PRINT ISSN 1119-8362
Electronic ISSN 2659-1502

Full-text Available Online at
https://www.ajol.info/index.php/jasem
http://www.bioline.org.br/ja

J. Appl. Sci. Environ. Manage.
Vol. 26 (10) 1689-1694 October 2022

Effect of MethanolicExtract of *Justicia flava* Leaves on Biochemical Markers in Male Wistar Rats Fed Crude Oil Contaminated Feed

[1]ONAKURHEFE, P; [1]ONYEUKWU, OB; [2]OHWOKEVWO, OA; *[2]ACHUBA, FI

[1]Department of Chemical Sciences, Faculty of Sciences, University of Delta, Agbor, Delta State, Nigeria
*[*2]Department of Biochemistry, Faculty of Science, Delta State University, PMB 1, Abraka, Nigeria*

**Corresponding Author Email: achuba@delsu.edu.ng*
Co-Authors Email: patience.onakurhefe@unidel.edu.ng;benjamin.onyeukwu@unidel.edu.ng; nyore4real@rocketmail.com

ABSTRACT: The medicinal potentials of plants have been documented. This study evaluated the capacity of the leaf of *Justicia flava* methanolic extract (JFME) to alter the biochemical distortions initiated by feeding on diet containing crude oil. Male Wistar albino rats, thirty six, were constituted into nine groups. Each group had six rats. Group 1 had untreated feed. Groups 2 to 4 had untreated feed but were given 100 mg, 200 mg and 300 mg/ kg b.wt of JFME, respectively. Group 5 had untreated feed and given 200 mg/kg b.wt of ascorbic acid as standard. Group 6 was fed with diet containing crude oil (4ml/100g v/w). Groups 7 to 9 were given contaminated feed and 100 mg, 200 mg and 300 mg/ kg b.wt of JFME, respectively. The rats were maintained on these treatments for thirty days and had water *ad libitum.* Thereafter exposure period, lipid profile, hematological and inflammatory markers in the blood were analyzed using standard methods. Petroleum in feed altered the lipid profile, hematological and inflammatory markers compared to values in positive control rats. However, treatment of the rats with JFME had a positive reversal of these markers close to values in control rats; which compared favorably with ascorbic acid, used as standard. This investigation discovered JFME as a candidate for managing crude oil- imposed health issues.

DOI: **https://dx.doi.org/10.4314/jasem.v26i10.11**

Cite this paper as: ONAKURHEFE, P; ONYEUKWU, O. B; OHWOKEVWO, O. A; ACHUBA, F. I (2022). Effect of Methanolic Extract of *Justicia flava* Leaves on Biochemical Markers in Male Wistar Rats Fed Crude Oil Contaminated Feed. *J. Appl. Sci. Environ. Manage.* 26 (10) 1689-1694

Keywords: Contaminated feed; Crude oil; *Justicia flava;* Methanolic extract; Wistar Rats

Petroleum is of economic importance due to the contribution to many a nation gross domestic product (GDP) through its export to earn foreign exchange and creation of employment (Usman *et al.*, 2015). It is equally a source of numerous chemicals/ solvents that are produced via fractional distillation (Eneh, 2011). These are commodity chemicals required by industries for production of other goods and for domestic uses (Achuba and Okoh, 2014). However, this economic benefit does not come without negative implications on the health of humans in areas of production (Ordinioha and Brisibe, 2013). Hydrocarbons toxicities are preceded by oxidative stress due to petroleum-induced generation of free radicals. The free radical generated stimulates the peroxidation of membrane lipids (Achuba, 2010). Lipid peroxidation, though the outcome of oxidative insult on the biomembrane, is important in the assessment of the functional state of the cell (Achuba, 2002). It is applied in conjunction with antioxidant enzymes in measuring physiological regions of cells (Achuba and Okoro, 2010). These biomarkers are also utilized in environmental monitoring and assessment (Achuba *et al.*, 2014); hydrocarbon-induced physiological as well as histological distortion in animals (Achuba, 2018a). Most importantly, the responses of animals to the protective influence of plant–derived products are catching the attention of so many scientists (Achuba 2018; Ichipi-Ifukor *et al.*, 2019; Achuba and Ichipi-Ifukor, 2020; Onakurhefe *et al.*, 2020; Mordi *et al*., 2021). Similarly, the usefulness of organic substances in mitigating chemically-induced toxicity has been published: honey and coconut water (Mordi et al., 2015; Akintola *et al.*, 2018), *Hibiscus sabdariffa* (Dahiru *et al.*, 2003) and Moringa oil (Ezedom, 2018).On this premise, it is evident, therefore, that the

