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Effect of crude oil pollution on the *in vitro* antioxidant potential of methanol extracts of *Magnifera indica* Leaf

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Abstract

Pollution continues to be a significant factor for adverse health implication and cause of death globally due to its strong ink to oxidative stress. Oxidative stress is a condition of Redox imbalance in vivo leading to oxidative damage by reactive oxygen/nitrogen species. Crude oil and its products in the water bodies of South ad Eastern Nigeria are a constant source of pollution to the environment. Plants have been used for thousands of years due to their high levels of antioxidants, which help in quenching free radicals damage to effectively treat both acute and chronic disorders. The aim of this study is to investigate the antioxidant status of *Magnifera indica* leaves in crude oil polluted sites compared to unpolluted sites. The antioxidant status is accessed through the DPPH assay which measures the free radical removal ability and the FRAP assay, which evaluates the reducing power of the extract. The result shows that DPPH radical inhibition (%) was 60.00, 90.00, and 100.00 for *Magnifera indica* polluted leaf, *Magnifera indica* unpolluted and ascorbic acid respectively. Similarly *Magnifera indica* polluted leaf, and *Magnifera indica* unpolluted leaf also showed significant free radical scavenging action over each other and the FRAP value (µM) was 0.1, 0.2, 1.1 for *Magnifera indica* polluted leaf, *Magnifera indica* unpolluted and ascorbic acid respectively. Similarly *Magnifera indica* polluted leaf, and *Magnifera indica* unpolluted leaf also showed significant FRAP values over each other. The results reveal important insights into the effect of environmental contamination and the vital role of *Magnifera indica* owing to its antioxidant properties. The findings indicate that crude oil pollution is real and buttresses the importance of plants in management of diseases.

Keywords: Oxidative stress; Crude oil; Pollution; Antioxidant; *Magnifera indica*

1. Introduction

Nigeria since the oil boom has suffered damage to aquatic and terrestrial life due to oil spillage. In the oil-abundant Niger Delta area, many people have been exposed to pollutants from water, land and air to become a health and environmental hazard. Heavy metals from crude oil like Arsenic, Chromium and others accumulate in water, crops and also in aquatic animals which are consumed by animals and humans, thus these heavy metals discover their path into the living system and wreak havoc.

Mango (*Mangifera indica* Linn.) belongs to the family *Anacardiaceae* has been recognized as the essential fruit crop of traditional importance and an economically significant tropical fruit globally (Barreto *et al.,* 2008). Mango leaves (MLs) are excellent sources of various minerals and vitamins, including nitrogen, potassium, phosphorus, iron, sodium, calcium, magnesium. Protein is a significant bio-macromolecule found in mango leaves, specifically types A, B, E, and C.

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MLs can be utilized as another source of livestock feeding in developing countries for alleviating the food shortage for livestock. Various components of the mango tree, like the leaves, flowers, bark, fruits, and seeds, contain important nutrients and bioactive compounds (Kulkarni and Rathod, 2018).

Extracts of the *Mangifera indica* is a potential traditional medicine to cure diabetes, bronchitis, diarrhea, asthma, kidney, scabies, respiratory problems, syphilis, and urinary disorders (Kulkarni *et al.,* 2014). Some bioactive constituent of *Mangifera indica* includes mangiferin, phenolic acids, benzophenones, and other antioxidants such as flavonoids, carotenoids, quercetin, isoquercetin, ascorbic acid, and tocopherols. Studying the effect of pollution caused by crude oil on the *in vitro* antioxidant potential of methanol extract of *Magnifera indica* leaf is significant due to the potential environmental implications and the importance of understanding the impact of pollution on natural resources. This research can shed light on the potential implication of pollution caused by crude oil on the antioxidant properties of an important medicinal plant like *Magnifera indica*. Understanding the antioxidant potential of *Magnifera indica* is not only relevant to ecological preservation but also holds potential human health benefits. Traditional medicine has utilized medicinal plants with antioxidants to treat health conditions caused by oxidative stress. The goal of this research is to investigate the antioxidant status of *Magnifera indica* leaves in crude oil polluted sites compared to unpolluted sites.

2. Materials and methods

2.1. Sample collection and identification

The polluted plant Samples were collected from Ibono (Mobil Terminal area) local Government Area of Akwa-Ibom, while the unpolluted sample were collected from Umudike in Ikwuano local Government Area of Abia State in June 2023. The plant samples were authenticated by Professor Garuba Omosun from the College of Natural Science at Michael Okpara University of Agriculture, Umudike, Nigeria. Voucher specimens of both samples were preserved in the herbarium of the Department of Veterinary Physiology and Pharmacology at Michael Okpara University of Agriculture, Umudike, Nigeria, under the reference number MOUAU/VPP/23/016.

