Phytoremediation of Petroleum-Contaminated Soils in the Tropics

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PDVSA INTEVEP

Phytoremediation is a non-destructive and economic *in situ* technology that uses plants to remove, degrade or stabilize contaminants in soil. In case of oil contamination, it is based on the stimulation of microbial degradation in the rhizosphere. Although phytoremediation is especially promising for the tropics due to climatic conditions that favour plant growth and microbial activity, research was so far mostly limited to the temperate zone. Furthermore, factors controlling the process and success of phytoremediation are still not well understood.

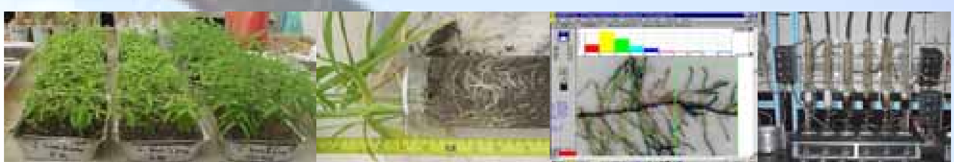
The present project was a cooperation of the University of Hohenheim and PDVSA-Intevep (Centro de Investigación y Apoyo Tecnológico de Petróleos de Venezuela S.A.). The studies were carried out with plants and soil from the savannah of eastern Venezuela. Results are expected to assist in the development and application of phytoremediation not only in Venezuela but also in other tropical countries.

After the pre-selection of plants collected on crude oil contaminated sites, species with characteristics promising for phytoremediation were screened in a greenhouse experiment for their ability to increase the degradation of petroleum hydrocarbons in soil. Soil planted with the pasture grass *Brachiaria brizantha* showed a significantly lower oil concentration after 180 days than unplanted soil. In subsequent expanded experiments with *B. brizantha*, fertilizer levels were adjusted to obtain best plant growth and highest oil dissipation.

Microbiological studies of rhizosphere and non-rhizosphere soil showed that *B. brizantha* had a prevalently increasing effect on microbial numbers, especially fungi. Since they tolerate lower pH values than bacteria, fungi are considered to play a central role in oil degradation, especially in acid savannah soils. Analysis of carbon source utilization patterns showed different microbial community structures in rhizosphere vs. non-rhizosphere soils. In particular, D,L-a-glycerol phosphate was more used in the rhizosphere, pointing to a higher availability of phosphorus, which is essential during oil degradation but scarce in savannah soils.

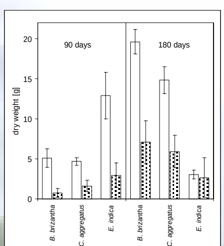
Pre-Selection/ Screening

Criteria for the pre-selection of potential phytoremediation species were population characteristics (frequency, density and importance score of a species at contaminated sites), ease of propagation (availability and handling of seeds, seed germination in contaminated soil, vegetative propagation), and root structure.



Subsequent screening experiments conducted in the greenhouse identified species that stimulate the degradation of the heavy crude oil Boscán in soil.

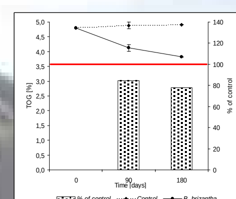
Phytoremediation Species

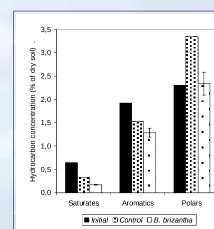


Although growth of *B. brizantha* was inhibited by the presence of heavy crude oil, it showed high biomass production of shoot and root compared to other tested species.

Left: Root biomass production of three graminoids. White bars: plants grown in uncontaminated soil. Grey bars: plants grown in soil with 5% crude oil. Values are means ± standard deviation.

B. brizantha caused highest dissipation of heavy crude oil in soil. Differences in oil contents in planted and unplanted soil could be determined for total oil and grease as well as for single fractions (saturates and aromatics).

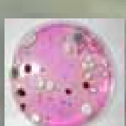




Top: Content of saturates, aromatics and polars in planted and unplanted soil (control).

Left: Total oil and grease (TOG) in planted and unplanted soil (control).

Phytoremediation Mechanisms

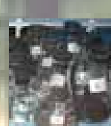


Microbial enumeration

The presence of *B. brizantha* had an initially inhibiting effect on bacteria but a promoting effect on fungi and oil degraders.

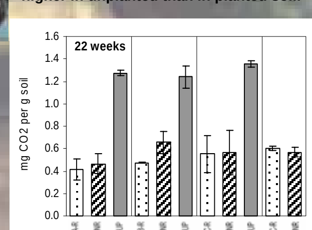
Source	14 weeks df	Mean squares		22 weeks df	Mean squares		30 weeks df	Mean squares	
Bacteria									
ZONE	2	4.3E-01	**	2	1.6E-01		2	8.6E-02	
FERT	2	9.5E-02		2	1.7E-01	*	1	4.9E-01	**
ZONE x FERT	4	6.1E-02		4	6.2E-02		2	3.0E-01	**
Error	14	5.8E-02		14	3.4E-02		10	3.8E-02	
Fungi									
ZONE	2	6.9E-02		2	3.0E-01	***	2	7.7E-01	***
FERT	2	1.3E-01	*	2	6.9E-01	***	1	7.3E-02	
ZONE x FERT	4	9.0E-03		4	1.4E-01	**	2	3.2E-02	
Error	12	1.9E-02		14	1.8E-02		10	2.6E-02	

Analyses of variance of a 3x3 factorial design: 3 sampling zones (ZONE: rhizosphere, non-rhizosphere and unplanted soil) by 3 fertiliser levels (FERT: A, B and C).

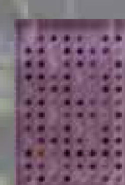


Microbial respiration

Substrate induced soil respiration was higher in unplanted than in planted soil.

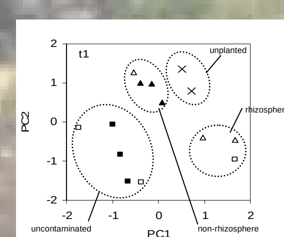


Soil respiration in contaminated soil with three fertiliser levels (A, B, C) and in uncontaminated soil (UC). Rhizosphere soil (R); non-rhizosphere soil (NR); unplanted soil (UP). Values are means ± standard deviation.



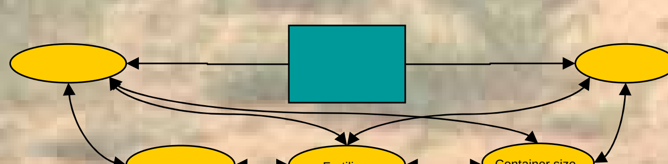
Microbial community structure

Analyses of substrate utilization patterns showed a division of bacterial communities after 14 weeks but not after 22 weeks and thereafter.



Principal component (PC) analysis of substrate utilization patterns.

None of the microbial parameters could fully explain the phytoremediation effect of *B. brizantha*. Fungi seem to play a particular role especially considering low pH values that occur in planted soil. Other factors stimulating degradation of oil in soil like oxygen supply and degradation by plant/plant enzymes have to be considered. Although greenhouse experiments help to clarify some important issues of phytoremediation factors and mechanisms, field trials are considered indispensable for the investigation of phytoremediation. Future research issues should furthermore include fertiliser composition and the particular role of fungi in phytoremediation of acid savannah soils.



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