**Corresponding Author Email: achuba@delsu.edu.ng*

importance of organic- derived substances in the prevention of chemically–induced cellular and metabolic distortions cannot be overemphasized. The present investigation examined the function of JFME against physiological changes in Wistar albino rats induced by petroleum in diet.

MATERIALS AND METHODS

*Plant material:*The plant sample, *Justicia flava* was collected from Obiaruku, Delta State. A sample of the plant is kept at the herbarium at Plant Biology and Biotechnology, University of Benin, Benin City, Nigeria with a specimen number UBH$_J$0386 after due identification by Dr. H.A. Akinnibosun.

*Experimental Animals.*Fifty-four male Wistar rats (weight: 150-170 g) used were inbreed from the animal house unit of Anatomy Department, Delta State University, Abraka, Nigeria. They were housed in plastic cages and fed with fish feed and water *ad libitum* for one month in order to allow for acclimatization to the new environment and feed. The rats were maintained at 12-12 h light/dark of normal day/night cycle. The rats were handled based on the care of experimental animals as outlined by the national research council on the use and care of experimental animals (NRC, 2011).

Plant extract preparation. A sizeable quantity of the leaves was collected and dried at laboratory condition (28 ± 2ºC) for two weeks and four days until constant weight was obtained. The leaves were chopped from the stalk, broken into coarse sizes with hand and ground by means of a warring blender into fine powder form. It was then subjected to extraction using 70% methanol by cold maceration technique. The extract was concentrated with a rotary evaporator which produced brownish-green slurry at 40ºC.The slurry obtained was then dried in an open water bath at 40ºC.The yield was determined and the extract kept in a sample container at 4ºC until needed.

*Treatment of animals.*Male Wistar albino rats, thirty six were constituted into nine groups. Each group had six rats. Group 1 had untreated feed. Groups 2 to 4 had untreated feed but were given 100 mg, 200 mg and 300 mg/ kg b.wt of JFME, respectively. Group 5 had untreated feed and given 200 mg/kg b.wt of ascorbic acid as standard. Group 6 was fed with feed that contained crude oil (4ml/100g v/w). Groups 7 to 9 were fed with contaminated feed and were given 100 mg, 200 mg and 300 mg/ kg b.wt of JFME, respectively. The rats were maintained on these treatments for thirty days and had water *ad libitum.*

Blood collection and determination of inflammatory markers. The rats were subjected to overnight fasting after thirty days of treatment and sacrificed under chloroform anesthesia. Blood samples were collected by cardiac puncture into EDTA containers and plain containers, respectively. The blood in plain sample containers were allowed to stand for 2 h to enable it clot and subjected to centrifugation at 4000rpm for 10 min to allow for collection of various sera of each treatment. The collected sera were stored at −4˚C and used within 12 h for assay. The serum lipid profile (triglyceride, total cholesterol, HDL-Cholesterol and LDL-Cholesterol concentrations in serum) were determined using Randox, UK, assay kits. The methods of Westegren and Singer (1957) were adopted for the determination of ESR and C-reactive protein (C-RP), respectively. While ceruloplasmin and creatinine concentrations were determined using the methods of Sunderman and Nomoto(1970) and Lustgarten and Wenk (1972), respectively. The bloods collected in EDTA-containers were used for hematological analysis using automated Mindray Hematology analyzer, model, BC-2300.

Statistical Analysis: All data were analyzed using Analysis of variance (ANOVA) and expressed as means ± SD. Significant difference between the control and treatment means were set at $P < 0.05$ confidence.