2.2. Preparation and extraction of the plant sample

The leaves were washed using tap water and were subsequently air-dried for a period of two weeks, with periodic turning to inhibit the growth of fungi, as outlined by Bonjar (2004). These dried plant materials were then finely ground into a powder using a milling machine. The resulting powdered plant samples were stored in a sealed, sterile container at room temperature. The Soxhlet apparatus set at 40 °C was used to extract the plant material exhaustively. The extract obtained was then concentrated in a water bath set at 40 ℃, resulting in concentrated extracts that had a dark green appearance. These concentrated extracts were stored in a refrigerator until they were required, following the methodology described by Bonjar (2004).

2.3. Antioxidant Test

2.3.1. 1 2, 2-Diphenyl-1-Picrylhydrazyl (DPPH) photometric assay

The free radical scavenging activity of the extract was investigated by the DPPH assay (Mensor *et al.*, 2001) using spectrophotometer.

2.3.2. Ferric reducing antioxidant power

The ferric reducing antioxidant power was carried out as described by Benzie and Strain, (1999).

2.4. Statistical analysis

The present data were statistically analyzed using SPSS Version 22. The mean values were separated using the Duncan multiple tests. The different levels of significance within the groups were examined using one-way analysis of variance (ANOVA). The data were expressed as mean±SD and considered significant at $p<0.05$.

3. Result

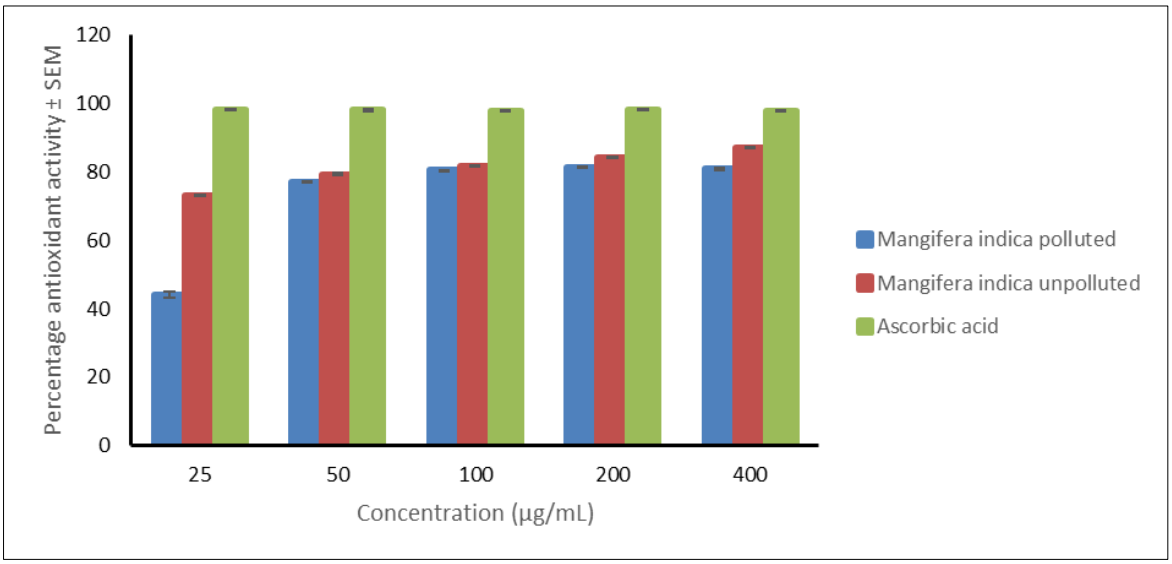

Figure 1 The DPPH radical scavenging property of crude oil polluted and unpolluted *Magnifera indica* leaf

The chart for DPPH assay results depicts the % inhibition of free radicals with increase in concentration of Ascorbic acid, *Magnifera indica* polluted leaf and *Magnifera indica* unpolluted leaf in µg/mL. This graph is representative of the antioxidant property of the formulation in terms of free-radical scavenging capacity. The IC_{50} value was determined using the graph. The DPPH radical inhibition (%) was 60.00, 90.00, and 100.00 for *Magnifera indica* polluted leaf, *Magnifera indica* unpolluted and ascorbic acid respectively. Similarly *Magnifera indica* polluted leaf, and *Magnifera indica* unpolluted leaf also showed significant free radical scavenging acti on over each other.

