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Phytoremediation in mangrove sediments impacted by persistent total petroleum hydrocarbons (TPH's) using *Avicennia schaueriana*

Icaro T.A. Moreira [a,*], Olivia M.C. Oliveira [a,b], Jorge A. Triguis [a], Antonio F.S. Queiroz [a], Sergio L.C. Ferreira [a,c,d], Cintia M.S. Martins [a], Ana C.M. Silva [a], Brunno A. Falcão [a]

[a] Núcleo de Estudos Ambientais, Instituto de Geociências, Universidade Federal da Bahia, Campus de Ondina, 40170-290 Salvador-BA, Brasil
[b] Instituto de Geociências, Departamento de Geofísica Aplicada, Universidade Federal da Bahia (UFBA), Campus de Ondina, 40170-290 Salvador-BA, Brasil
[c] Instituto de Química, Universidade Federal da Bahia (UFBA), Campus de Ondina, 40170-290 Salvador-BA, Brasil
[d] Instituto Nacional de Ciência e Tecnologia, INCT, de Energia e Ambiente, 40170-290 Salvador-BA, Brasil

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ABSTRACT

This study evaluated the efficiency of *Avicennia schaueriana* in the implementation of phytoremediation compared with intrinsic bioremediation in mangrove sediments contaminated by total petroleum hydrocarbons (TPHs). The experiment was conducted for 3 months at a pilot scale under conditions similar to a mangrove: the dynamics of the tides were simulated, and physical, chemical, microbiological and biogeochemical parameters were monitored. After the 90 days, it was found that the phytoremediation was more efficient in the degradation of the TPHs compared to bioremediation, reducing the initial concentration of 32.2–4.2 mg/g. *A. schaueriana* was also more efficient in mediating the degradation of different fractions of hydrocarbons, achieving a removal efficiency of 87%. The microbiological results consisted of a higher growth in the model with the plants, demonstrating the phytostimulation ability of the plants. Finally, the experiment showed that phytoremediation is a promising alternative in mangrove impacted by oil.

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1. Introduction

Mangrove swamps are coastal ecosystems of great ecological importance to tropical countries. These environments return biomass and nutrients to the sea and act as ecologically nurseries of marine organisms. Such regions are also relevant to geochemical studies because, in addition to their economic importance and protection against erosion, a large portion of nutrients accumulate in these areas (Alongi, 2002; Lee et al., 2005; Duke et al., 2007; Santos et al., 2011). However, according to the Environmental Sensitivity Index for Coastal Areas published by NOAA (2010), the mangrove habitat is classified as a tropical habitat sensitive to oil spills due to the difficulties of implementing a contingency plan.

The existing remediation techniques in these environments that are impacted by total petroleum hydrocarbons (TPHs) are expensive and limited when applied in situ at the field scale (Atlas, 1982; Ke et al. 2005; Seabra, 2008; Burns et al., 2000; Huang et al., 2004; Yu et al., 2005; Oliveira, 2008; Brito et al., 2009; Moreira et al., 2010a). As toxic compounds, TPHs represent some of the most common and persistent contaminants in the environment and are highly toxic to the mangrove forests that are particularly difficult to protect. Because many of the techniques will be applied in the other sites, subsequently they may cause other secondary impacts (McNicoll and Baweja, 1995; EPA, 2000).

Advances in biotechnology have prompted several researchers to employ phytoremediation, which can be an alternative that is potentially more effective for soils and sediments contaminated with TPHs and more profitable when compared to the traditional remediation methods (Salt et al., 1998; Gunther et al., 1996; Susarla et al., 2002). Phytoremediation is well defined in the literature as a method that uses plants to stabilize, extract, accumulate, degrade or transform contaminants in sediments, soils or aquatic environments. The plants utilized process the phytodegradation, phytostabilization or phytoextraction of the contaminants (Gunther et al., 1996). In the case of sediments contaminated with oil, the effect of phytoremediation is based mainly on rhizosphere microorganisms that stimulate degradation, a process known as rhizodegradation (Alkorta and Garbisu, 2001; Santos et al., 2011).

For maximum success in the implementation of phytoremediation, it is essential to use plants that are endemic to the areas requiring treatment; these plants offer significant advantages because they are adapted to the local environmental conditions and possess established interactions with the microorganisms in the area (Anderson et al., 1993). However, the diversity of plant species

* Corresponding author at: Núcleo de Estudos Ambientais, Instituto de Geociências, Universidade Federal da Bahia, Campus de Ondina, 40170-290 Salvador-BA, Brasil. Tel./fax: +55 71 3283 8632.
E-mail addresses: icarotam@gmail.com, icarotam@ufba.br (I.T.A. Moreira).

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is very low in mangrove ecosystems. It was found that *Avicennia schaueriana* (black mangrove) is a plant that does exhibit sensitivity to the presence of sediments contaminated by oil, suggesting a promising species for such remediation (Moreira et al., 2010a,b). This plant species has a discontinuous distribution and is found mainly in Brazil (90% of the occupied coastal area), at the southern end of the Leeward Islands, the Atlantic Coast of the northern region of South America, South of Guyana and Suriname (Dodd and Afzal-Rafii, 2002; Wilkie and Fortuna, 2003).

The purpose of this study was to evaluate the application of *A. schaueriana* in phytoremediation when compared to intrinsic bioremediation in mangrove sediments contaminated by TPHs in a pilot study using controlled laboratory conditions that closely simulated the environmental conditions of a mangrove ecosystem.