RESULTS AND DISCUSSION

The consumption of feed containing crude oil altered the lipid profile of rats (Table 1). There are increases in total cholesterol and LDL-Cholesterol but a significant ($P<0.05$) decrease in HDL-cholesterol and triglyceride in rats fed crude contaminated feed against rats fed normal feed. On the contrary, the administration of JFME reversed these parameters close to the values in control rats and rats given the extracts but fed untreated feed. Lipid profile distortion is a major contributor in the initiation of cardiovascular disease (Einarson *et al*., 2018). The higher lipid level in plasma of rats fed diet containing crude oil (Table 1) noted here is in agreement with previous study (Achuba *et al.,* 2018). The abnormal high concentration of serum lipids in crude oil exposed rats is due to petroleum stimulated hypoglycemia (Achuba *et al.,* 2005). And in a bid to meet energy requirement there is increase in the mobilization of free fatty acids from the fatty depots in the body and hormone-activated lipolysis (Achuba*et al.,* 2005). The trend observed in the plasma lipid profile in the crude oil exposed rats was consistent as the plasma LDL-C was significantly higher relative to control (Table 1).

Table 1. Effect of administration of various doses of methanolic extract of *Justicia flava* leaf on serum lipid profile in male Wistar albino rats fed crude oil contaminated feed

Groups	TC(mg/dL)	HDL-C(mg/dL)	LDL-C(mg/dL)	TG(mg/dL)
GP1 Control	75.98 ± 7.49[a]	30.07 ± 2.755[a]	19.41 ± 2.60[a]	91.64 ± 6.02[a]
GP2 +100 mg/kg b wt JFME	72.74 ± 14.62[a]	60.24 ± 19.66[b]	18.88 ± 2.87[a]	91.79 ± 1.44[a]
GP3 +200 mg/kg b wt JFME	82.12 ± 4.56[b]	34.74 ± 2.35[a]	17.86 ± 2.16[a]	89.88 ± 2.43[a]
GP4+ 300 mg/kg b wt JFME	86.85 ± 4.17[b]	36.96 ± 1.76[c]	17.41 ± 2.62[a]	86.99 ± 2.90[b]
GP5+ Std AA	87.53 ± 6.36[b]	38.58 ± 1.67[c]	21.06 ± 2.44[b]	85.93 ± 1.59[b]
GP6 +CO	102.65 ± 6.53[c]	27.12 ± 2.30[d]	33.37 ± 4.72[c]	98.90 ± 2.40[c]
GP7 CO +100 mg/kg b wt JFME	92.79 ± 3.18[d]	32.07 ± 1.04[c]	31.71 ± 5.92[c]	94.37 ± 0.61[a]
GP8 +200 mg/kg b wt JFME	89.75 ± 2.30[b]	34.29 ± 1.14[c]	27.340 ± 5.47[d]	93.46 ± 0.96[a]
GP9 +300 mg/kg b wt JFME	89.73 ± 0.80[b]	35.323 ± 1.30[c]	25.91 ± 4.01[d]	92.05 ± 1.31[a]

TC= Total cholesterol; HDL-C=High Density Lipoprotein-Choleserol; LDL-C=Low Density lipoprotein-cholesterol; TG= Triglyceride Each datum is the mean of six determinations and expressed as mean± SD. Different superscripts in a column signifies significant difference at 5% confidence limit

Table 2. Effect of administration of various doses methanolic extracts of *Justicia flava* leaves on hematological indices in male Wistar albino rats fed crude oil contaminated feed.

