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Research Article

Phytoremediation of Crude Oil Polluted Water by *Pistia stratiotes* L.

Ochekwu Edache Bernard and Ezekwe Thadeus Chukwuemeka

Department of Plant Science and Biotechnology, Faculty of Science, University of Port Harcourt, Nigeria

Abstract

Background and Objective: The exploration and exploitation of crude oil in the Niger Delta has led to the pollution of water bodies in this area of Nigeria. This study was carried out to determine the phytoremediation potential of aquatic macrophyte in a crude oil polluted aquatic habitat. *Pistia stratiotes* (water lettuce) was used as a phytoremediating aquatic macrophyte to evaluate its remediation potential on crude oil polluted water. **Materials and Methods:** Four different levels of pollutions of 0, 50, 100 and 150 mL were polluted in a 20 L of water and these pollutions were replicated 5 times. Physicochemical parameters as temperature, conductivity, total dissolved solid (TDS), dissolved oxygen (DO), salinity, turbidity, pH, total hydrocarbon content (THC) were analyzed. Also, heavy metals Cu, Zn, Cd and Pb were analyzed in the water and plant samples. **Results:** Statistical ($p \leq 0.05$) analysis reveals that *Pistia stratiotes* despite its low level of tolerance and negative response to high doses of crude oil pollution reduced the THC of water and heavy metals in a negligible amount which are non-significantly ($p \leq 0.05$) different except for Pb which is significantly ($p \leq 0.05$) lower than other heavy metals (Cu, Zn and Cd). **Conclusion:** Results from this experiment shows that the aquatic weed *Pistia stratiotes* has potential for phytoextraction and rhizofiltration which are forms of phytoremediation.

Key words: Crude oil, physicochemical, phytoremediation, water lettuce, rhizofiltration, aquatic habitat

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Corresponding Author: Ochekwu Edache Bernard, Department of Plant Science and Biotechnology, Faculty of Science, University of Port Harcourt, Nigeria

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Crude oil is a natural occurring toxic flammable liquid which varies in appearance depending on its composition. Crude oil is composed mostly of alkenes, cycloalkanes, aromatic hydrocarbons, trace amount of metals such as iron, nickel, copper, lead and vanadium and other organic compounds such as nitrogen, oxygen and sulphur. Crude oil is a black or dark brown liquid consisting of a mixture of hydrocarbons of various molecular weights and other organic compounds that are found in geological formation beneath the earth's surface¹. Crude oil in surface and ground water does not only affect natural but also causes a major strain in aquatic ecosystem². Remediation of crude oil contaminated site using conventional practices such as pump-and-treat and dig-and dump techniques are very expensive and has limited potential and is usually applicable to small areas. Conventional approaches to remediate crude oil polluted aquatic environment often make water unfit and unsuitable for agriculture and other uses. Phytoremediation is an efficient use of plants to remove, detoxify or immobilize environmental contaminants in growth matrix (soil, water or sediments) through the natural, biological, chemical or physical activities and processes by plants³. It also gives the solution to environmental problems through the use of plants which mitigate the environmental problems without the need to excavate the contaminated materials and dispose of it elsewhere⁴. Plants are unique organisms equipped with remarkable metabolic and absorptive capabilities as well as transport systems that can take up nutrients or contaminants selectively from the ground matrix-soil or water⁵.

Pistia stratiotes L. belong to the family Araceae and often called water cabbage or water lettuce. Its native distribution is uncertain but was first describe from the Nile near Lake Victoria but now present in nearly all tropical and subtropical fresh water way^{6,7}. *Pistia stratiotes* as an aquatic, floating, rosette-forming stemless stoloniferous herb, with sessile and glaucous leaves. The leaves can be up to 14 cm long and they are light green with parallel veins, wavy margins covered in short hairs which form basket-like structures which trap air bubbles increasing the plant buoyancy. Pollutants are phytotoxic and can affect the normal physiological and genetic constituent of plants¹. However, plants have evolved a great diversity of adaptation to handle accumulated pollutants that occur in the environment. Some plants are tolerant and tend to accumulate toxic chemicals through the production of antioxidants such as ascorbic acid, phenolic compounds, beta carotene etc and also through the process of phytochelatin and metalthionin⁸. Hence the objectives of the study are to access the tolerant level of *P. stratiotes* to crude oil contaminated water and to determine the

accumulation of heavy metals in crude oil polluted water using *P. stratiotes*.

MATERIALS AND METHODS

Study area: The experiment was conducted in a controlled environment at Port Harcourt, Rivers State for 1 months from the duration of May, 2017 to August, 2017. The crude oil used for the study was obtained from Niger Delta Petroleum Resources (NDPR) Ahoada, Rivers State while the plant species were collected from a pond in Opolo, Yenagoa in Bayelsa state. The 17 L of fresh water fetched from New Calabar River in Choba Rivers state was poured into 16 containers of 20 L size each.