2. Materials and methods

2.1. Sediment preparation and residual oil addition

Surface sediment samples were collected at a depth of 0–30 cm using a stainless steel sampler in a mangrove ecosystem northern Bay of Todos os Santos, between the cities of Candeias and São Francisco do Conde, Bahia, Brazil (Fig. 1). The choice of collection sites was random. These sediment samples were sieved through a 4 mm sieve to remove rocks and plant material and were immediately homogenized to ensure uniformity. It was collected five sub-samples of sediment, which were homogenized and freeze-dried in a cold for 72 h and then sieved through 2 mm mesh for the analysis of the selected sediment physical and chemical properties (Table 1). The organic matter in the sediment was assessed using a modified Mebius method (Nelson and Sommers, 1982). The total *N* was determined by the Kjeldahl distillation/digestion and titration method (Bremner and Mulvaney, 1982), and the available *P* was determined by the Olsen extraction method (Olsen and Dean, 1982). The particle size distribution was determined after the organic matter was removed with 30% H2O2, according to the method of Folk and Ward (1957). After homogenization, the sediment samples were mixed in a 1:10 ratio (weight) with oil residue found in the same area, a relatively light paraffinic oil originating the Recôncavo basin, a region with many petroleum industry activities (extraction, transportation and refining). Then, five replicates of the homogenized sediment samples were collected to analyze the initial concentrations of TPHs (Table 2). Uncontaminated sediment was also collected in a reference area, as described elsewhere (Moreira et al., 2010b), for comparisons of the parameters analyzed in this study.

Table 1
Some selected physicochemical properties of the sediment used in the experiment.

Parameters	Value
Textural class	Sandy mud
Particle-size distribution	
Sand (%)	23.65
Silt (%)	73
Clay (%)	3.25
Organic matter (%)	5.73
Organic carbon (%)	3.32
Total *N* (%)	0.36
Available *P* (mg/L)	1.8

2.2. Simulation of the mangrove ecosystem

The dynamics of a mangrove ecosystem was simulated with a tidal regime in the sediment used for the application of the remediation techniques. This experiment was conducted in a greenhouse

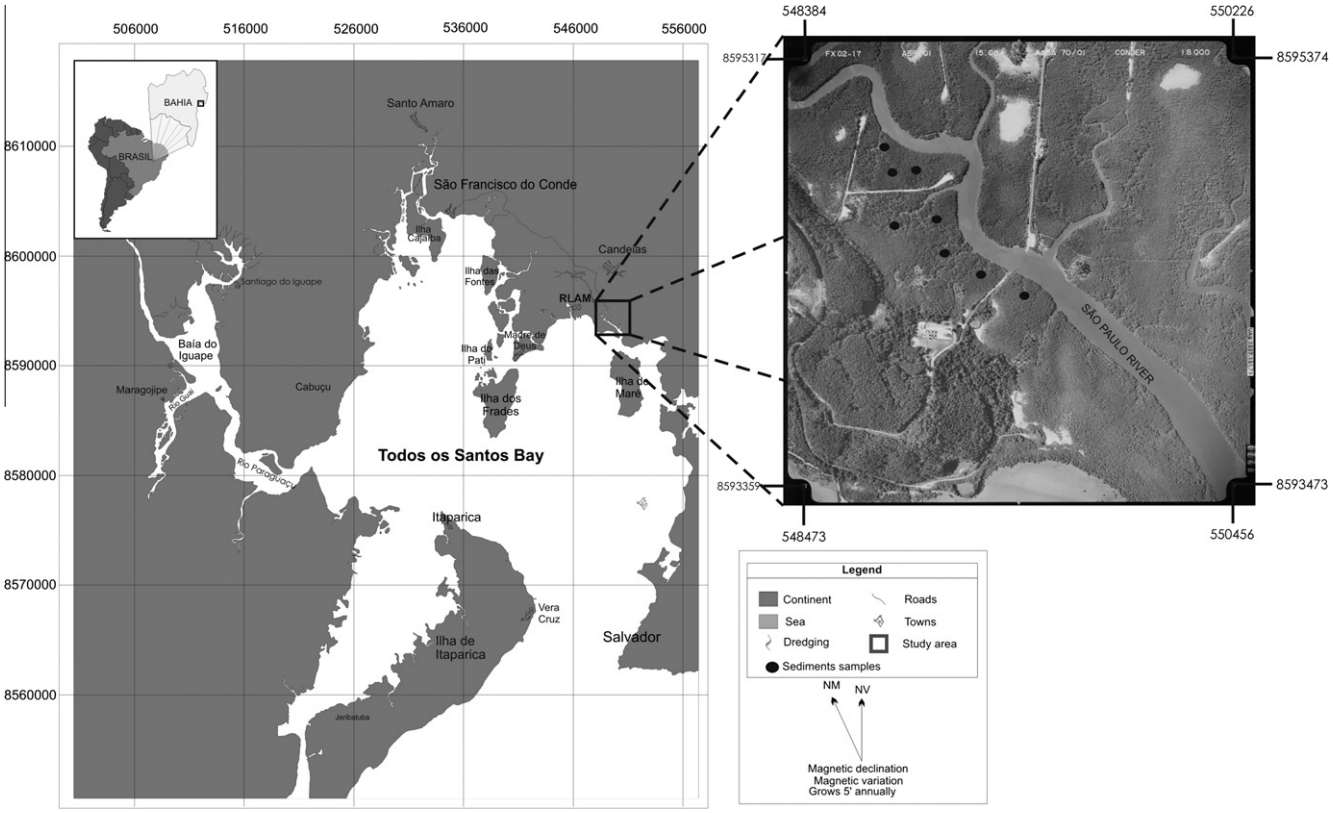

Fig. 1. Map showing the study area.

Table 2
Concentrations average of TPH's (mg/kg^{-1}) and standard deviation (±) in the different treatments (bioremediation and phytoremediation with *A. schaueriana*) during the experiments.