Groups	PCV (%)	RBC(x10^6/µL)	Hb (g/dL)	MCV(fL)	MCH (pg)	MCHC(g/dL)	WBC(x10^3 /µL)
GP1 Control	39.20 ± 2.10[a]	7.20 ± 0.82[a]	13.77 ± 1.39[a]	48.96 ± 1.19[a]	22.33 ± 1.96[a]	44.32 ± 1.37[a]	8.03 ±0.61a
GP2 +100 mg/kg b wt JFME	43.94 ± 1.79[a]	8.35 ± 0.72[a]	15.79 ± 2.11[a]	49.70 ± 1.64[a]	23.49 ± 1.37[a]	45.25 ± 1.37[a]	8.50 ± 0.68a
GP3 +200 mg/kg b wt JFME	45.06 ± 1.43[b]	9.08 ± 0.38b	16.62 ± 1.21[b]	49.51 ±1.58[a]	25.88 ± 1.26[a]	45.51 ± 0.71[a]	8.53 ± 0.86a
GP4+ 300 mg/kg b wt JFME	46.08 ± 1.08[b]	9.09 ± 0.33[b]	17.61 ± 0.96[b]	49.91 ±1.678[a]	24.49 ± 1.79[a]	45.87 ± 1.13[a]	8.77 ± 0.93a
GP5+ Std AA	38.07 ± 1.54[a]	9.01 ± 0.91[b]	14.62 ± 1.26[a]	47.81 ±0.69[a]	24.10 ± 1.21[a]	45.30 ± 1.54[a]	7.99 ± 0.89a
GP6 +CO	31.51 ± 2.60[c]	5.39 ± 0.85[c]	10.38 ± 0.61[c]	43.995 ±1.34[b]	19.73 ± 1.19[b]	38.45 ± 1.97[b]	6.53 ± 0.83b
GP7 CO +100 mg/kg b wt JFME	34.64 ± 3.45[c]	5.77 ± 0.61[c]	12.39 ± 1.03[d]	47.638 ±0.86[a]	22.17 ± 1.63[a]	40.24 ± 1.64[c]	8.05 ± 0.15a
GP8 +200 mg/kg b wt JFME	37.00 ± 0.61[a]	6.10 ± 0.30[d]	13.39 ± 1.16[d]	47.688 ±1.01[a]	22.55 ± 1.63[a]	40.77 ± 1.65[c]	8.06 ± 0.19a
GP9 +300 mg/kg b wt JFME	37.16 ± 0.65[a]	6.02 ± 0.74[d]	12.85 ± 0.92[d]	47.845 ±1.09[a]	22.19 ± 1.85[a]	41.34 ± 1.55[c]	7.96 ± 0.19a

PCV= Packed cell volume; RBC= Red blood cell count; Hb= Hemoglobin content; MCV= Mean cell volume; MCH=Mean cell haemoglobin; MCHC= Mean cell haemoglobin concentration; WBC= White blood cell count. Each datum is the mean of six determinations and expressed as mean± SD. Different superscripts in a column signifies significant difference at 5% confidence limit

This finding is not unexpected as a high LDL-C has been reported to occur in blood of crude oil exposed rats (Achuba, 2005). The lowering of the plasma lipid profile in the control rats and rats treated with the extract is an indication of a hypolipidemic effect of the plant extract. The increase in plasma HDL-C in control rats treated with the extract is noteworthy as it did benefit crude oil intoxicated rats. This observation agrees with Achuba, (2005). Earlier publication indicated that high HDL-C protects against cardiovascular disease (Ganjali *et al*., 2017).This is predicated on phytochemicals that are shown to decrease blood lipid levels (Pires *et al.,* 2018; Onakurhefe *et al.,* 2019).This explains the positive modulation of the lipid profile by the plant extract. The consequence of consuming feed containing crude oil on hematological indices and the impact of the administration of various doses of methanolic extracts of *Justicia flava* in male Wistar albino rats are indicated in Table 2. The consumption of feed containing crude oil altered the hematological indices of male Wistar rats. This was exhibited by the significant ($P<0.05$) decreases in haemoglobin (Hb) content, red blood cell (RBC), packed cell volume (PCV), mean cell volume (MCV), Mean cell haemoglobin (MCH), mean cell haemoglobin concentration (MCHC) and white blood cell count(WBC). The administration of various doses of *Justicia flava* stimulated the increase in Hb, RBC, PCV, MCV, MCH and MCHC compared to the rats fed crude oil-contaminated diet only. Moreover, administration of various doses of *Justicia flava* significantly restored white blood cell count to values in rats fed with diet without crude oil. The modulations of these hematological indices are comparable to those of standard ascorbic acid given concurrently. Hematological profile is one index of the internal environment of animals which has been reported to be

predicated on nutritional composition and environmental milieu (Achuba *et al.,* 2018; Achuba, 2018c). The decrease in hematological profile, which is in tandem with previous study, has been attributed to a number of factors (Achuba, 2018c). One of the factors responsible for the reduced hemoglobin concentration, packed cell volume and red blood cell count is hydrocarbon-mediated decreased cell blood cell synthesis and increased hemolysis (Achuba, 2018c). However, the administration of *Justicia flava* leaves extract conferred protection against crude oil-mediated hematotoxic imports of crude oil. Hematoprotective propensity of *Justicia flava* is no surprise as plant materials with antioxidant potentials have been reported to protect animals from the hematotoxicity of crude oil (Achuba, 2019).

