
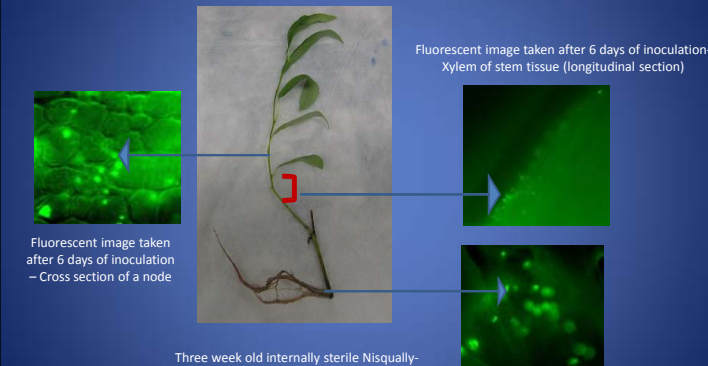


IMPROVING BIOMASS PRODUCTION AND PHYTOREMEDIATION USING NATURAL ENDOSYMBIONTS OF POPLAR AND WILLOW



Sharon L. Doty, Ph.D.
Associate Professor

Endophytes are microbes living within plants without causing disease





Fluorescent image taken after 6 days of inoculation – Xylem of stem tissue (longitudinal section)

Fluorescent image taken after 6 days of inoculation – Cross section of a node



Three week old internally sterile Nisqually-1 plant inoculated with *gfp*-WP9

Fluorescent image taken after 2hrs of inoculation (root emergence)





Endophytes are found in most plants and can be added for improved

- *Stress tolerance
- * Growth with less fertilizer
- *Remediation potential

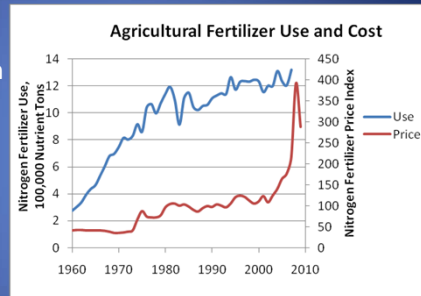



Fixed nitrogen is a limiting nutrient for plant growth



Chemical Fertilizers

- Synthesized nitrogen fertilizer requires high levels of fossil fuel.
- Price is tied to cost of fossil fuels
- Harmful to the environment



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Climate change mitigation

- Increase C sequestration if N is not limiting
- Using more chemical fertilizers is inconsistent with mitigation

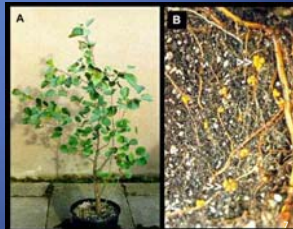


Biological Nitrogen Fixation

Nitrogenase



EXPENSIVE REACTION → Usually plant-associated



NO ROOT NODULES ON THESE PLANTS



In their native habitat, poplar and willow grow under low-nitrogen conditions



Poplar Endophytes Most are putative N-fixers

- *Rhizobium tropici*
- *Burkholderia vietnamiensis*
- *Herbaspirillum*
- *Pseudomonas graminis*
- *Rahnella* sp.
- *Acinetobacter* sp.
- *Enterobacter* sp.
- *Sphingomonas* sp.
- *Rhodotorula* sp.



Doty, S. L., et al. (2005) *Symbiosis* 39: 27-35.

Doty, S. L., et al. (2009) *Symbiosis* 47: 23-33.

Xin, G., et al. (2009) *Mycological Research* 113:973-980



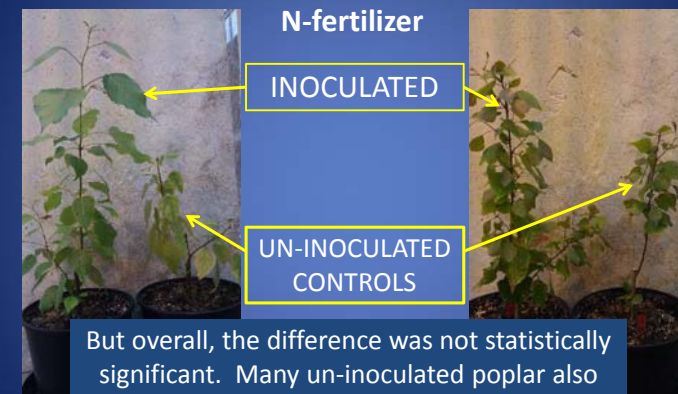
Poplar (Nisq-1) inoculated with endophyte consortia had more than TWICE the root mass as the control

- UNPUBLISHED DATA WAS REMOVED

J Knoth, SH Kim, and SL Doty, manuscript in preparation



Addition of wild poplar endophytes to cultivated hybrid poplar clones improved growth without



