

Bioremediation of Oil-Contaminated Brackish Salty Water and its Safe use in the Cultivation of Woody Trees and Economic Plants

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Abstract

Oil-mining companies have to subject waste water to expensive treatment before it can be discharged on land or at sea to comply with environment regulations. This study aims at developing an economically valid and applied comprehensive solution that takes advantage of oil-contaminated brackish salty water disposed by the General Petroleum Company in Egypt, and maximizes its economic value and ensures its safe use in the environment. Three fields in Ras Sudr site of the company were inspected. Two main common plant species to Ras Sudr, *Tamarix nilotica* tree and *Phragmites australis* grass that is tolerant to salinity. These plants together with their associated bacteria of endophytes and rhizosphere that utilize crude oil as a carbon and energy source was considered a useful combination of bioremediation agents. Initially, soil characteristics were determined by analyzing soil samples taken at depths of 25cm and 50cm, and bacterial content of soil around the roots and within plant tissues was examined. Discharged water (@50 m³ day⁻¹) was used in irrigating plant fields in amounts sufficient to plant needs only. Growth parameters of plants were assessed four times in an interval of two months. Preliminary results indicated that growth rates in plant length, number of branches and stem girth, and chlorophyll content of oil-polluted water irrigated plants of the two plant species were not significantly different ($p \leq 0.05$) of plants irrigated with fresh water. The number of bacteria in the soil increase significantly ($p \geq 0.05$) over time, and the color of residual oil in the soil was fading, indicating its decomposition. Soil under *Tamarix nilotica* contained similar quantities of microorganisms in both coastal saline-alkali soil and inland arid region indicating that colonization of the plant provided stable growth conditions for microorganisms. These plants and endophytes and rhizosphere combination played the main role in the in-situ bioremediation process, and were efficient in removing around 70 % of the initial traces of crude oil within two months. They also provide safe environment and promote plant growth. They were able to decompose hydrocarbons and residues of crude oil as they possess special physiological mechanisms (PGPR) turns polluted water to safe water for human and environment, and meanwhile achieving the objectives of this work. These results indicated that *Tamarix nilotica* and *Phragmites australis* are promising agents for treating oil-polluted salty wastewater in other fields of crude oil mining.

Keywords: bioremediation, endophytes, oil contamination, *Phragmites australis*, rhizosphere, *Tamarix nilotica*

Introduction

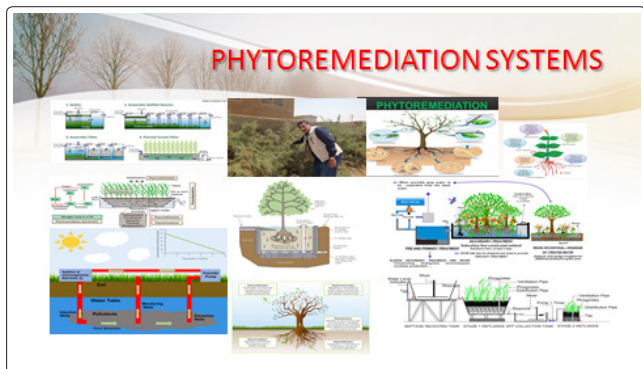
Crude oil is extracted with a quantity of brackish water. The crude oil is washed once it is stored in large tanks in additional quantities of water from the sea. This salty water is separated from crude oil by physical separation methods (separation of oil from water). At the end of the separation process, the crude oil is transported to the shipping ships and the salt water remains with the remains of oil. The problem lies in the fact that this water is in violation of the specifications of the environmental requirements in Egypt and globally in terms of high salinity and mixing with some crude oil. It must undergo expensive purification processes to conform to environmental requirements that allow it to be discharged on land or at sea.

This applied study aims to find a scientific solution achieves Economical:

1. Benefit from this water and to maximize its economic value.
2. Protect the environment from pollution and application of environmental requirements.
3. Facilitate the work of the oil companies and the continuity of safe production and prevent conflict between them and environmental protection institutions.

In the framework of scientific research published locally and internationally in the scientific journals approved in the field of (Phytoremediation) and based on the long experience of the researcher in this area, the proposed economic solution depends on the plants development of specific plants, that are resistant to high salinity and able to get rid of the remnants of crude oil, (Microorganisms) that feed on these hydrocarbon compounds and then

benefit from these plants economically and achieve the rest of the desired objectives of the research.



Eman A. Diab and Reham K.A. Badry (2011), Abstracted that, during a scientific visit to a coastal area at Suez, Egypt, it was observed that *Tamarix nilotica* plant naturally dominated on oil polluted site in this area, indicating that this plant is a tolerant of the combined adverse effects of salinity and petroleum pollutants. This observation stimulated a study to investigate the rhizosphere effect of this plant on the degradation and removal of petroleum aromatic hydrocarbons (PAH) compounds from this coastal saline soil. Accordingly, samples were collected from the rhizosphere and from the non-rhizosphere soil and studied. The results show that the rhizosphere soil of *Tamarix nilotica* was rich in total heterotrophic bacteria and oil-degraders. In the rhizosphere soil oil-degraders were of higher percentage (30.7%) compared to the non-rhizosphere soil (4.6%). Residual total petroleum hydrocarbons (TPH) in the non-rhizosphere soil were 2.25% (w/w), while in the rhizosphere soil the percentage was 0.9% (w/w).