Table 3. Effect of administration of various doses methanolic extracts of *Justicia flava* leaves on inflammatory markers in male Wistar albino rats fed crude oil contaminated feed.

Groups	ESR (mm/hr)	CRP(mg/dL)	Creatinine (mg/dL)	Ceruloplasmin (mg/dL)
GP1 Control	2.04 ± 0.27^{a}	5.04 ± 1.04^{a}	1.84 ± 0.15^{a}	64.48 ± 1.79^{a}
GP2 +100 mg/kg b wt JFME	2.00 ± 0.28^{a}	5.00 ± 1.00^{a}	1.83 ± 0.20^{a}	61.08 ± 1.07^{b}
GP3 +200 mg/kg b wt JFME	1.85 ± 0.09^{a}	5.22 ± 0.69^{a}	1.86 ± 0.12^{a}	61.28 ± 2.66^{b}
GP4+ 300 mg/kg b wt JFME	1.98 ± 0.21^{a}	4.92 ± 0.36^{a}	1.85 ± 0.14^{a}	61.58 ± 0.74^{b}
GP5+ Std AA	2.09 ± 0.28^{a}	4.67 ± 0.52^{a}	1.86 ± 0.05^{a}	62.50 ± 1.69^{b}
GP6 +CO	2.41 ± 0.28^{b}	6.54 ± 1.07^{b}	2.47 ± 0.12^{b}	76.65 ± 5.10^{c}
GP7 CO +100 mg/kg b wt JFME	1.98 ± 0.03^{a}	5.32 ± 0.50^{c}	2.00 ± 0.15^{c}	68.35 ± 1.64^{d}
GP8 +200 mg/kg b wt JFME	2.02 ± 0.06^{a}	5.38 ± 0.49^{c}	1.99 ± 0.03^{c}	67.58 ± 1.99^{d}
GP9 +300 mg/kg b wt JFME	2.02 ± 0.18^{a}	5.28 ± 0.27^{c}	1.92 ± 0.11^{c}	66.58 ± 2.11d

Each datum is the mean of six determinations and expressed as mean± SD. Different superscripts in a column signifies significant difference at 5% confidence limit

The crude oil in rat feed altered the inflammatory indicators (Table 3). This was exhibited by the significant ($P<0.05$) increases in erythrocytes sedimentation rate (ESR), and the concentrations of C-reactive protein (CRP), creatinine and ceruloplasmin. The administration of various doses of *Justicia flava*stimulated decreases in ESR as well as the concentrations of CRP, creatinine and ceruloplasmin compared to the values in rats fed diet containing crude oil only. These modulations of these inflammatory indicators are comparable to those of standard ascorbic acid given concurrently. Some important indices of inflammation in animals include erythrocyte sedimentation rate, C-reactive protein, ceruloplamine and creatinine, and the relationship between inflammatory markers and crude oil intoxication had been documented (Achuba and Obaremi, 2018). The health promoting potentials of *Justicia flava* extract is expressed in the positive alterations of petroleum-induced changes in inflammatory markers. Similarly, the medicinal capability of *Justicia flava* leaves was reported earlier (Bafor*et al.,* 2019). The anti-inflammatory disposition of *Justicia flava* leaves extract is not out of place since induction of free radical production is one mechanism that accounts for the initiation of petroleum-stimulated tissue damages (Achuba, 2018d; Achuba, 2019). The generation of free radical is quenched by the plant extract due to its richness in antioxidants (Bafor *et al*., 2019). This explains the anti-inflammatory propensity of *Justicia flava*leaves extract because disease progression is associated with free radical generation cum oxidative stress.

*Conclusion:*This investigation revealed that JFME as a natural product for managing crude oil contaminated feed- imposed health issues. Thus, inhabitants of crude oil bearing areas of the world can take advantage of this plant that is readily available in the tropics.

