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Comparative Study on Effect of *Chrysophyllum albidum* Medicinal Plant from Crude Oil Polluted and Non-crude Oil Polluted Areas on Selected Biochemical Parameters in Rats

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Authors' contributions

This work was carried out in collaboration among all authors. Authors GE and BA designed the study. Authors EA and OM performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors GE, BA and EA managed the analyses of the study. Author PD managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Comparative study on effect of *Chrysophyllum albidum* medicinal plant from crude oil polluted and non-crude oil polluted areas on selected biochemical parameters in rats was evaluated. Leaves of *C. albidium* were collected from a botanical garden (non-crude oil polluted site) and Okrika (crude oil polluted site), and studied. The leaves passed for heavy metals analyses and heavy metals such as mercury (0.38 ±0.07 mg/100 g), lead (3.06±0.40 mg/100 g), cadmium (0.09±0.00 mg/100 g), copper (1.00±0.18 mg/100 g), chromium (0.23±0.01 mg/100 g), and cobalt (4.90±1.22 mg/100 g) were observed in leaves of *C. albidum* from crude oil polluted area. Rats placed on compounded feed of *C. albidium* leaves from crude oil polluted area revealed marked degeneration in

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haematological indices, liver enzymes, urea and creatinine of the kidney. These observed degeneration could be linked to toxicity of the heavy metals found in the leaves of *C. albidum* from crude oil polluted area since leaves of *C. albidum* are known to have medicinal potency that could protect the integrity of internal organs and tissues. There is need to properly inform those who rely on medicinal plants from crude oil polluted areas of the possible dangers such may pose in the body. This study has evaluated the comparative effect of *C. albidum* medicinal plant from crude oil polluted and non-crude oil polluted areas on selected biochemical parameters in rats.

Keywords: Medicinal plant; biochemical parameters; Chrysophyllum albidum; crude oil polluted area.

1. INTRODUCTION

The importance of medicinal plants has long been noted [1-5]. Different authors have reported evaluations on medicinal plants and their products in relation to the body [5-9]. Different studies have authenticated the potency of so many medicinal plants against different disease conditions [1, 3,10-20]. It has been reported that natural compounds found in plants or their synthetic forms are the basis of modern pharmacopeia [21-22]. According to Ogunkunle and Ladejobi [11], and Sofowora [18], a medicinal plant is one whose one or more of its organs contains substances that can be used for therapeutic purposes or which are precursors for the synthesis of useful drugs. These substances are phytochemicals and phytonutrients [23-24], and are associated to the medicinal potency of medicinal plants [25-31]. Adebayo et al. [32] and Taylor et al. [33], noted that over 5000 plants are known to be used for medicinal purposes in Africa, but only a few have been described or studied.

Chrysophyllum albidum is among the 5000 plants used for medicinal purposes, which has been studied. The plant contains phytochemicals and phytonutrients. *C. albidum*, belongs to the family Sapotaceae. The plant is found in Eastern, Central and Western Africa [34]. The plant is called “agbalumo” and “udara” by Yoruba and Igbo tribes of Nigeria [35-37]. Different parts of *C. albidum* are used traditionally as medicinal plants throughout the world for a number of ailments. Their parts are extensively employed as raw materials in herbalism. Due to proven medicinal importance of the plant, not much has been done to consider the sites where it grows or where it is gotten from. It is not a gainsaying to state that when any plant is urgently needed, it is indiscriminately collected for use.

The Niger Delta is an area that produces the crude oil for which Nigeria is associated with. The area is highly associated with environmental degradation resulting from crude oil pollution. Herbalists in the area rely on medicinal plants that grow within for ailments. Okrika is among the towns found in Niger Delta area where herbalists rely on medicinal plants that grow within for ailments *C. albidum* is amongst the medicinal plants commonly employed against ailments in Niger Delta area.

This study comparatively evaluated the effect of *C. albidum* medicinal plant from polluted and non-polluted areas on selected biochemical parameters in rats.