Methodology: Measuring cylinder was used to dispense 0 mL (control), 50, 100 and 150 mL of crude oil giving rise to 0% pollution, 0.33% pollution, 0.67% pollution and 1% pollution (v/v), respectively. The different levels of pollution serve as the treatment and were replicated 5 times. A completely randomized experimental design was employed in the experiment. Water physico-chemical analysis conducted in the water before and after pollutant's introduction includes pH, total dissolved solids (TDS), dissolved oxygen (DO), temperature, conductivity, turbidity and salinity using a Multiparameter water checker (HANNA H19828).

Pistia stratiotes were introduced to the non-polluted and polluted containers 2 weeks after pollution and this was left till the next 8 weeks. At the end of the experiment the fresh and dry weights of *P. stratiotes* were determined by placing the plant samples on a weighing balance (Model TE 1533) and the corresponding weights recorded as the fresh weight. The plant samples were then placed in different paper bags and kept in the oven (Model No: Carbolite, Sheffield England) for 48 h at 80°C.

Hydrocarbon content: Total hydrocarbon content (THC) of water samples were determined by weighing the samples into a clean test tube. Chloroform was added and a glass rod was used for proper stirring. The extract was decanted and read at a wavelength of 370 nm in a data logging spectrophotometer using chloroform as a blank.

Chemical compounds: Heavy metals (Cu, Zn, Cd and Pb) were determined by cutting into smaller portions of plant samples and placing them into foil papers at 100°C for 48 h. The dried plant samples were then grinded into fine particles using clean acid wash mortar and pestle. The procedure employed for the digestion of the plant samples was the standard procedure⁹. The metallic content of the digested samples was read using the atomic absorption spectrophotometer (AAS).

Statistical analysis: Data collected were subjected to Analysis of Variance and the means were separated at 5% level of significance using least significant difference (LSD) using statistical package¹⁰.

RESULTS

The mean temperature level of the water was not different statistically with the introduction of different levels of concentrations (Table 1). This was not the case with mean conductivity level in which case, 50 mL concentration was significantly ($p \leq 0.05$) higher than other concentrations. No significant ($p \leq 0.05$) difference exists between 100 and 150 mL concentration while the control (0 mL) was seen to be significantly ($p \leq 0.05$) lower than the other concentrations. The same trend recorded for conductivity was also recorded for total dissolved solid (Table 1). This was not the case with mean dissolved oxygen levels that had the control (0 mL) significantly ($p \leq 0.05$) higher than other concentrations and among the other concentrations no significant ($p \leq 0.05$) difference was recorded (Table 1). The reverse statistical implication was observed in the mean values of salinity as opposed to dissolved oxygen in which case concentrations 50, 100 and 150 mL were not significantly ($p \leq 0.05$) different from each other but was significantly ($p \leq 0.05$) higher than the control (Table 1). Turbidity had 150 mL concentration has the highest mean statistically ($p \leq 0.05$) with concentration

100 placed next but 50 and 0 mL had no difference statistically. The effect of the 150 mL concentration was seen as a result of the high level of concentration among the rest i.e., 0, 50 and 100 mL (Table 1). Mean pH value had the same trend like dissolved oxygen and this shows that the introduction of the different levels of concentrations affect the pH value, while the pH of the control was almost neutral the others were slightly acidic. The control (0 mL) was significantly ($p \leq 0.05$) higher than other concentrations level (Table 1).

Dry weight of the plant samples at the end of the experiment reveals that concentrations 50 and 150 mL had a more stable weight than other concentrations. Even though, the aquatic environment was polluted the plants absorbed the pollutants and out competed the control which was not polluted. The control was significantly ($p \leq 0.05$) lowest in ranking. Total hydrocarbon content had concentration 150 mL to be significantly ($p \leq 0.05$) highest followed by 100 mL then 50 mL and finally the control. This is correct because the more the level of pollutant the high the Total petroleum content (Table 2).

Trace amount of heavy metals were detected in the polluted water environment (50, 100 and 150 mL). No significant ($p \leq 0.05$) difference was recorded for all the heavy metals in 50 but 100 and 150 mL had the same trend in which case Pb was significantly ($p \leq 0.05$) lower than the other heavy metals which were not significantly different from each other i.e., Cu, Cd and Zn (Table 3).

Table 1: Mean values of water physio-chemical parameters

Treatments	Mean						
	Temperature (°C)	Conductivity ($\mu\text{S cm}^{-1}$)	TDS (mg L^{-1})	DO (mg L^{-1})	Salinity (mg L^{-1})	Turbidity (NTU)	pH
0 mL (Control)	26.160 ^a	32.80 ^c	16.40 ^c	2.07 ^a	0.010 ^b	2.042 ^c	8.19 ^a
50 mL	26.130 ^a	71.40 ^a	36.20 ^a	1.67 ^b	0.028 ^a	5.292 ^c	7.59 ^b
100 mL	26.570 ^a	65.00 ^b	32.40 ^b	1.45 ^b	0.030 ^a	24.720 ^b	7.52 ^b
150 mL	26.190 ^a	66.80 ^b	33.80 ^b	1.21 ^b	0.030 ^a	58.800 ^a	7.72 ^b
LSD	0.8803	3.10	2.01	0.33	0.006	3.861	0.212