*Acknowledgements:*The authors appreciate the kind assistance of Mr. Murphy Dumbiri Ogwumu for the technical support and Dr. Israel Okoro, who took the Plant sample to University of Benin, Benin City, Nigeria, for identification and documentation.

REFERENCES

Achuba, FI; Okoro, IO (2010). Role of antioxidants in the palatability and production potentials of water yam (*Diascorea alata poir*cv kurudu) *Elect. J. Environ. Agric. Food Chem.* 9(2):364-368.

Achuba, FI; Okoh, PN (2014). Effect of petroleum products on soil catalase and dehydrogenase activities. *Open J. Soil Sci.* (4):399-406.

Achuba, FI;Ebokaiwe, P; Peretiemo-Clarke, BO (2014). Effect of environmental pollution on oxidative stress in African catfish (*Clarias*

heterobranchus), *Int. J. Environ. Monitor Anal*. 2(6): 297-301.

Achuba, FI; Obaremi, C (2018). Effects of selenium fortified diet on inflammatory markers in Wistar albino rats exposed to crude oil. *Nig. J. Pharmaceut. Biomed. Res*. 3(3):209-216.

Achuba, FI;Peretiemo-Clarke, BO; Okolie, TC (2005). Oxidative stress in the brain of rabbits with petroleum-induced hypoglycaemia. *Biol. Lett.* 42 (1): 33-39.

Achuba, FI;Ubogu, LA; Ekute, BO (2018). *Moringa oleifera* treatment prevents crude oil tainted diet imposed toxicity in rats Sokoto *J. Med. Lab. Sci.* 3(3): 99 - 105.

Achuba, FI (2002). Superoxide dismutase and Lipid peroxidation levels in fish from the Ethiope River in Southern Nigeria. *Bull. Environ. Contam. Toxic.* 69(6): 892 – 899.

Achuba, FI (2005). Effect of vitamins C and E intake on blood lipid concentration, lipid peroxidation, superoxide dismutase and catalase activities in rabbit fed petroleum contaminated diet. *Pak. J. Nutr*. 4 (5): 330-335

Achuba, FI (2010). Spent engine oil mediated oxidative stress in cowpea (*Vigna unguiculata*) seedlings. *Elect. J. Environ. Agric. Food Chem*. 9(5): 910-917.

Achuba, FI (2018a). Role of bitter leaf (*Vernonia amygdalina*) extract in prevention of renal toxicity induced by crude petroleum contaminated diets in rats. *Intl. J. Vet. Sci. Med.* 6(2): 172–177

Achuba, FI (2018b). Protective Influence of *Elaesis guineensis* leaf in diet on petroleum-mediated kidney damage in rat. *Nig. J. Pharmaceut. Appl. Sci. Res.* 7(2): 33-38.

Achuba, FI (2018c). Effect of *Moringa oleifera* on crude oil mediated hematotoxicity in Wistar albino rats. *Nig. J. Pure Appl. Sci*. 31(2):3192-3196.

Achuba, FI (2018d). Powdered oil palm (*Elaesis guineensis* Jacq) leaf as remedy for hydrocarbon induced liver damage in rats. *Nig. J. Pharmaceut. Appl. Sci. Res.* 7(3):89-95.

Achuba, FI (2019). Protective r ole of Elaesis guineensis leaves against crude oil tainted diet–induced hematotoxicity in Wistar rats. *Iranian J. Toxicol*. 13(4): 1-4.

Achuba, FI; Ichipi-Ifukor, PC(2020) Protection effects of *Vernonia amygdalina* methanolic extracts against hepatocellular damage induced by petroleum contaminated diets in male rats*Iraqi J. Sci*. 61 (11): 2820-2830.

Akintola, AO; Kehinde, BD; Fakunle, JO; Ajayi, AF (2018). Synergistic and ameliorative effect of honey and coconut water on crude oil induced toxicity in rats. *Res. J. Environ. Toxicol*. 12: 24-33

Bafor, EE;Ukpebor, F; Omoruyi, O; Ochoyama, E; Odega, K (2019). Acute toxicological evaluations of the methanol leaf extract of *Justicia flava* (Vahl) Acanthaceae in mouse models. *Trop. J. Nat. Prod. Res*. 3(4):138-144.