But overall, the difference was not statistically significant. Many un-inoculated poplar also had N-fixing endophytes

Baum and Doty, unpublished



Field Trial at Boardman, Oregon Greenwood Resources



Ph.D. graduate student, Amy Baum, inoculated half of the 60 hybrid poplar trees for a field trial of the impact of the endophytes

Many poplar endophytes produce plant hormones (auxins) that increase root growth

UNPUBLISHED DATA WAS REMOVED

Doty, SL, Doty, CM, Khan, Z, and Isebrands, JG, manuscript in preparation



7300501 (*P. deltoides*) at 2 mths



Uninoculated controls

CONSORTIA

Statistically significant difference in total root mass at 3 months

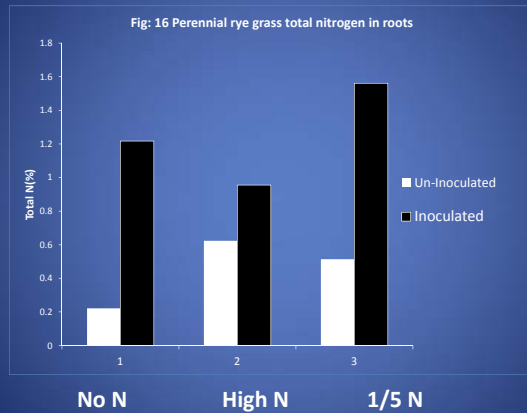
Doty, SL, Doty, CM, Khan, Z, and Isebrands, JG, manuscript in preparation

Inoculating other plants with endophytes from poplar and willow improves plant growth in low-nutrient soils



Khan, Z, Guelich, G., Phan, H., Redman, R., and Doty, S. L. 2012. ISRN Agronomy (in press)¹⁶

Endophytes can increase total N



Khan, Z, Guelich, G., Phan, H., Redman, R., and Doty, S. L. 2012. ISRN Agronomy (in press)

Endophytes can improve plant growth under water stress

60% more root biomass and 48% more shoot biomass



Perennial rye grass after two weeks of water stress. Plants on the left were colonized by an endophyte consortium.

Khan, Z, Guelich, G., Phan, H., Redman, R., and Doty, S. L. 2012. ISRN Agronomy (in press)

When plants are grown on large- scale commercially, their natural symbiotic partners may be lost



Adding the microbial partners back may lessen the reliance on chemical fertilizers, increasing plant growth and stress tolerance



NSF Energy for Sustainability program



AFRI bioenergy program

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Part 2: Phytoremediation Research

