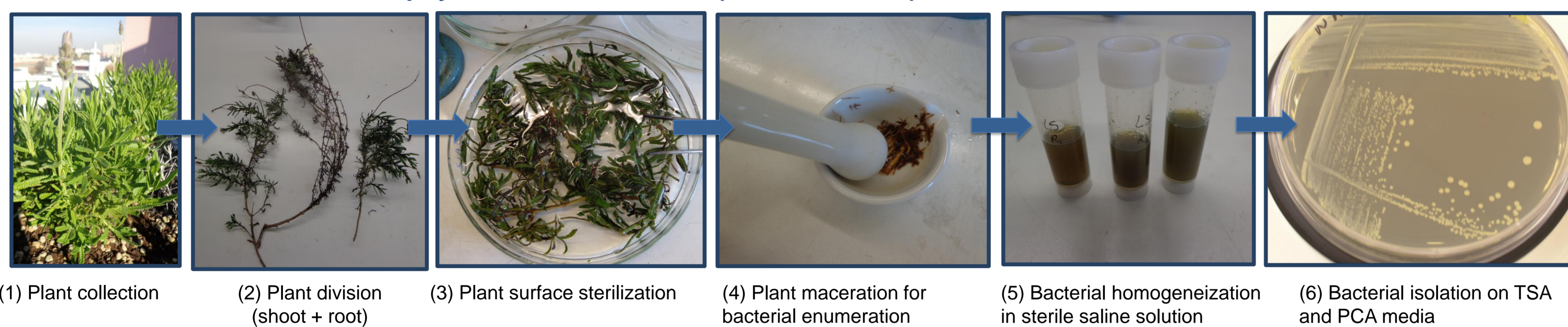
Introduction

Green roofs are a type of multilayer construction that uses vegetation on top of buildings. In the last decade, their use has become more frequent due to the environmental advantages they offer in impervious urban areas, such as stormwater retention, removal of atmospheric pollutants, attenuation of the urban heat island effect, among others. Rooftops covers a significant area of hard surfaces in cities, being thus an alternative green technology to bring vegetation back to urban areas, and to take advantage of their positive effects in the nearby microclimate. Selection of vegetation able to growth on the harsh environment of a rooftop is of major importance for the successful establishment of a green roof.

The use of plant growth promoting bacteria (PGPB) as bioinoculants is a sustainable alternative to improve plant establishment and growth on green roofs. The present study aimed to isolate endophytic bacteria from *Lavandula dentata* L. plants and to characterize isolates for their *in vitro* growth promoting traits. The effect of the inoculation of a selected mixture of four endophytic bacteria in aromatic plants development is being monitored on two established green roofs pilot systems.

Research Setup

Lavandula dentata L. - Endophytic bacteria isolation (shoot and root)



Aromatic plants:

- ✧ *Armeria maritima*
- ✧ *Santolina chamaecyparissus*
- ✧ *Santolina Lemon Queen*
- ✧ *Satureja montana*
- ✧ *Festuca glauca*
- ✧ *Sempervivum tectorum*

Plant growth promotion (PGP) traits of bacterial endophytes:

- ✓ Indol acetic acid (IAA)
- ✓ Siderophores
- ✓ Ammonia
- ✓ Phosphate solubilization
- ✓ Extracellular enzymes (proteases, cellulases, pectinases and lipases)

Endophytic selection for inocula

- LR 1-9 *Pseudomonas graminis*
- LR 2-11 *Paenibacillus kribbensis*
- LS 2-1 *Pseudomonas congelans*
- LS 1-2 *Bacillus aryabhatai*

Results and Discussion

Table 1 – Phylogenetic affiliation to the closest relative and characterization of bacterial endophytes isolated from *Lavandula dentata* L.