Values above are means of 5 replicates, means with the same letters are not significantly ($p \leq 0.05$) different from each other

Table 2: Mean dry weight (g) at the end of the experiment and mean total hydrocarbon content (mg L^{-1})

Treatments	Dry weight (g)	THC
0 mL (Control)	45.31 ^c	0.0 ^d
50 mL	70.73 ^a	5.1 ^c
100 mL	67.04 ^b	8.3 ^b
150 mL	69.74 ^a	12.6 ^a
LSD	1.858	2.01

Means with the same letters are not significantly ($p \leq 0.05$) different from each other

Table 3: Mean heavy metals (g) of the different levels of pollution

Heavy metals	0 mL	50 mL	100 mL	150 mL
Cu	0	0.246 ^a	0.023 ^a	0.05 ^a
Cd	0	0.175 ^a	0.05 ^a	0.07 ^a
Zn	0	0.237 ^a	0.24 ^a	0.26 ^a
Pb	0	0.0204 ^a	0.12 ^b	0.17 ^b
LSD	0	0.18808	0.0441	0.0445

Means with the same letters are not significantly ($p \leq 0.05$) different from each other

DISCUSSION

Phytoremediation using *P. stratiotes* was seen to remediate the polluted aquatic environment. Though, the experiment was conducted in a controlled environment, it showed promises that in the natural environment it can help in the breakdown of crude oil and use C and H for its growth and development. As evident in the statistical interpretation, there was a clear difference between the polluted treatments (50, 100 and 150 mL) and the non-polluted treatment (control or 0 mL) in all the physicochemical analysis conducted apart from temperature and this may be as a result of the closeness in percentage pollution between the control and other concentrations.

The heavy metals (Cu, Cd, Zn and Pb) were present only in the polluted treatments but they were all in trace amount. *P. stratiotes* showed ability to absorb heavy metals in crude oil polluted water. The tolerance of *P. stratiotes* was variably sensitive to the effect of pollution levels. Result of this controlled experiments also showed that *P. stratiotes* can remediate slightly polluted water environment. Variations in tolerant levels of polluted condition could be attributed to nutrient enhancement of the aquatic habitat following successive crude oil degradation because some plants can tolerate certain concentration levels and *P. stratiotes* is one of those plants. This is in confirmation who reported that crude oil pollution could interfere with plant growth and development but a slight contamination in the order of one percent of crude oil will improve growth and development^{11,12}. Researchers have also observed improvement in growth of plants on sites contaminated with oil, despite the damaging effect of oil pollution. This commentary agrees with the findings of a study on the impact of oil concentration in *Scirpus pungens* (a fresh water wetland plants) which showed a stimulated growth at concentration less than 4.59 kg sediment as against controlled plant growth and had decreased growth at 27.49 kg sediment concentration, meaning that plants were likely to survive and grow in sediments contaminated with crude oil in a concentration rate comparable to oil spill incidence¹³.

The biomass of species within pollution levels and control recorded a significant ($p \leq 0.05$) difference. There was increase in biomass against control in a decreasing trend as pollution level increased. The incidence of crude oil on macrophyte could endanger either positive or negative response on this plant as it could as nutrient or toxicant additive. Similar investigation of sub lethal plant stress has been often

detected by the net CO₂ exchange response (decrease photosynthesis and water-use efficiency) but longer term or integrated stress response are best assessed in terms of shift in biomass allocation patterns including increased root to shoot ratios, increased proportions of dead tissue and decreased total production^{14,15}.

The implication of this study is that the aquatic macrophyte employed for this study has the potential to phytoaccumulate contaminants in crude oil polluted water. It should be noted that the abundance and distribution of the aquatic macrophyte shouldn't be limited and the quantity of oil spill determines the phytoaccumulating potentials of the aquatic macrophyte. It is highly recommended that *P. stratiotes* though an invasive species can help mitigate the perturbation and destruction of aquatic ecosystem by crude oil. A limitation of this study is that it was carried out in controlled environment.

CONCLUSION

This study has helped to see the usefulness of *P. stratiotes* in crude oil polluted water in the Niger Delta Area of Nigeria. The more the number of *P. stratiotes* in polluted water the greater the ability in remediating. Also, less concentration of crude oil spills can be easier achieved than heavily polluted water bodies.

SIGNIFICANCE STATEMENT

This study discovers the potentials of *Pistia stratiotes* that can be beneficial for the remediation of crude oil polluted water. This study has helped in understanding that *P. stratiotes* in a crude oil polluted water is sensitive to various pollution concentrations. *Pistia stratiotes* performed well in terms of heavy metals accumulation and it showed potentials for phytoremediation of polluted aquatic environment. This study will help the researcher to uncover the critical areas of phytoremediation that many researchers were not able to explore. Thus a new theory on potentials of *P. stratiotes* on crude oil polluted water may be arrived at.

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