2. MATERIALS AND METHODS

2.1 Collection and Identification of Plant Materials

The plant materials used in this study were collected from a crude oil polluted site in Okrika Rivers State, and a botanical garden (Non-crude oil polluted site) found in Owerri, Imo State, both in Nigeria. The plant materials were identified by Professor Ferdinand Nkem Mbagwu of Department of Plant Science and Biotechnology, Imo State, University Owerri, Nigeria as *C. albidum*. Their leaves were collected, air dried and crushed with pestle and mortar, then sieved to obtain the coarse powder, which was used to compound the feed used for further studies.

2.2 Heavy Metal Analysis

The identified leaves were analysed for heavy metals using atomic absorption spectrophotom-eter (Model: Unicam 9939/959) method to ascertain their level of heavy metals.

2.3 Experimental Animals

Thirty-six albino rats of Wistar strains weighing between 90-112 g were purchased from the animal colony of Department of Biochemistry, Gregory University, Uturu, Nigeria. The rats

were allowed to acclimatize in their new environment for five days before they were used for studies. The rats were separated into three major groups of I-III, with each group having two subgroups designated “a” and “b”. Each of the subgroup housed six rats. The rats were given compounded feed of *C. albidum* and rat feed. The rats` feed was a brand of commercial grower freshly obtained from a feed dealer along Abayi road, Aba.

Treatment given to the rats are as follows:

Group Ia: 5% of *C. albidum* (non-crude oil polluted area) + 95% normal feed + potable water.
Group Ib: 5% of *C. albidum* (crude oil polluted area) + 95% normal feed + potable water.
Group IIa: 25% of *C. albidum* (non-crude oil polluted area) + 75% normal feed + potable water.
Group IIb: 25% of *C. albidum* (crude oil polluted area) + 75% normal feed + potable water.
Group IIIa: 50% of *C. albidum* (non-crude oil polluted area) + 50% normal feed + potable water.
Group IIIb: 50% of *C. albidum* (crude oil polluted area) + 50% normal feed + potable water.

The treatments of experimental rats were in accordance to the National Institute of Health (NIH) guidelines for the care and use of laboratory animals [38]. The treatment lasted for 28 days.

2.4 Biochemical Studies

Rats from the various groups were weighed and sacrificed while under chloroform anesthesia after the treatment period. Blood was collected by direct cardiac puncture into heparin treated tubes for haematology analysis, while the blood for creatinine, urea and liver enzyme studies were collected in anticoagulant free tubes. The tubes were properly labeled for analysis.

Haematology indices such as Packed Cell Volume (PCV) was estimated using micro-haematocrit method as described by Alexandar and Griffths [39], haemoglobin level (Hb) was determined using cynomethaemoglobin as described Alexandar and Griffths [39], whereas white blood cells count (WBC) was estimated by visual means using the new improved Neubauer counting chamber as described by Dacie and Lewis [40]. Mean cell volume (MCV), Mean corpuscular, haemoglobin (MCH), and mean corpuscular haemoglobin concentration (MCHC) were estimated using the methods as described by Jain [41]. Urea, creatinine, as well as the liver enzymes considered such as aspartate aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase (ALP) were spectrophotometrically determined using the standard ready to use kits from Rondox Laboratory Ltd. Co. Antrim, United Kingdom.

2.5 Statistical Analysis

Results were presented as mean and standard deviations. Significant difference was established using students t-tests between two subgroups “a” and “b” of a main group at $p<0.05$.

3. RESULTS AND DISCUSSION

Result of heavy metals as presented in Table 1 shows the presence of heavy metals such as mercury (0.38 ±0.07 mg/100g), lead (3.06±0.40 mg/100 g), cadmium (0.09±0.00 mg/100 g), copper (1.00±0.18 mg/100 g), chromium (0.23±0.01 mg/100 g), and cobalt (4.90±1.22 mg/100 g) in leaves of *C. albidum* from crude oil polluted. Only copper (0.67±0.10 mg/100 g) was observed in leaves of *C. albidum* from non-crude oil polluted site. These heavy metals become very important when their toxicity to organs of the body is considered.

