





## Oil and Gas Endophyte-Enhanced Tree Phytoremediation plus Mycoremediation

Crude oil and gas pollution originate from many sources and can include activities such as unintentional spills of organic pollutants, leaking storage tanks, and oil and gas exploration, extraction, and transportation that can contaminate soils and sediments, groundwater, and surface water. Traditional cleanup of petroleum hydrocarbon pollutants is costly, not only financially but also environmentally. In some cases, traditional remediation treatments are unsuccessful at removing a sizable portion of pollutants that were accidentally released into the environment. In other cases, low, yet reportable, levels of recalcitrant petroleum hydrocarbon pollutants persist after initial cleanup, making it difficult to close sites. Our team provides remediation assessment, direction and new solutions to assist in the stabilization and remediation of these contaminants from polluted environments, and the mitigation of risks associated with these sites. Intrinsyx and PPCU utilize poplar tree-endophytic bacteria in many different groundwater loving trees combined with the Remediators fungal soil mycelium to effectively remove and degrade petroleum hydrocarbon pollutants (BTEX, TPH, PAH's, etc.) in groundwater and soil using a combined poplar tree phytoremediation and mycoremediation system.

## Advantages of using plants inoculated with endophytic bacteria that degrade petroleum

hydrocarbons. Trees inoculated with our highlyspecialized bacteria significantly increase the degradation of petroleum pollutants in soil and water by as much as 40% versus controls containing un-inoculated plants, and considerably more than no treatment at all. In addition, plants containing these specialized endophytic bacteria have demonstrated higher root and shoot growth as well as no signs of phytotoxic effects from pollutants, even at traditionally petroleum these phytotoxic concentrations. In fact,



endophytic bacteria even facilitate increased uptake of pollutants into the plant tissues for degradation, which is especially important for recalcitrant hydrocarbons. In the image shown above, willow trees were inoculated (left 3 trees), or un-inoculated (right 3 trees), and grown in soil containing phytotoxic concentrations of phenanthrene (Khan et al. 2014). Thankfully, these specialized endophytic bacteria can be used with any plant species, and the inoculation of plants can occur at the time of planting or on established trees, shrubs, herbs and grasses!

Plant endophytic bacteria that degrade chlorinated solvents and pesticides. Some petroleum hydrocarbon impacted sites also contain chlorinated solvents or persistent organic pesticides, or even explosives! In addition to our petroleum hydrocarbon degrading poplar endophytes we have tree endophytes that degrade chlorinated molecules and explosives like TNT and RDX.







Soil mycoremediation. Fungi naturally breakdown organic compounds from the soil in which they reside. They inherently degrade a variety of petroleum hydrocarbons including aromatic (PAHs, dioxins) and chlorinated (PCBs, DDT) compounds. Degradation of these organic pollutants results in the creation of water and carbon dioxide, leaving no contaminants behind. In addition to the degradation activities of these beneficial fungi for remediation of organic pollutants, they also provide benefits to the plants used for phytoremediation by helping to make mineral nutrients more bioavailable as well as confer greater environmental stress tolerance to biotic and abiotic factors.

Combined Tree Bio-Phytoremediation and Mycoremediation Applications. Combining endophyte-enhanced phytoremediation with mycoremediation has the potential to dramatically increase the remediation efficiency and effectiveness of organic polluted sites over any other green technology on the market; and, this system is vastly less expensive than traditional remediation approaches. Our system is designed to work together to increase remediation efficiency from the time of implementation to closure and reduces the total time to remediate using biological organisms. To top it all off, the technologies discussed in this paper are isolated from nature and are completely safe to humans and the environment, and do not require specialized permitting for use. Many sites we encounter are contaminated with multiple pollutants and we have found that this multifaceted approach is ideal because we can address multiple contaminants of concern concomitantly.

Our endophytic plant bacteria and soil fungi are compatible with most plant species. That means we can customize our remediation approach specific to the site's geographic region, site conditions, chemical characteristics, and depth of pollutant(s). Plant selection can take into consideration any desire for native plants as well as future plant biomass use for timber or bio-fuel related applications. This combined system allows us to address multiple pollutants at many depths. We can address:

- Soil contamination at shallow depths and deeper due to the trees
- Groundwater contamination at 30 feet below ground using high-transpiration water loving trees. Trees like Poplar, Willow, Ash and Alder are quite useful in this regard. These trees generally grow in freshwater aquifers where the water table depth is not more than ten meters.
- Aquatic systems requiring water and/or sediment remediation

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