Treatments	Time, day	TPH's (mg/kg^{-1}) average	Standard deviation
Bioremediation	1	33215.2	±2594.1
Phytoremediation	1	33215.2	±2594.1
Bioremediation	7	24351.3	±4363.1
Phytoremediation	7	29494.3	±4035.9
Bioremediation	15	20098.7	±7261.2
Phytoremediation	15	25253.3	±2747.4
Bioremediation	30	19437.7	±2110.4
Phytoremediation	30	18396.8	±4072.0
Bioremediation	60	18698.5	±3888.0
Phytoremediation	60	16041.2	±2031.3
Bioremediation	90	9225.1	±1616.7
Phytoremediation	90	4223.0	±826.05

(a site to conduct research developed within the network RECUPETRO/UFBA – Cooperative Network Recovery in Areas Contaminated by Petroleum Activities, linked to the Federal University of Bahia) near the area where the sediment samples were collected. The conditions approximated the original ecosystem, with an average temperature of 24.6 °C. For this, simulation units were constructed of glass (50 × 30 × 40 cm). Within each unit, 6 tubes of glass (30 × 10 × 10 cm) were applied to the two models of remediation compared in this study. These glass tubes were suspended in the simulation unit, allowing the simulation of a tidal regime, with water runoff. The tubes were closed at the bottom to prevent the loss of chemical residue when adding water. As in a mangrove ecosystem, all of the units received the treatment simulation of a daily tidal regime of an adequate amount of water (approximately 10 L) to maintain a constant humidity of the sediment. The experimental blocks consisted of three replicates of each treatment and the analysis of three samples from each repetition. To assess the efficiency of the phytoremediation of the TPHs in the sediment, each of the remediation models described below were tested separately, taking into consideration public safety and local environmental compartments.

2.3. Application of remediation models

Two models were used to evaluate the remediation of the contaminated sediment by assessing the removal of TPHs. The species *A. schaueriana* (black mangrove) was selected to evaluate the efficiency of phytoremediation in the mangrove sediment, a choice that was based on preliminary tests conducted earlier by our group and on other studies suggesting the use of this plant for phytoremediation (Eysink, 1997; Moreira et al., 2010a,b). Seedlings of *A. schaueriana* were collected at low tide, taking into consideration their height (average of 3 months old), defining a standard sampling to standardize the plant samples collected. The plants were subjected to sediments mixed with waste oil from the study area. In the laboratory simulation, the species were planted in the glass tubes, and morphophysiological monitoring was conducted for 90 days. During the growth period, the plants were watered twice a week with bottled water as needed. The other type of remediation for the removal of TPHs was intrinsic bioremediation (natural attenuation monitored) – in which the degradation of the hydrocarbons by bacteria present in the sediment was monitored. The density of the bacterial community was characterized to compare the presence of microorganisms in the phytoremediation experiment.

2.4. Sediment extraction and analysis of TPHs

The TPH levels in the sediment were determined by assaying for total hydrocarbons. The extraction of analytes was performed using a soxhlet equipment, according to the method USEPA 8015. Sediment samples (approximately 50 g) from the remediation models were collected at 0, 7, 15, 30, 60 and 90 days after the start of the experiments and were stored at 4 °C until analysis. The storage time for the collected samples was no longer than 10 days, and the storage had no effect on the TPH levels in the soil (data not shown), according to the preservation protocol sediment samples (ASTM D3694-2011). The sediment samples were dried in a lyophilizer at a constant temperature of −50 °C. The dried sediment (5 g), without previous treatment, was extracted with dichloromethane/hexane (1:1, v/v). The extracts were concentrated to allow the solvent to evaporate completely, and then the amount of extracted sludge was determined gravimetrically. The extracted oil was weighed to approximately 0.02 g for the aliphatic fraction using an activated silica gel column and eluted with ultrapure hexane (30 mL). The eluate was evaporated, and the volume was adjusted to 1 mL with the same solvent. The extracts were quantified using a Varian CP 3800 (Varian Inc., CA, USA) gas chromatograph equipped with a DB-5 capillary column (15 m length, 0.25 mm ID, 0.25 μm film thickness) and flame ionization detector (FID). The GC conditions were as follows: injector temperature, 300 °C; starting oven temperature, 40 °C; 40 °C (hold 2 min) ramp of 10 °C min^{-1} to 300 (hold 12 min); detector temperature, 300 °C. Helium was used as the carrier gas at a flow rate of 1.0 ml min^{-1}, and a split ratio of 10:1 was used. A calibration standard was prepared from the same TPH (C10–C40) stock chemicals (ASTM D2887 and D6352). The concentration values of the target compounds always fell within the 95% confidence interval of the assigned reference value for concentrations of selected hydrocarbons. The precision of the measurements obtained through replicates of the reference materials was better than 10% for all target compounds.

2.5. Bacterial density in the two models

The quantification of the bacterial density was assessed in the two remediation models studied. For the microbiological analysis, 25 g of the different sediments samples were transferred to Erlenmeyer flasks containing 90 mL of sterile 0.1% peptone water. Each sample was stirred at 200 rpm for 30 min. For the colony counting, the "microgota" (Romeiro, 2001) plating technique was used with decimal serial dilutions on nutrient agar (NA), as follows (in g/L): beef extract, 3; bacteriological peptone, 5; NaCl, 3 and agar, 13. The plates were incubated at 25 °C ± 1 °C for 24 h. After incubation, the plates containing between 3 and 30 colonies were selected, and the number of colonies counted was multiplied by the reciprocal of the dilution. The results are expressed as colony-forming units (CFUs).