Table 1. Result of heavy metals in leaves of *C. albidum* from non-crude oil polluted and crude oil polluted sites

Heavy metal (mg/100 g)	Leaves from non-crude oil polluted site	Leaves from crude oil polluted site
Mercury	ND	0.38 ±0.07
Lead	ND	3.06±0.40
Cadmium	ND	0.09±0.00
Copper	0.67±0.10	1.00±0.18
Chromium	ND	0.23±0.01
Cobalt	0.05±0.01	4.90±1.22

Results are mean and standard deviation of triplicate determination; ND: Not Detected

Haematology is of diagnostic importance for short and long-term detection of future disease conditions in the body [41-44]. Haematological results as presented in Table 2, shows that aside Ia rats that had insignificant RBC increase ($p>0.05$) when compared to those of Ib, subgroups IIa and IIIa rats had significantly ($p<0.05$) increased RBC levels against IIb and IIIb rats respectively. This could be indication that rats placed on leaves of *C. albidum* from crude oil polluted area may have had a reduced rate of erythropoiesis (Red cell production) against those placed on leaves *C. albidum* from non-crude oil polluted area. Hence, the balance between the rate of erythropoiesis and destruction of the blood corpuscles in rats placed of leaves of *C. albidum* from crude oil polluted area may have been altered. The increase observed in Hb of subgroup IIIa rats was significant ($p<0.05$) against IIIb rats. PCV and its relationship with Hb was maintained in all the groups. However, only the observed increase in subgroups IIa and IIIa were significant ($p<0.05$) against IIb and IIIb subgroups respectively. Leucocytosis is directly proportional to the severity of the causative stress condition [17,25,27] and the severity may have contributed to a significant ($p<0.05$) increase in leucocyte mobilization observed in rats placed on leaves of *C. albidum* from crude oil polluted area (Ib, IIb, and IIIb) against those placed on leaves of *C. albidum* from non-crude oil polluted area in this study (Ia, IIa, and IIIb) respectively. MCHC, MCH and MCV are related to individual red blood cells while Hb, RBC and PCV are associated with the total population of red blood cells [26,43-44]. Levels of WBC, MCV, and MCH reduced significantly ($p<0.05$) in subgroups Ia, IIa and IIIa against Ib, IIb and IIIb subgroups. MCHC of rats in subgroups IIa and IIIa had reduced significantly ($p<0.05$) against those of subgroups IIb and IIIb. The significant effect ($p<0.05$) observed in MCV and MCH of rats placed on leaves of *C. albidum* from non-crude oil polluted area against those placed on leaves of *C. albidum* from crude oil polluted could be indication that incorporation of haemoglobin into red blood cells, the morphology and osmotic fragility of the red blood cells in rats placed on leaves of *C. albidum* from crude oil polluted area may have been altered.

The consequences of tissue damage is the release of specific enzymes associated with the affected tissue or organ into the circulation [45-46]. AST levels in rats placed on leaves of *C. albidum* from crude oil polluted site increased significantly ($p<0.05$) when compared to those of rats placed on leaves of *C. albidum* from non-crude oil pollute site. The same order was also followed in levels of ALT and ALP as observed.

Table 2. Haematological results of rats given *C. albidum* from non-crude oil polluted and crude oil polluted sites

	Group I		Group II		Group III	
Parameters	**Ia**	**Ib**	**IIa**	**IIb**	**IIIa**	**IIIb**
RBC($\times10^{12}$/L)	5.45±0.25	5.09±0.27	5.77±0.29	4.24±0.20*	6.17±0.90	4.43±0.14*
Hb (g/dl)	14.01±0.70	14.05±0.71	14.17±0.66	13.80±0.11	15.00±0.16	13.74±0.26*
PCV (%)	42.01±2.10	42.61±2.13	43.93±2.03	40.02±0.13*	46.80±1.10	39.98±0.17*
WBC($\times10^{9}$/L)	4.43±0.12	6.58±0.03*	5.14±0.33	7.29±0.42*	5.78±0.61	8.20±0.13*
MCV	77.08±0.19	83.71±1.01*	76.14±1.09	94.38±2.11*	75.85±3.02	90.24±2.16*
MCH	5.70±0.16	27.69±0.18*	24.56±0.20	32.55±1.19*	24.31±2.07	31.02±1.03*
MCHC	3.43±3.04	32.97±1.13	32.26±0.12	34.48±0.10*	32.05±0.19	34.37±0.18*

