

# Research on Bacterial Endophytes at the Institute for Advanced Learning and Research



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## Background

Bacterial endophytes live inside plants and represent a largely unexplored resource for enhancing sustainable agricultural production. They are also recognized as an untapped reservoir of novel natural products. We have demonstrated significant growth promotion of the bioenergy crop switchgrass by bacterial endophytes (*Burkholderia phytofirmans* strain PsJN and *Pantoea agglomerans* strain PaKM) under *in vitro*, growth chamber, greenhouse and field conditions, particularly in low fertility soil. Currently, we have established a library of bacterial endophytes isolated from plants grown in various environments and characterized their abilities in N fixation, P solubilization and auxin production. Many endophytes exhibited one or several of these plant beneficial traits.

## Results

### Characterization of a library of bacterial endophytes

1. Auxin production: Auxin is a plant growth promoting hormone and plays important roles in plant growth and development. The ability of bacterial endophytes to produce auxin was determined using Salkowski reagent (Fig. 1).

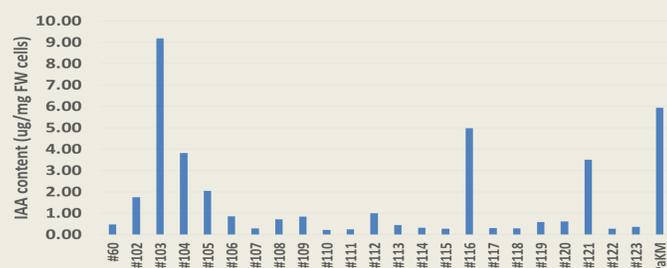


Fig. 1. Auxin levels produced by representative bacterial endophytes.

2. Phosphate solubilization: The ability of bacterial endophytes to solubilize phosphate was observed using Pikovskaya's agar medium containing insoluble calcium phosphate and quantifying with the method of Murphy and Riley (1962) (Fig. 2).

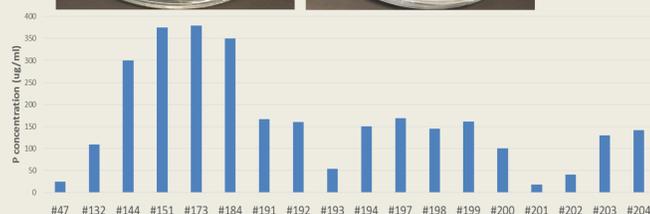
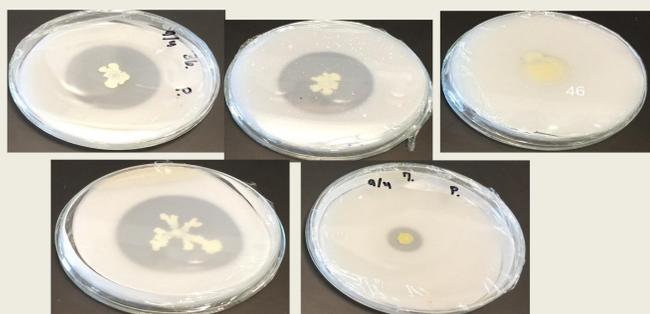


Fig. 2. Phosphate solubilization by representative bacterial endophytes.

3. Nitrogen fixation: The ability of nitrogen fixation by bacterial endophytes was determined by growing bacteria on Norris N free medium with at least 3 subcultures (Fig. 3)

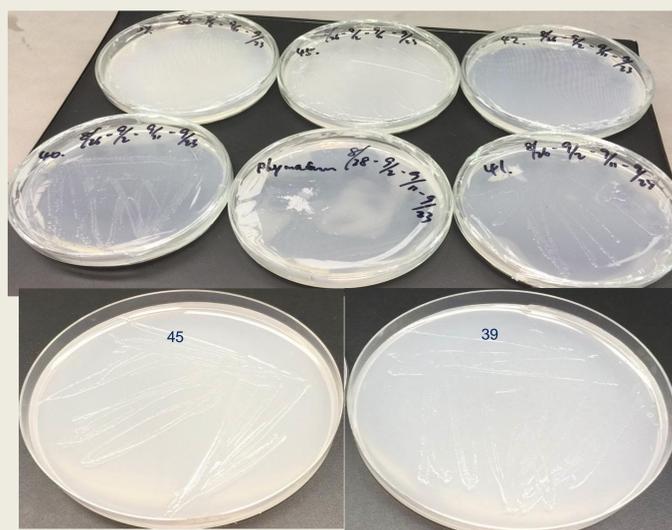


Fig. 3. Representative bacterial endophytes grown in Norris N free medium.