PESTICIDES



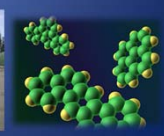
SOLVENTS




EXPLOSIVES



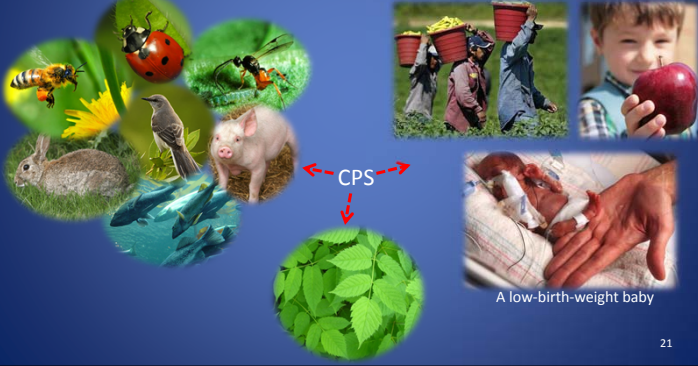
PAH'S



CPS is a broad-spectrum insecticide

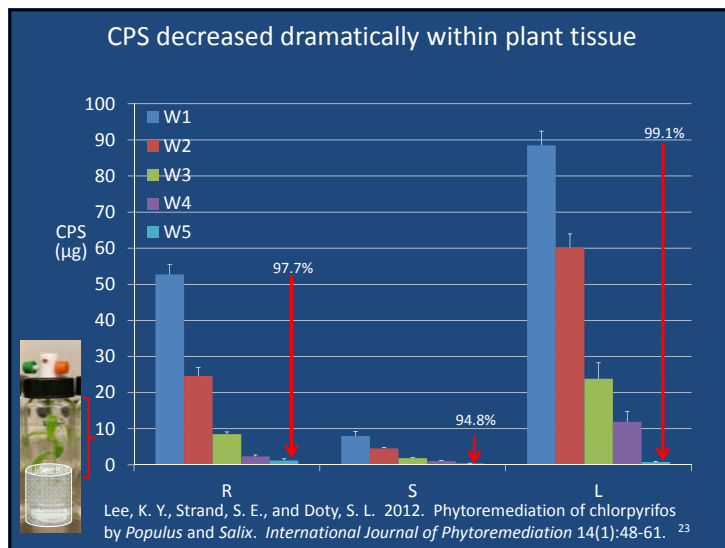
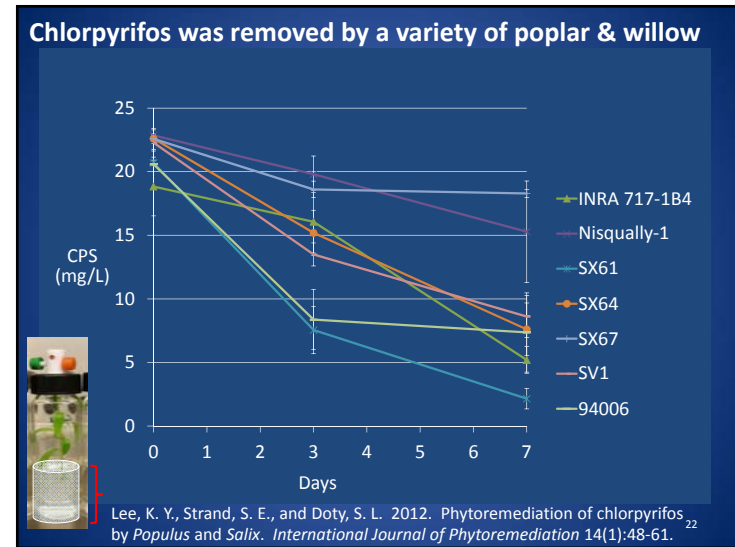


Adverse impact on environmental and human health





A low-birth-weight baby

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Phytoremediation Research



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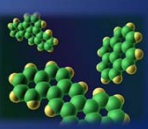
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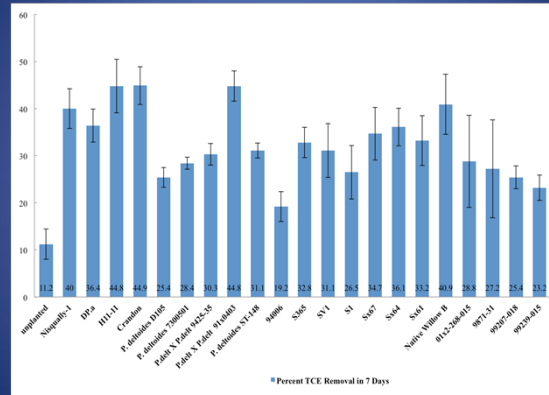
EXPLOSIVES

PAH'S

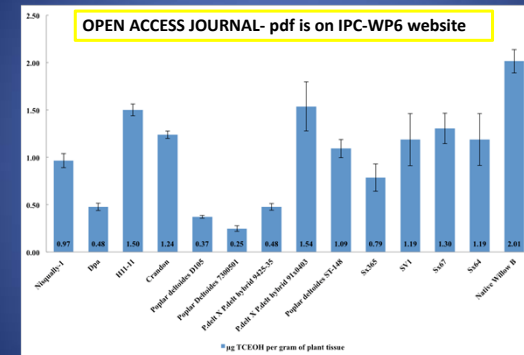



Removal of TCE by a variety of poplar and willow clones

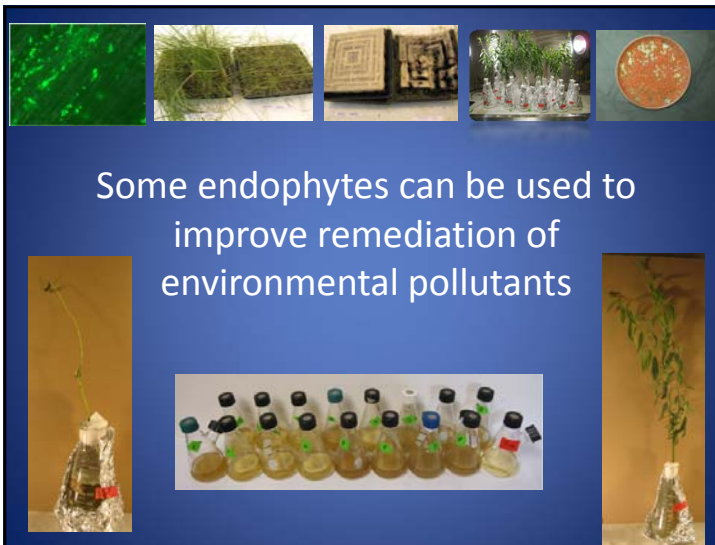


Miller, RM, Khan, Z, and Doty, SL. 2011. *Journal of Bioremediation and Biodegradation* S7:001. Doi:10.4172/2155-6199.S7-001.