Tamarix seed germination

This indicates a reduction of 60% of the TPHs. The saturates fraction in the rhizosphere as compared to the non-rhizosphere soil was reduced by 87.5%, while the aromatics were reduced by 60.7%. It is of interest to find that the non-degradable asphaltenes and resins were reduced in the rhizosphere by 1.1% and 2.5% respectively. As a total the amount of PAHs (mgkg⁻¹ soil) were 1073.5 and 541.94 in the non-rhizosphere and rhizosphere soil respectively, i.e. with a loss of 49.5% in the rhizosphere. Chrysene and dibenzo(ah) anthracene as compared to the other PAHs were more frequent in the non-rhizosphere soil. These two compounds were reduced by 55.7% and 24.3% respectively in the rhizosphere. As a total the four-ringed PAHs as compared to other PAH groups were highly reduced (60.3%) in the rhizosphere, this was followed by the three-ringed PAH group (52.5%). The five-ringed and the six-ringed groups were weakly reduced (37.8% and 33.8% respectively). The 8 carcinogenic PAH

groups were collectively reduced in the rhizosphere by 49.1%. A particular notable distinction of the rhizosphere of *Tamarix nilotica* is the greater efficiency to degrade the carcinogenic PAH compounds especially flouranthene (75.4%), benzo(a)anthracene (63.4%) and pyrene (60.2%).

Results of Gas Chromatography (GC) analysis for the detection of the accumulated PAHs in the shoot tissue of *Tamarix nilotica* plant growing in the polluted area as compared to that growing in non-polluted area show that the identified peaks in the tissue of both plants were 15 and 14 peaks respectively. The sum of the 15 PAHs was 528 mgkg⁻¹ dried tissue, whereas the sum of the 14 PAHs was 769 mg kg⁻¹ dried soil. This result indicate an accumulation value of 1.46.

The creative idea & bioremediation map

- The creative idea lies in the ability of the research team to select these suitable plants for the problem and conduct research on them and then transfer them to reality in field applied experiments in accordance with all the environmental conditions associated with each site where oil is extracted globally and then arrive at a scientific map at extraction sites and appropriate plants to purify water associated with These sites. (BIOREMEDIATION NATIONAL MAP).

Visit the company's headquarters to view the solution

- During several days, meeting was held in the company and was attended by the team working in the field of environmental protection at the company and offered them several suggestions for the solution and were discussed about the nature of the problem and statistics related to it and agree to visit site Ras Sudr as one of the company's most suitable sites for the first project.

Field visit to Ras Sudr site and our activities

- The visit took place on Thursday, June 30, 2016, and is the starting point for the project. At the site, a geographical and environmental survey was conducted for all three fields on the site and how to collect the water from one area to another until it stabilizes at the terminal at the final basin.
- Monitoring of all naturally occurring natural plants has been conducted on the site, particularly the tow common plants (*Tamarix nilotica*, & *Phragmits australis*).
- On Friday morning, a hearing was held for most of the employees on the site to present our ideas about the project and a scientific dialogue between the two communities resulted in the conviction of allof the first project as an integrated application experience measured and used when repeating work in other sites of the company.



Research on plant purification processes involves several engineering designs for purification work, all of which end up in a large reservoir where water is suitable for biological or human use. These systems are usually applied in wastewater purification for use in agriculture. However, the situation here is very different because we are dealing with natural saline water that has no contaminants in the harmful sense and has not been added any chemicals, pigments, fertilizers, pesticides or heavy elements, but only brackish saline water. A natural substance with known specifications and a small amount of heavy salts.



Old method



Elmeleigy new method

Based on the results of many research, the water will be used directly in the irrigation of the selected plants to deal with salinity and hydrocarbons according to the possession of special physiological mechanisms and microorganisms around the (plant growth promoter rhizobacteria) PGPR.

It is planned that irrigation with sufficient doses only for the needs of the cultivated plants will hardly allow a leakage of water or evaporation.

- The *Tamarix* trees were selected as a basic category for this study because of their environmental presence in the area and their tolerance to the surrounding climate and the nature of the water used and available for planting. The study was also studied in addition to other suitable species such as *Phragmites* and other trees studied. In order to ensure its success in Egypt and its good economic returns to maximize the existing industries.
- The in-situ technology of using endophytes and rhizosphere bacteria for remediation include cost-effective advantages, the benefit to apply in-situ processes without the need for removal or transport of contaminated soil. Rhizosphere bacteria can be used successfully in an in-situ bioremediation process to degrade

contaminants and prevent harm and potentially provide nutrients for the plant. They are associated with the plant's rhizosphere through attachment to plant roots as plants have the ability to secrete nutrients of organic acids, amino acids and sugars through root systems. They partly control the plant's physiology by delivering nutrients, impacting hormone levels, protecting against toxic compounds by degradation and against pathogens endophytes are non-pathogenic bacteria that occur naturally in internal tissues of plants and have evolved to provide a safe environment that can promote plant growth.

- The ongoing work of the project from 30-6-2016
- carry out several field visits to the site and submit a final draft of the project in conjunction with scientific and technical and financial economic study, and to obtain the approval of the official environmental authorities implemented the following:
- First, the water drainage was completely diverted from drainage in the sea to the plantation area for use in plant irrigation (50 cubic meters of water per day).
- Second: Before the start of irrigation with water separated from the oil and partially treated by the oil separation ponds, the soil samples were collected under surface plants and at depths of 25 cm and 50 cm to study and analyze the soil characteristics
- Samples of soil associated with plant's rhizosphere were collected to study the bacteriological content of the soil, around the roots and within plant tissues.
- Third: Samples of Tamarix and Phragmites were collected to study growth status and physiological specifications.
- Fourth: Sampling are repeated for all studies every two months



Results

- First: all plants grow in good condition and increase in growth rate (length - branch -Stems diameter - chlorophyll content).
- Second: the number of bacteria in the soil increases significantly due to regular irrigation.
- Third: In the case of the presence of patches of oil in the soil samples, a decaying its black color compared to the original color is an indication of decomposition by bacterial activity.

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