### Plant growth promotion by bacterial endophytes

1. Switchgrass growth promotion: We have worked on *Burkholderia phytofirmans* strain PsJN and *Pantoea agglomerans* strain PaKM for more than 5 years and found that both of them significantly promoted switchgrass growth in various conditions (Fig. 4)

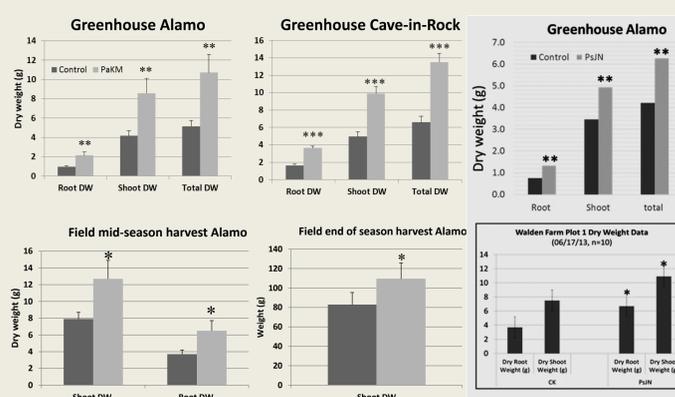


Fig. 4. Switchgrass growth promotion by bacterial endophytes in greenhouse and field.

2. Tall fescue growth promotion: We have tested several tall fescue germplasm lines and found *Pantoea agglomerans* strain PaKM greatly promoted *in vitro* tall fescue growth (Fig. 5)



Fig. 5. Tall fescue growth promotion by *Pantoea agglomerans* strain PaKM *in vitro*.

3. Tobacco growth promotion: We have found that both of PsJN and PaKM significantly promoted tobacco growth (Fig. 6)

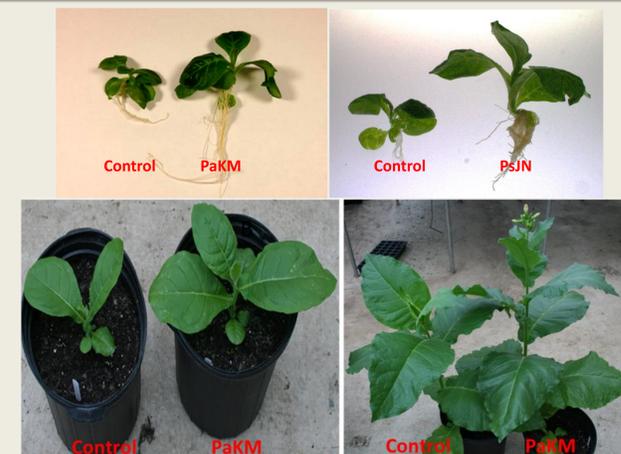


Fig. 6. Bacterial endophytes promoted tobacco plant growth under *in vitro* and greenhouse conditions.

### Phytoremediation with bacterial endophytes

We are involved in Altavista's 6-acre Petri Dish of Polychlorinated biphenyl (PCB) remediation with switchgrass and bacterial endophytes. We isolated bacterial endophytes from plants grown in that contaminated area and found some bacteria could degrade the PCB analog biphenyl (Fig. 7A). We also work on bioremediation of polycyclic aromatic hydrocarbons (PAH) (Fig. 7B, C)

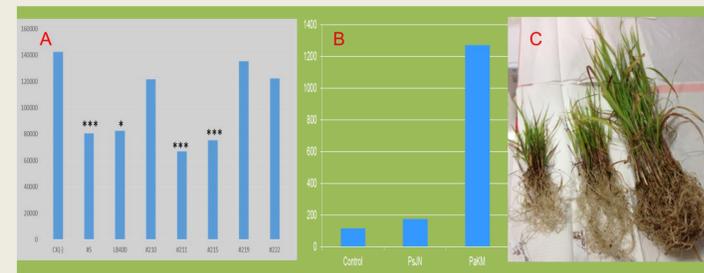


Fig. 7. A: Biphenyl degradation by bacterial endophytes at 5-day culture. B and C: Switchgrass-inoculated with bacteria grew in PAH contaminated soil for two months.

### Drug discovery from bacterial endophytes

Bacterial endophytes were screened from our bacterial library. Four to eight liters of bacterial cultures were prepared for extraction and bioactivity guided fractionation and isolation. Structure characterization, structure modification and total synthesis will be conducted for potential candidates.