2.6. Statistical analysis

An analysis of variance was used to verify the existence of significant differences between the two remediation models. To test the homogeneity of variances, the Bartlett test was used when the parametric analysis of variance did not show a significant difference. The Kolmogorov–Smirnov test was applied to assess the normality of the data; by chi-square, this test indicated that there was no significant difference between the variances of the samples. When the variances were homogeneous, ANOVA was applied to a single parametric classification, which showed significant differences between the two models. An 'a posteriori' Tukey–Kramer test for multiple parameters was applied to confirm significant differences between the models. These statistical analyses were performed using the GraphPad software.

3. Results

3.1. Removal of TPHs in the remediation models

With the objective of evaluating the removal of TPHs from contaminated mangrove sediments in the remediation models employed in this study (intrinsic bioremediation and phytoremediation), a pilot experiment was conducted to compare the different methods of correction. After 90 days, the results showed that the intrinsic bioremediation (natural attenuation monitored) was able to remove 70% of individual TPHs, whereas *A. schaueriana* (phytoremediation) removed approximately 89%, indicating a statistically significant TPH removal by *A. schaueriana*. These results indicate that the phytoremediation with *A. schaueriana* has a greater capacity to remove TPHs from mangrove sediments. The phytoremediation using *A. schaueriana* was able to remove approximately 19% more TPHs from the sediment than intrinsic bioremediation, with contaminants levels being reduced from 33.2 to 4.2 mg/g, whereas the intrinsic bioremediation only lowered the levels to 9.2 mg/g over 3 months (Table 2).

3.2. Removal of different fractions of TPHs in the remediation models

According to Huang et al. (2005), fractions 3A (C16–23), 3B (C23–34) and 4 (C34–40) are the most recalcitrant TPH contaminants in sediment. Our results indicate that the phytoremediation with *A. schaueriana* was more effective than the intrinsic bioremediation in the removal of all of the fractions of the TPHs. However, the efficiencies of both remediation models were similar for fraction 3A (C16–C23) (81% and 77%, respectively). For fraction 3B (C23–34), the results showed that the degradation efficiency of phytoremediation was moderately higher (78%) than that of intrinsic bioremediation (63%), whereas this difference in effectiveness was much greater for fraction C24–C40 (phytoremediation: intrinsic bioremediation, 61%: 21%). Taking into account the levels of total TPH remediation, after 90 days, the phytoremediation with *A. schaueriana* showed decreases in the major components of fractions 3A, 3B and 4, with an efficiency of approximately 73%; the intrinsic bioremediation showed decreases of only approximately 55% (Fig. 2).

3.3. Temporal analysis of the remediation models for the removal of TPHs

The temporal degradation of the two models of remediation was evaluated based on the total content of TPHs in the mangrove sediment (Fig. 3). The removal rate remained relatively constant for phytoremediation, resulting in pseudo-zero-order kinetics for the entire period of 90 days. This behavior of the phytoremediation became a more effective model for remediation than the intrinsic bioremediation, which, despite having received a higher rate at the beginning of the experiment, failed to maintain these initial rates of recovery during the experiment. After 3 months, the total amount of TPHs removed by *A. schaueriana* was approximately 89%, whereas it was approximately 70% for bioremediation, showing a strong decrease in the rate of removal.

3.4. Bacterial communities in the two models of remediation

During the 90 days of the experiment, the total numbers of viable bacteria in the two remediation models were quantified in six pre-established samples. The initial average counts were between 0.1 and $0.2 \times 10^6 \times 10^6$ CFU g^{-1}, and there was a significant increase in the number of microorganisms after the 7th day in the two models, showing significant differences compared to the initial sediment sample of 8.3×10^6 (intrinsic bioremediation) and 1.5×10^6 (phytoremediation by *A. schaueriana*) CFU g^{-1}. After the 30th day, there was an increase in the microbial community in the phytoremediation model, with values from 8×10^6 to $1^6 \times 10^6$ CFU g^{-1}; however, there was a drastic decrease in the number of microorganisms in the intrinsic bioremediation model (1.8×10^6 CFU g^{-1}). Fig. 4 presents the total count of bacteria during the 90 days, with the data expressed in polynomials, with a coefficient of determination R^2 of 100% for the total bacterial counts.

3.5. Evaluation of the physiology of A. schaueriana used for phytoremediation

It is very important to investigate the physiology of a plant used in the application of phytoremediation, as the contaminants found in polluted sediments may often affect the growth of the plant species, making it unclear whether the plant is able to degrade the toxic compounds. The effects of TPHs in the sediment on the growth of *A. schaueriana* were evaluated by measuring the sizes of plants and their roots and by comparing the growth of plants in the contaminated sediment with the sediment reference (Fig. 5). Unexpectedly, there was a higher amount of growth observed in the phytoremediation experiments compared to the reference sediment, with an increase of 24% (18 cm) in plant growth and a 7% (3 cm) larger root. Thus, the biomass accumulation in the plants growing in the contaminated sediments was higher than the plants in non-contaminated sediment reference. Therefore, this plant species is very promising for the application of this technique, as the increased growth in the black mangrove plants most likely indicates that the plant has a good adaptation to the conditions found in contaminated sediments.

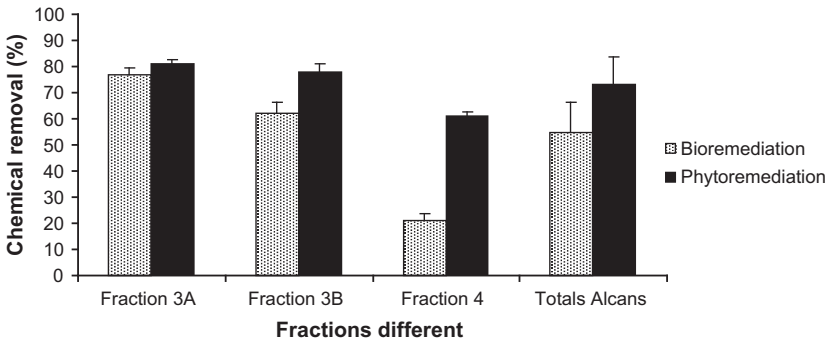

Fig. 2. Chemical removal (%) from the fractions in the different in remediation models after 90 days.