Results are presented as mean and standard deviation of triplicate determinations. Values of "b" subgroup asterisked against those of "a" subgroup under a main group on the table are statistically significant at p<0.05

Table 3. Liver enzyme studies of rats given *C. albidum* from non-crude oil polluted and crude oil polluted sites

	Group I		Group II		Group III	
Parameters	**Ia**	**Ib**	**IIa**	**IIb**	**IIIa**	**IIIb**
AST (U/L)	38.02±2.00	42.23±1.80*	42.40±0.85	46.1±0.17*	43.02±0.20	48.02±0.98*
ALT (U/L)	50.06±0.86	51.23±2.56	48.23±2.56	56.23±2.00*	54.38±1.10	62.69±3.13*
ALP (U/L)	12.99±0.65	19.79±0.99*	17.05±0.19	21.79±0.84*	15.79±2.08	24.04±1.20

Results are presented as mean and standard deviation of triplicate determinations. Values of "b" subgroup asterisked against those of "a" subgroup under a main group on the table are statistically significant at p<0.05

Table 4: Urea and creatinine levels of rats given leaves of *C. albidum* from non-crude oil polluted and crude oil polluted sites

	Group I		Group II		Group III	
Parameters	**Ia**	**Ib**	**IIa**	**IIb**	**IIIa**	**IIIb**
Creatinine (mg/dl)	0.65±0.08	0.98±0.01	0.61.±0.06	1.20±0.02*	0.48±1.10	1.21±0.03*
Urea(mg/dl)	46.00±0.10	56.03±0.80*	50.00±0.15	58.05±0.12*	67.13±0.23	72.47±0.18*

Results are presented as mean and standard deviation of triplicate determinations. Values of "b" subgroup asterisked against those of "a" subgroup under a main group on the table are statistically significant at p<0.05

ALT is a cytoplasmic enzyme found in very high concentration in the liver and an increase of this specific enzyme indicates hepatocellular damage, while AST is less specific than ALT as an indicator of liver function [47]. ALP has been associated with the prostrate [48].

Creatinine is the major catabolic products of the muscle and it is excreted in the kidneys [1]. Creatinine levels are used as indicator of renal failure [45-49]. The serum creatinine levels increased significantly ($p<0.05$) in rats placed on leaves of *C. albidum* from crude oil polluted area (IIb and IIIb) against those of rats placed on leaves of *C. albidum* from non-crude oil polluted area (IIa and IIIa) respectively. The observed significant increase could be indication of toxic nature of the leaves of *C.albidum* from crude oil polluted area in relation to the body. Urea levels increased significantly $p<0.05$) rats from all the subgroups placed on *C. albidum* from crude oil polluted site when respectively compared to rats from the subgroups placed on *C. albidum* from non-crude oil polluted area. Observed increased level of urea could be indication of azotaemia. High blood urea is associated with increased tissue protein catabolism, excess breakdown of blood protein and diminished excretion of urea [1, 45-49].

4. CONCLUSION

Leaves of *C. albidum* from crude oil polluted area contained high levels of heavy metals. The observed heavy metals become very important when their impacts are considered in a biological system. Rats placed leaves of *C. albidum* from crude oil polluted area showed marked clinical signs of fast degeneration against those placed on leaves of *C. albidum* from non-crude oil polluted area. These observed clinical signs could be linked to toxicity of the heavy metals found in the leaves of *C. albidum* from crude oil polluted area since leaves of *C. albidum* are known to have medicinal potency that could protect the integrity of internal organs and tissues. This study has evaluated the comparative effect of C*hrysophyllum albidum* medicinal plant from crude oil polluted and non-crude oil polluted areas on selected biochemical parameters in rats.

ETHICAL APPROVAL

As per international standard written ethical approval has been collected and preserved by the author(s).

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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