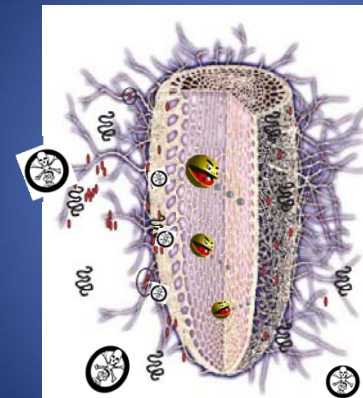
Degradation of TCE by a variety of poplar and willow clones varies



Miller, RM, Khan, Z, and Doty, SL. 2011. *Journal of Bioremediation and Biodegradation* S7:001. Doi:10.4172/2155-6199.S7-001.




Endophyte-Assisted Phytoremediation




Can take advantage of the natural ability of microbes to rapidly adapt to pollutants and to colonize plants that can take up the pollutant


Picture from Jun Won Kang



Improving phytoremediation using endophytes in partnership with plants for more effective phytoremediation of the pollutants, TCE and PAHs



 National Science Foundation
WHERE DISCOVERIES BEGIN

NSF Environmental Engineering Program


 USDA

McIntire-Stennis Program

1. Isolation of endophytes

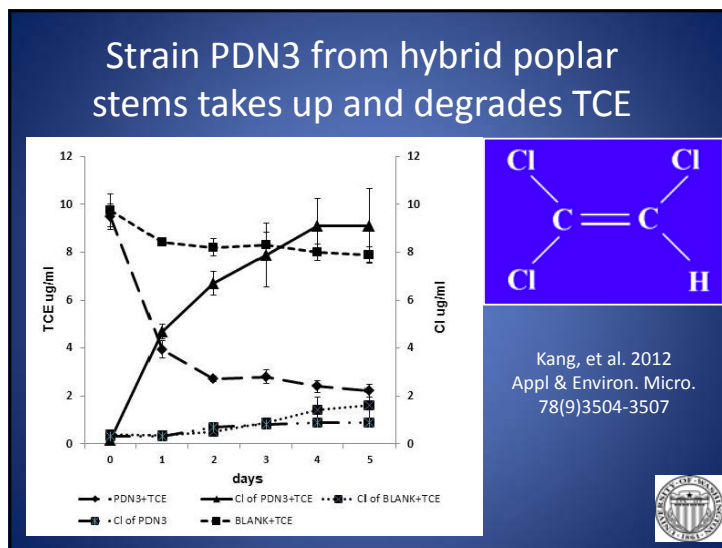



2. Screen for resistance to high levels of TCE




Kang, et al. 2012. Appl & Environ. Micro. 78(9)3504-3507

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Phytoremediation Research


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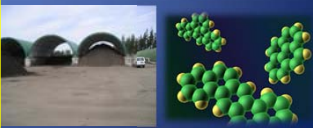
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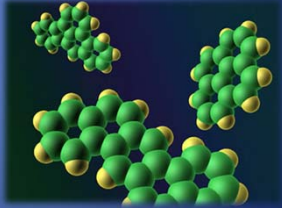
PAH'S



PAHs

- Polycyclic aromatic hydrocarbons
- Multiple fused rings
- Formed through the incomplete combustion of carbon compounds: coal, oil, wood, etc
- Phytotoxic



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Endophyte strain, PD1, isolated from poplar stems, consistently performed best on PAHs (pyrene, naphthalene, phenanthrene)

UNPUBLISHED DATA WAS REMOVED

Zareen Khan, David Roman, and Sharon Doty, manuscript in preparation



PD1 reduces PAH phytotoxicity

Day 1

Day 19

Day 1

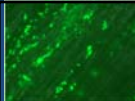




Day 19

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Inoculated with PD1

Uninoculated control

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SUMMARY

Endophytes can improve plant growth, fix nitrogen, and produce phytohormones

A variety of poplar and willow clones can remove the common pesticide, chlorpyrifos. There is variation in the ability of different poplar and willow clones to degrade the most common pollutant found in Superfund sites, TCE

Some endophytes can degrade environmental pollutants including TCE, PAHs, & explosives, and may aid in phytoremediation

Acknowledgements



DOTY LAB
Zareen Khan- Research Scientist III
Regina Redman- RS IV

Graduate Students
Jun Won Kang
Keum Young Lee
Jenny Knoth
Amy Baum

Undergraduate Researchers: N-fixation:
Andy Tiegen, Grant Guelich, Falon
Miranda, Josefina Narvaja, Alex Thomas

Phytoremediation and Bioenergy:
David Roman, Hannah Morrison

Other temp. researchers: Ronald Cuie
and Yoshitaka Goto, Christina Doty

Co-PI's: Soo-Hyung Kim, Greg Ettl, Renata
Bura, and Stuart Strand

NSF-BioE, NSF-EE, AFRI, UW C4C, McIntire-Stennis, NIEHS, CPBR