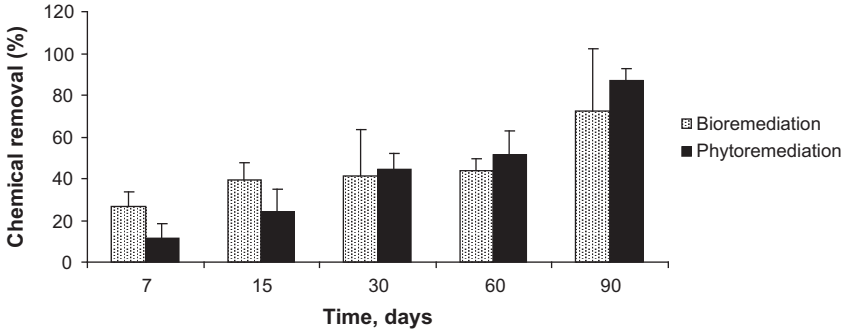

Fig. 3. TPH removal in the mangrove sediment as a function of time.

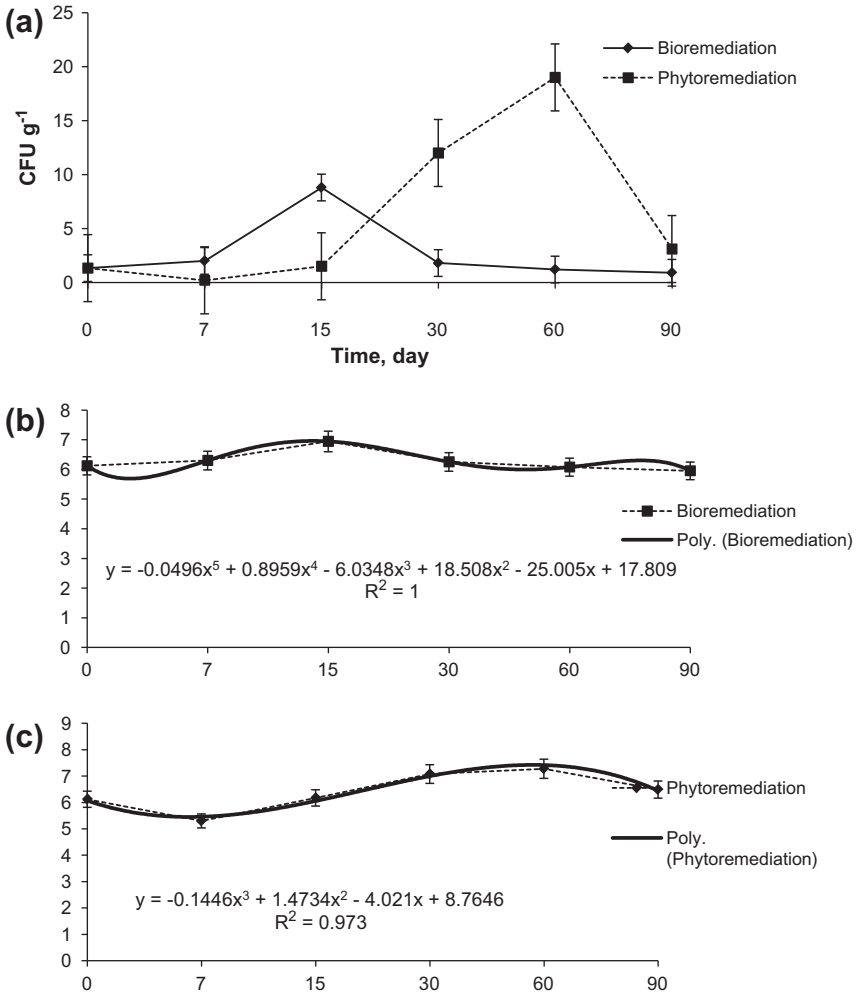

Fig. 4. Total bacterial count over 90 days, with the data expressed in a polynomial trend and a coefficient of determination R^2 at 100%. (a) Indicates the two remediation models; (b) indicates in the bioremediation model; (c) Indicates in the phytoremediation model.

4. Discussion

In this research, two models of treatment were developed using the intrinsic bioremediation and phytoremediation by *A. schaueriana* to assess the capability of removing organic compounds (TPHs) form contaminated mangrove sediments. The results showed that phytoremediation was more efficient for the degradation of the organic compounds, corroborating other studies that evaluated similar situations (Yuan et al., 2001, Huang et al., 2004, Tam and Wong, 2008; Yergeau et al., 2009). However, despite the fact that many recent studies found an efficient use of plants for the removal of TPHs, the traditional remediation techniques are still the most used in these situations (Jorgensen et al., 2000; McCarthy et al., 2004; Huang et al., 2005). Some of the difficulties encountered by these traditional techniques in the removal of TPHs in polluted industrial areas are the different concentrations of organic compounds in soils, sediments and groundwater, the application of isolated remediation processes and high costs (Mcnicoll and Baweja, 1995).