Dahiru, D; Obi, OJ; Umaru, H (2003). Effect of Hibiscus sabdariffa calyx extract on carbon tetrachloride induced liver damage. *Biokemistri.* 15 (1): 27-33

Einarson, TR; Acs, A; Ludwig, C; Panton, UH (2018). Prevalence of cardiovascular disease in type 2 diabetes: A systematic literature review of scientific evidence from across the world in 2007-2017. *Cardiovasc. Diabetol.* 17(1): 83

Eneh, OC(2011). A review on petroleum: Source, uses, processing, products and the environment. *J. Appl. Sci.* 11: 2084-2091.

Ezedom, T (2018). Coconut (*Cocos nucifera*) and Moringa (*Moringa oleifera*) oils protect against cadmium-induced toxicity in albino rats. Trop. J. Nat. Prod. Res. 2(4):158-161. 2(4):158-161.

Ganjali, S; Dallinga-Thie, GM; Simental-Mendía, LE; Banach, M; Pirro, M; Sahebkar, A (2017). HDL functionality in type 1 diabetes. *Atherosclerosis.* 267: 99–109.

Ichipi-Ifukor, PC; Asagba, SO; Nwose, C (2019). Potentiating role of palm oil (*Elaeis guineensis*) and its extracts in cadmium-induced alteration of aminotransferases in albino rats. *Thai J. Pharmaceut. Sci*. 43(1):36-48.

Lustgarten, JA, Wenk, RE (1972). Simple, rapid, kinetic method for serum creatinine measurement. *Clin. Chem.* 18(11): 1419-1422.

Mordi, JC; Achuba FI; Ichipi-Ifukor, PC; Emete, G; Mokogwu, ATH; Nmanedu, AC; Aruoren, O; Ohwokevwo, OA (2021).Protective influence of *Costus afer* aqueous extract in rats fed with crude oil contaminated diet as measured by employing biochemical indices. *Iraqi J. Sci.* 62(12): 4639-4648.

Mordi, JC;Uzuegbu, UE; Opajobi, AO; Ojieh, AE (2015). Hepatoprotective effects of palm oil and coconut water in the serum of Wistar rats exposed to cadmium chloride contaminated diet. *Biokemistri* 27 (2) 79–84.

NRC (National Research Council) (2011). Guide for the care and use of laboratory animals. 8th ed. Institute of Laboratory Animal Resources, National Academy Press. pp 246.

Onakurhefe, P;Achuba, FI; George, BO(2020) Assessment of different regimen of oil palm leaf extracts against crude oil-adulterated feed mediated nephrotoxicity *Trop. J. Nat. Prod. Res.* 5(1): 199-204.

Onakurhefe, P;Achuba, FI; George, BO (2019). Phytochemical analysis and chemical characterization of extracts and blended mixture of palm oil Leaf. *Trop. J. Nat. Prod. Res*. 3(9):282-297.

Ordinioha, B; Brisibe, S (2013). The human health implications of crude oil spills in the Niger delta, Nigeria: An interpretation of published studies. *Nig. Med. J*. 54(1):10-16.

Pires, VA; Cardozo-Junior, EL; Ortmann, CF, Maraschin, JC; Favreto, WA; Donaduzzi, CM; Assreuy, J (2018). Lipid lowering and antiatherogenic effects of Vitex megapotamica (Spreng) Moldenke in a mice experimental model. *J. Ethnopharmacol*. 215:14–20.

Singer, JM;Plotz, CM; Pader, E; Elster, SK(1957). The latex-fixation test. III. Agglutination test for C-reactive protein and comparison with the capillary precipitin method. *Am. J. Clin. Pathol.* 28:611-617

Sunderman, FW;Nomoto, S (1970). Measurement of human ceruloplasmin by its p-phenylenediamine oxidase activity. Clin. Chem. 16(11):903-910.

Usman, A;Madu, I;Abdullahi, F (2015). Evidence of petroleum resources on Nigerian economic development (2000-2009). *Bus. Econ.* J. 6:149.

Westergren, A (1957). Diagnostic test: the erythrocyte sedimentation rate range and limitation of the technique. *Triangle*3(1): 20-25.