Strain	NCBI accession No.	Phylogenetic affiliation	Closest E/Taxone match (accession number)	Similarity (%)
LR 1-1	KP407086	γ-Proteobacteria	<i>Pseudomonas brassicacearum</i> subsp <i>neourantica</i> ATCC 49054 (EU391388)	100
LR 1-2	KP407087	α-Proteobacteria	<i>Rhizobium nepotum</i> 397 (FR870231)	100
LR 1-5	KP407088	γ-Proteobacteria	<i>Pseudomonas moorei</i> RW101 (AM295566)	100
LR 1-6B	KP407089	β-Proteobacteria	<i>Variovorax boronicumulans</i> BAM-48 (AB300597)	100
LR 1-7	KP407090	β-Proteobacteria	<i>Variovorax soli</i> GH9-3 (DQ432053)	98
LR 1-9	KP407091	γ-Proteobacteria	<i>Pseudomonas graminis</i> DSM 11363 (Y11150)	99
LR 1-11	KP407092	Firmicutes	<i>Bacillus cereus biovar toyoi</i> CNCM 1-1012 (AJ310100)	100
LR 1-12	KP407093	α-Proteobacteria	<i>Caulobacter vibrioides</i> CBS1 (AJ009957)	100
LR 1-13	KP407094	β-Proteobacteria	<i>Variovorax boronicumulans</i> BAM-48 (AB300597)	100
LR 1-16	KP407095	γ-Proteobacteria	<i>Pseudomonas brassicacearum</i> subsp <i>neourantica</i> CIP 109457 (EU391388)	100
LR 2-1	KP407096	Firmicutes	<i>Bacillus thuringiensis</i> IAM 12077 (D16281)	100
LR 2-2	KP407097	γ-Proteobacteria	<i>Pseudomonas kilonensis</i> 520-20 (AJ292426)	100
LR 2-3	KP407098	γ-Proteobacteria	<i>Pseudomonas jessenii</i> CIP 105274 (AF98259)	99
LR 2-4	KP407099	Firmicutes	<i>Bacillus cereus biovar toyoi</i> CNCM 1-1012 (AJ310100)	100
LR 2-6	KP407100	Firmicutes	<i>Bacillus cereus biovar toyoi</i> CNCM 1-1012 (AJ310100)	100
LR 2-9	KP407101	γ-Proteobacteria	<i>Pseudomonas punonensis</i> LMT03 (JQ344321)	98
LR 2-10	KP407102	β-Proteobacteria	<i>Variovorax boronicumulans</i> BAM-48 (AB300597)	100
LR 2-11	KP407103	Firmicutes	<i>Paenibacillus kribbensis</i> AM49 (AF391123)	99
LR 3-1	KP407104	γ-Proteobacteria	<i>Pseudomonas frederiksbergensis</i> JAJ28 (AJ249382)	100
LR 3-2	KP407105	γ-Proteobacteria	<i>Pseudomonas fuscovaginae</i> ICMP 5940 (FJ483519)	99
LR 3-3	KP407106	Firmicutes	<i>Bacillus cereus biovar toyoi</i> CNCM 1-1012 (AJ310100)	100
LR 3-6	KP407107	γ-Proteobacteria	<i>Pseudomonas fuscovaginae</i> ICMP 5940 (FJ483519)	99
LR 3-7	KP407108	Firmicutes	<i>Bacillus cereus</i> ATCC 14579 (AE016877)	100
LR 3-8	KP407109	Firmicutes	<i>Bacillus aerophilus</i> 28K (AJ831844)	100
LR 3-9	KP407110	Firmicutes	<i>Bacillus deontensis</i> LMG 21831 (AJ542596)	99
LR 3-10	KP407111	γ-Proteobacteria	<i>Pseudomonas frederiksbergensis</i> JAJ28 (AJ249382)	99
LR 3-11	KP407112	α-Proteobacteria	<i>Rhizobium grahamii</i> CCCE 502 (JF424608)	99
LS 1-1	KP407113	γ-Proteobacteria	<i>Pseudomonas graminis</i> DSM 11363 (Y11150)	100
LS 1-2	KP407114	Firmicutes	<i>Bacillus aryabhatai</i> BSW22 (EF114313)	100
LS 1-4	KP407115	Firmicutes	<i>Bacillus cereus biovar toyoi</i> CNCM 1-1012 (AJ310100)	100
LS 1-5	KP407116	γ-Proteobacteria	<i>Pseudomonas graminis</i> DSM 11363 (Y11150)	99
LS 2-1	KP407117	γ-Proteobacteria	<i>Pseudomonas congelans</i> DSM 14939 (AJ492828)	100
LS 2-2	KP407118	γ-Proteobacteria	<i>Pseudomonas graminis</i> DSM 11363 (Y11150)	100
LS 2-3	KP407119	γ-Proteobacteria	<i>Pseudomonas latea</i> OK2 (AY364537)	100
LS 3-1	KP407120	Firmicutes	<i>Bacillus cereus biovar toyoi</i> CNCM 1-1012 (AJ310100)	99
LS 3-2	KP407121	γ-Proteobacteria	<i>Pseudomonas latea</i> OK2 (AY364537)	100
LS 3-5	KP407122	γ-Proteobacteria	<i>Pseudomonas koreensis</i> Ps 9-14 (AF468452)	98
LS 3-8	KP407123	γ-Proteobacteria	<i>Xanthomonas vesicatoria</i> ATCC 35937 (Y10761)	100

>56 Total culturable endophytic bacteria isolated;

>38 different bacterial strains;

HIGHER IAA levels
+
ANTIFUNGAL ACTIVITY
Paenibacillus kribbensis (LR 2-11)