The successful implementation of remediation for sediments contaminated by the organic compounds in oil depends on the

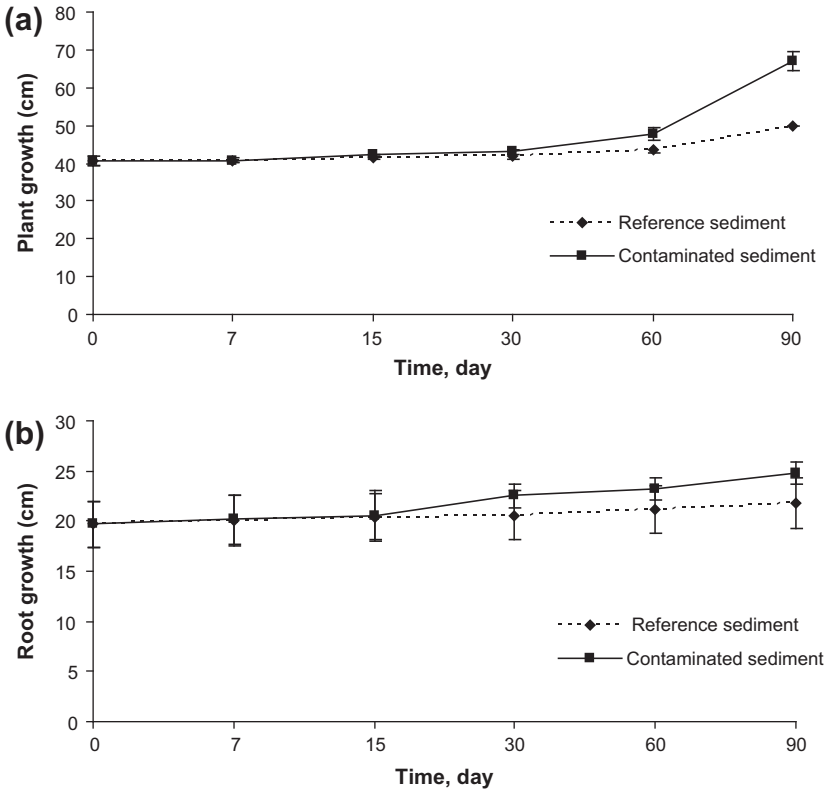

Fig. 5. Growth of *A. schaueriana* was evaluated by measuring the sizes of plants (a) and their roots (b).

efficiency of the degradation of the different fractions of alkanes present in sediments contaminated by TPHs, mainly the most recalcitrant alkanes, as in fraction 3B (C23–C34) and 4 (C34–C40) (Huang et al., 2001). The results of the present study showed that the phytoremediation by *A. schaueriana* was more effective at removing all of the fractions of organic compounds evaluated (3A (C16–23), 3B (C23–34) and 4 (C34–40)) in the experiment after 90 days. However, it is worth noting that the intrinsic bioremediation procedure was almost as efficient in the removal of fractions 3A (77%) and 3B (62%) as phytoremediation (3A – 3B and 81–72%), suggesting that the two models of remediation can be used in areas contaminated by these fractions of TPHs. However, a very significant degradation by *A. schaueriana* (73%) compared to bioremediation (21%) was found for fraction 4, which was most likely due to the plant's capacity to act in conjunction (phytostimulation) with the microorganisms in the rhizosphere, thus promoting rhizodegradation. It is also important to consider the degradation and transformation of compounds by plants that grow in mangrove sediments contaminated by TPHs (Rock, 1997; Cunningham et al., 1996, 1995).

Another important theme in this research was the evaluation of the number of viable bacteria that grew in the rhizosphere of *A. schaueriana*, compared to the amount that grew on the model of intrinsic bioremediation. The growth was higher during the first 15 days in the bioremediation model, but a higher concentration of bacteria in the rhizosphere was found from the 30th day until the 90th day. These results corroborate those for the 30th day of the experiment in which the phytoremediation model demonstrated a higher efficiency in the degradation of TPHs in relation to the bioremediation model. This result suggests that the plant species used in the experiment have a high ability to stimulate the degradation of organic compounds by bacterial communities, perhaps through some allelopathic compounds that are similar to organic compounds and stimulate the defenses of the communities, described by other studies (Rovira and Davey, 1979; Anderson et al., 1993, Walton et al., 1994; Espinosa et al., 2005). Other compounds that are extruded by plant roots act by stimulating the microorganisms that degrade TPH; these compounds include carbohydrates, organic acids and amino acids (Joner et al., 2002). The oxygenation promoted by the presence of mangrove roots in the black anoxic sediment of the mangrove forest may be another factor stimulating biodegradation (Siciliano et al., 2001; Weibner et al., 2002).

The results of the comparison of plant growth in the sediment reference with respect to the contaminated sediment showed that this plant species developed the greatest amount of biomass and roots in the substrate contaminated with the TPHs. These results confirm that *A. schaueriana* was not influenced by the toxic effects of the petroleum compounds present in the sediment, which differs from the reports of some researchers for sensitive species (Dowty et al., 2001; Culbertson et al., 2008; Peng et al., 2009; Nie et al., 2010).

The Tukey–Kramer test for multiple parameters showed a not significant difference between the developed models. The parametric analysis showed there was homogeneity of variances. The statistical treatment that was used showed the existence of significant differences between the two remediation models on efficiency the sediments remediation.