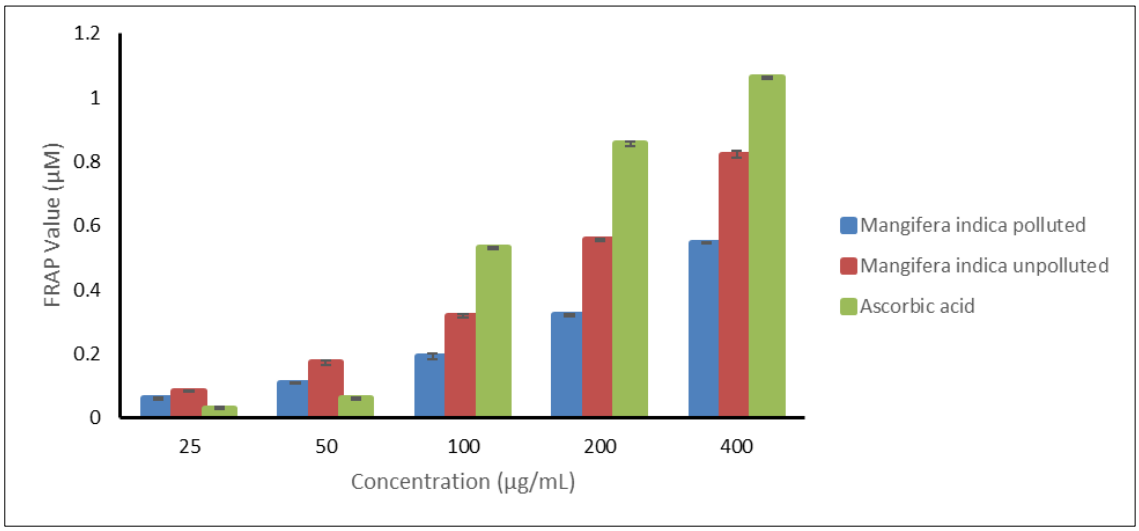

Figure 2 The ferric reducing antioxidant power (FRAP) of crude oil polluted and unpolluted *Magnifera indica* leaves

Chart for FRAP assay result shows the FRAP value of free radicals with increase in concentration of Ascorbic acid, *Magnifera indica* polluted leaf and *Magnifera indica* unpolluted leaf in µg/mL. The FRAP value (µM) was 0.1, 0.2, 1.1 for *Magnifera indica* polluted leaf, *Magnifera indica* unpolluted and ascorbic acid respectively. Similarly *Magnifera indica* polluted leaf, and *Magnifera indica* unpolluted leaf also showed significant FRAP values over each other.

4. Discussion

Ascorbic acid, also called vitamin C, is a well-established antioxidant. The 100.00% DPPH radical inhibition observed for ascorbic acid in this study is consistent with the extensive body of research that confirms its potent antioxidant

properties. Ascorbic acid's ability to counteract free radicals is well-documented, making it a standard for comparing the antioxidant potential of other substances. The extract derived from *Magnifera indica* that were not exposed to pollution demonstrated a high level of DPPH radical inhibition, at 90.00%. This suggests that this extract contains significant quantities of bioactive compounds with strong antioxidant properties. Previous studies have also highlighted the antioxidant potential of *Magnifera indica,* corroborating the results obtained in this study. For example, research by Chen *et al*. (2015) demonstrated the presence of phenolic compounds and also flavonoids in *Magnifera indica*, which are known for their antioxidant effects (Chen *et al*., 2015). The extract from *Magnifera indica* exposed to pollution showed a moderate level of DPPH radical inhibition, at 60.00%. While this inhibition is lower compared to the unpolluted leaf extract, it is important to note that even the polluted leaf extract exhibited significant antioxidant activity. The results demonstrate that both polluted and unpolluted *Magnifera indica* possess antioxidant activity, as indicated by their increasing FRAP values with higher concentrations. This discovery aligns with past research that has also identified *Magnifera indica* as a potential source of antioxidants. A study by Ajayi *et al*. (2018) investigated the antioxidant potential of *Magnifera indica* and reported significant free radical scavenging properties. The current study reinforces and extends this understanding. Comparison with Ascorbic Acid: Ascorbic acid, a well-known antioxidant, exhibited the highest FRAP values among the tested samples. This observation is consistent with the recognized strong antioxidant activity of ascorbic acid, which has been extensively documented in previous studies (Saeed *et al*., 2019). The comparison acts as a point of reference for evaluating the relative antioxidant potency of *Magnifera indica* leaves. The graph and data shown in the study indicate that both polluted and unpolluted *Magnifera indica* leaves showed significant differences in their FRAP values compared to each other. These differences suggest that the pollution status of the leaves may impact their antioxidant potential. Previous studies have also explored the implication of environmental factors, such as pollution, on the phytochemical composition and antioxidant activity of plants (Nabavi *et al*., 2015). The results in this study support the notion that the pollution status of the plant can influence its antioxidant properties.

5. Conclusion

This research examined the implication of pollution caused by crude oil on the *in vitro* antioxidant potential of methanol extract from *Magnifera indica* leaves. The results reveal important insights into the effect of environmental contamination on the antioxidant properties of this medicinal plant. The findings indicate that crude oil pollution had a significant influence on the antioxidant potential of *Magnifera indica* leaves. The *in vitro* antioxidant activity, as assessed by DPPH radical scavenging and total phenolic content, was notably reduced in the polluted samples compared to the unpolluted ones. This suggests that the exposure to crude oil contaminants resulted in a decrease in the plant's ability to combat oxidative stress and neutralize free radicals.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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