INOCULATION

> Good plant establishment and development after bacteria inoculation

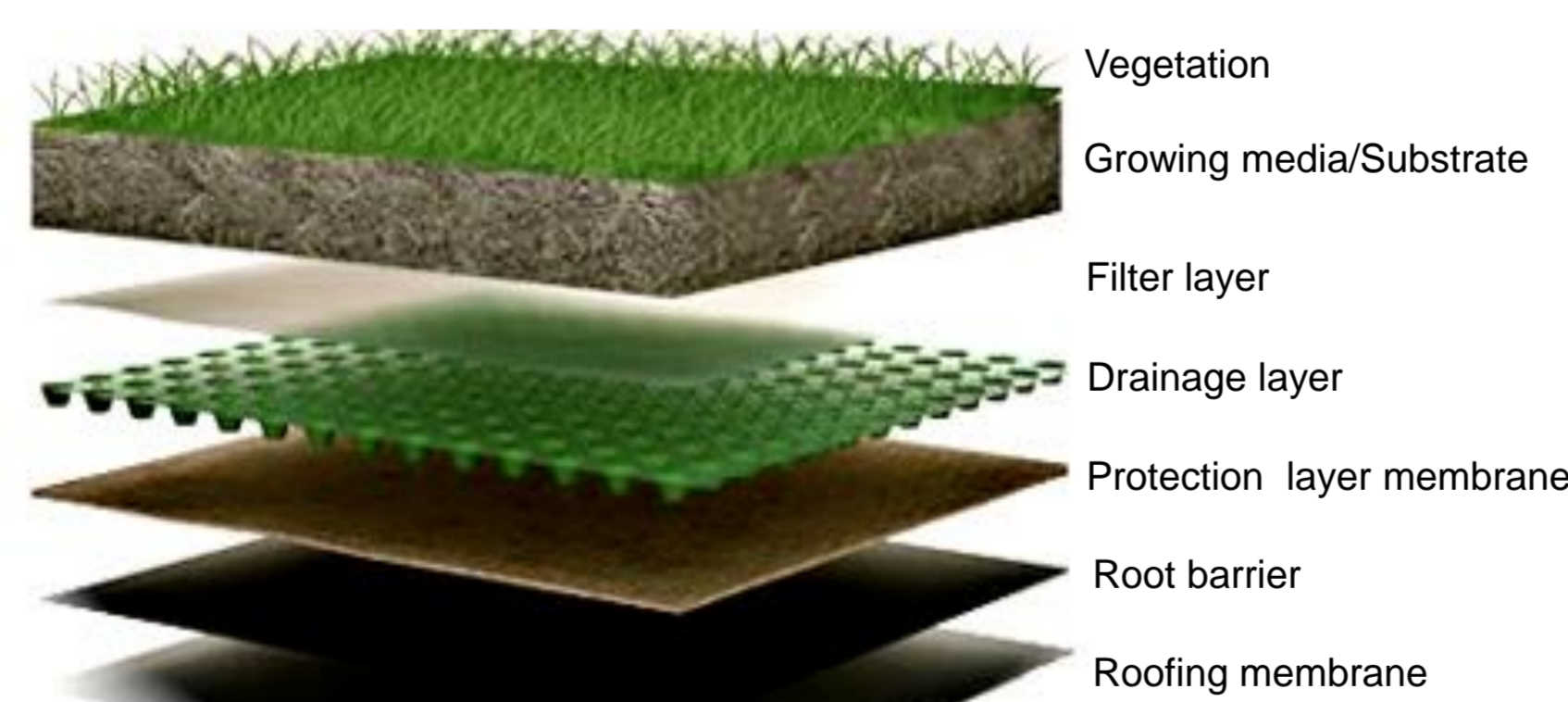


Figure 1: Green roof scheme (<http://godfreyroofing.com>)

Table 2 - Characterization for multiple plant growth promoting (PGP) traits

Strain	Closest relatives	NI ₂	Siderophores	P solubilization	IAA (mg l ⁻¹)	HCN	Extracellular enzymes		
							Cellulase	Pectinase	Protease
LR 1-9	<i>Pseudomonas graminis</i>	+	+	-	51.4 ± 3.58	+	+	+	-
LR 2-11	<i>Paenibacillus kribbensis</i>	-	+	-	7.3 ± 1.43	++	+	++	+
LS 1-2	<i>Bacillus aryabhatai</i>	+	+	+	18.4 ± 1.48	+++	+	++	-
LS 2-1	<i>Pseudomonas congelans</i>	-	+	-	74.7 ± 8.20	++	+	++	+



Figure 2: Green roof pilot systems

Conclusions

Bacterial endophytes colonize differently lavender tissues (root and shoot), some exhibiting interesting growth promoting traits. Aromatic plants presented successful growth after bacterial inoculation during their establishment in the green roof pilot structures. Plants selected proved to be resistant to the harsh conditions of a rooftop and able to establish and grow in the Mediterranean climate.

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