5. Conclusions

The results of this study show that the phytoremediation model using *Avicennia schaueriana* was more efficient in the degradation of different fractions of TPHs, although intrinsic bioremediation has also been effective for the lighter fractions. It is noteworthy that the phytoremediation by the plant species used was shown to be promising for the decontamination of sediments contaminated by the activities of the oil industry and indirectly to

contribute to the minimization of global warming via carbon sequestration; this phytoremediation technology is highly ecologically beneficial. Our study also found that intrinsic bioremediation has low efficacy with regard to the individual recalcitrant fractions of alkanes; in addition, its maximum efficiency only occurred in the first 30 days. Furthermore, data from the microbiological analysis revealed that the association of plants with the community of microorganisms in the rhizosphere increased the degradation of organic compounds in the sediment, thus promoting the development of plant biomass in the sediments contaminated by TPHs. However, whether the model of phytoremediation produced on this pilot scale will be as effective in situ as was observed under laboratory conditions remains to be evaluated. A more detailed study could combine these processes into a new procedure for the remediation of mangrove sediments contaminated by TPHs, especially with regard to heterogeneous sediment contamination at different depths. In addition, new research on the transformation of TPHs in the environment is necessary to evaluate whether the transformation processes produces toxic by-products that are detrimental to the resident organisms and human health.

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References

Alkorta, I., Garbisu, C., 2001. Phytorremediation of organic contaminants in soils. Bioresearch Technology 79, 273–276.
Alongi, D.M., 2002. Present state and future of the worlds mangroves forests. Environmental Conservation 29, 331–349.
Anderson, T.A., Guthrie, E.A., Walton, B.T., 1993. Bioremediation in the rhizosphere. Environmental Science and Technology 27, 2630–2636.
ASTM – American Society of Testing and Materials, 2011. D3694–2011 standard practices for preparation of sample containers and for preservation of organic constituents. West Conshohocken, PA.
Atlas, R.M., 1982. Microbial degradation of petroleum hydrocarbons: an environmental perspective. Microbiological Reviews 45, 180–209.
Bremner, J.M., Mulvaney, C.S., 1982. Nitrogen–Total. In: Page, A.L., Miller, R.H., Keeney, D.R. (Eds.), Methods of soil analysis, Part 2. Chemical and microbiological properties; SSSA Madison WI, pp. 595–624.
Brito, E.M., Duran, R., Guyoneaud, R., Goni-Urriza, M., Garcia de Oteyza, T., Crapez, M.A., 2009. A case study of in situ oil contamination in a mangrove swamp (Rio De Janeiro, Brazil). Marine Pollution Bulletin 58, 418–423.
Burns, K.A., Codi, S., Duke, N.C., 2000. Gladstone, Australia field studies:weathering and degradation of hydrocarbons in oiled mangrove and salt marsh sediments with and without the application of an experimental bioremediation protocol. Marine Pollution Bulletin 41, 392–402.
Culbertson, J.B., Valiela, I., Pickart, M., Peacock, E.E., Reddy, C.M., 2008. Long-term consequences of residual petroleum on salt marsh grass. Journal of Applied Ecology 45, 1284–1292.
Cunningham, S.D., Berti, W.R., Huang, J.W., 1995. Phytoremediation of contaminated soil. Trends in Biotechnology 13, 393–397.
Cunningham, S.D., Anderson, T.A., Schwab, A.P., Hsu, F.C., 1996. Phytoremediation of soil contaminated with organic pollutants. Advance in Agronomy 56, 55–71.
Dodd, R.S., Afzal-Rafii, Z., 2002. Evolutionary genetics of mangroves: continental drift to recent climate change. Trees: Structure and Function 16, 80–86.
Dowty, R.A., Shaffer, G.P., Hester, M.W., Childers, G.W., Campo, F.M., Greene, M.C., 2001. Phytoremediation of small-scale oil spills in fresh marsh environments: A mesocosm simulation. Marine Environmental Research 52, 195–211.
Duke, N.C., Meynecke, J.O., Dittmann, S., Ellison, A.M., Anger, K., Berguer, U., 2007. A world without mangroves? Science 317, 41–42.
Espinosa, E., Martinez, M.E., Torres, E.F., Rojas, M.G., 2005. Improvement of the hydrocarbon phytoremediation rate by *Cyperus laxus* Lam. inoculated with a microbial consortium in a model system. Chemosphere 59, 406.
Eysink, G.G.J., 1997. Recuperação de áreas de manguezais degradados através do uso de propágulos de *Rizophora mangle* acondicionado em estufa. Arquivo do Instituto Biológico 24, 1–65.
Folk, R.L., Ward, W.C., 1957. Brazos river bar: A study of significant of grain size parameters. Journal of Sedimentary Petrology 27, 3–26.
Gunther, T., Dornberger, U., Fritsche, W., 1996. Effects of ryegrass on biodegradation of hydrocarbons in soil. Chemosphere 33, 203–215.
Huang, X.-D., Glick, B.R., Greenberg, M.B., 2001. Combining remediation technologies increases kinetics for removal of persistent organic contaminants from Soil. In: Greenberg, B.M., et al. (Eds.), Environmental Toxicology and Risk Assessment, vol. 10. ASTM, pp. 271–278.
Huang, X.D., El-Alawi, Y., Penrose, D.M., Glick, B.R., Greenberg, B.M., 2004. A multi-process phytoremediation system for removal of polycyclic aromatic hydrocarbons from contaminated soils. Environmental Pollution 130, 465–476.
Huang, X.D., El-Alawi, Y., Gurska, J., Glick, B.R., Greenberg, B.M., 2005. Microchemical Journal 81, 139–147.
Joner, E.J., Corgie, S., Amellal, N., Leyval, C., 2002. Nutritional constraints to PAH degradation in a rhizosphere model. Soil Biology and Biochemistry 34, 859–864.
Jorgensen, K.S., Puustinen, J., Suortti, A.-M. 2000. Bioremediation of petroleum hydrocarbon-contaminated soil by composting in biopiles 107, 245–254.
Ke, L., Yu, K.S., Wong, Y.S., Tam, N.F., 2005. Spatial and vertical distribution of polycyclic aromatic hydrocarbons in mangrove sediments. The Science of the Total Environment 340, 177–187.
Lee, J.U., Kim, S.M., Kim, K.W., Kim, I.S., 2005. Microbial removal of uranium in uranium-bearing black shale. Chemosphere 59, 147–154.
McCarthy, K., Walkerb, L., Vigorenc, L., Barteld, J., 2004. Remediation of spilled petroleum hydrocarbons by in situ landfarming at an arctic site. Science and Technology 40, 31–39.
Mcnicoll, D.M., Baweja, A.S., 1995. Bioremediation of Petroleum-Contaminated Soils: An Innovative, Environmental Friendly Technology. Environment Canada, 15p.
Moreira, I.T.A, Freitas, P.F., Nascimento, R.S.A., Oliveira, O.M.C., Triguis, J.A. 2010a. Selection of species plant mangrove for assessment phytoremediation of contaminated sediments by in oil and derivatives. In: Rio Oil & Gas Expo and Conference, IBP2899_10.
Moreira, I.T.A, Martins, C.M.S., Oliveira, M.C., Oliveira, O.M.C., Triguis, J.A., 2010b. Detailed and confirmatory investigation in an area with contaminated sediment, in order for the application of phytoremediation. In: XII Workshop and Congress Geochemistry Organic American Latin Association (ALAGO).
Nelson, D.W., Sommers, L.E., 1982. Total carbon, organic carbon and organic matter. In: Page, A.L., Miller, R.H., Keeney, D.R. (Eds.), Methods of Soil Analysis, Part 2. Chemical and Microbiological Properties. SSSA Madison, WI, pp. 539–577.
Nie, M., Yang, Q., Jiang, L.F., Fang, C.M., Chen, J.K., Li, B., 2010. Do plants modulate biomass allocation in response to petroleum pollution? Biology Letters. http://dx.doi.org/10.1098/rsbl.2010.0261.
Oliveira, O.M.C. 2008. Processos de Biorremediação em áreas influenciadas por atividades petrolíferas. Rio Oil & Gás. IBP174_08.
Olsen, S.R., Dean, L.A., 1982. Phosphorus. In: Page, A.L., Miller, R.H., Keeney, D.R. (Eds.), Methods of Soil Analysis Part 2 Chemical and Microbiological Properties. SSSA Madison, WI, pp. 1035–1049.
Peng, S.W., Zhou, Q.X., Cai, Z., Zhang, Z.N., 2009. Phytoremediation of petroleum contaminated soils by *Mirabilis jalapa* L. in a greenhouse plot experiment. Journal of Hazardous Materials 168, 1490–1496.
Rock, S., 1997. Phytoremediation. In: Freeman, H. (Ed.), Standard handbook of hazardous waste treatment and disposal, 2nd ed. McGraw Hill Inc, New York, USA, pp. 93–112.
Romeiro, R.S., 2001. Métodos em bacteriologia de plantas. Viçosa, UFV, pp. 279.
Rovira, A.D., Davey, C.B., 1979. In: Carson, E.W. (Ed.), The plant root and its environment. University Press of Virginia, Charlottesville, pp. 153–204.
Salt, D. E., Smith, R. D., Raskin, I. 1998. Annual Review of Plant Physiology 49, 643–668.
Santos, H.F., Carmo, F.L., Paes, J.E.S., Rosado, A.S., Peixoto, R.S., 2011. Bioremediation of mangroves impacted by petroleum. Water, Air, and Soil Pollution 216, 329–350.
Seabra, P.N. 2008. Biorremediação de solos contaminados por petróleo e derivados. In: Microbiologia Ambiental 2, 548–570.
Siciliano, S.D., Fortin, N., Mihoc, A., Wisse, G., Labelle, S., Beaumier, D., Ouellette, D., Roy, R., Whyte, L.G., Banks, M.K., Schwab, P., Lee, K., Greer, C.W., 2001. Selection of specific endophytic bacterial genotypes by plants in response to soil contamination. Applied and Environment Microbiology 67, 2469–2475.
Susarla, S., Medina, V.F., McCutcheon, S.C., 2002. Phytoremediation: An ecological solution to organic chemical contamination. Ecological Engineering 18, 647–658.
Tam, N.F., Wong, Y.S., 2008. Effectiveness of bacterial inoculum and mangrove plants on remediation of sediment contaminated with polycyclic aromatic hydrocarbons. Marine Pollution Bulletin 57, 716–726.
U.S. EPA, 2000, Introduction to phytoremediation. EPA/600/R-99/107, Washington, DC, (February).
Walton, B.T., Guthrie, E.A., Hoylman, A.M. 1994. Toxicant degradation in the rhizosphere. In: Anderson, T.A., Coats, J.R. (Eds.), Bioremediation through Rhizospere Technology. ACS Series 563, Washington DC., pp. 11–26.
Weibner, A., Kuschk, P., Stottmeister, U., 2002. Oxygen release by roots of *Typha latifolia* and *Juncus effuses* in laboratory hydroponic systems. Acta Biotechnologica 1–2, 209–216.
Wilkie, M.L. and Fortuna, S. 2003. Status and trends in mangrove area extent wordwide. FAO Working Paper FRA 63.
Yergeau, E., Arbour, M., Brousseau, R., Juck, D., Lawrence, J.R., Masson, L., et al., 2009. Microarray and real-time pcr analyses of the responses of high-arctic soil bacteria to hydrocarbon pollution and bioremediation treatments. Applied and Environmental Microbiology 75, 6258–6267.
Yu, X., Zhou, P., Liu, Y., Hu, H., 2005. Detoxification of cyanide by woody plants. Archives of Environmental Contamination and Toxicology 49, 150–154.
Yuan, S.Y., Chang, J.S., Chang, B.V., 2001. Biodegradation of phenanthrene in river sediment. Chemosphere 43, 273–278.