Full Length Research Paper

Effects of Nigeria Ekete light crude oil on plasma electrolytes, packed cell volume (PCV) and lipids profile in wistar (Rattus norvegicus) rats

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Hydrocarbon is known to alter blood parameters; some of these blood parameters may affect the activities of certain systems like cardiovascular system. Hydrocarbon gets into man and animal either through ingestion of contaminated food and water, bio-concentration through food chain, occupational exposure or by using hydrocarbon products. Effects of crude oil on plasma electrolytes, packed cell volume (PCV) and plasma lipid profile were examined in male Rattus norvegicus in this study. Crude oil was administered orally at dosages of 5 ml/kg (group I), 10 ml/kg (group II) and 20 ml/kg (group III) for two weeks while the control group received drinking water only. The result shows a reduction in packed cell volume in all treated groups compared to control (37.14±0.85, P < 0.05) and a significant (P < 0.05) increase in plasma sodium level in treated groups compared to control (22.486±2.983 mg/L) while there was no significant alteration in plasma lipids profile of the treated groups relative to the control. Oral administration of crude oil significantly altered the plasma levels of electrolytes and PCV.

Key words: Electrolytes, crude oil, plasma lipids profile, packed cell volume.

INTRODUCTION

Crude oil, the corner stone of Nigeria’s economy has been posing a threat through contamination of immediate environment during exploration and exploitation (Amadi et al., 1993). Hydrocarbons eventually get into man and animal through ingestion of contaminated food and water, bio-concentration through food chain, occupational exposure or by using hydrocarbon products (Amadi et al., 1993; Egborge, 1991). However, many people in some communities ingest crude oil directly as remedy for various conditions such as snake poisoning, convulsion, treatment of skin infection (Adesanya et al., 2009), gastrointestinal disturbances (Eyong et al., 2004) and arthritis. Crude oil has been shown to be well absorbed through all routes of contact such as dermal, oral or respiratory, because it is highly lipophilic (Bohlen et al., 1997). Impurities in crude oil include phenolic acid, napthelic acid and small quantities of most known elements except: sulphur, nitrogen, nickel, molybdenum which is present in relatively large quantities (Mason, 1966).

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Environmental pollutants are known to alter hematological parameters which in turn affect the functioning of vital organs like heart, blood vessels or the cardiovascular system as a whole by inducing; endothelial oxidative stress and vasodilator dysfunction (Podlutsky et al., 2010), dyslipidemia (Lemonine, 1911). Also increase in plasma electrolytes is known to contribute to incidence of hypertension.

The ingestion of crude petroleum contaminated diet imposed a reciprocal relationship between high-density lipoprotein (HDL)- cholesterol and low-density lipoprotein (LDL)- cholesterol in the plasma of rabbit and reduced blood glucose (Achuba, 2005; Ben-David et al., 2001). Presence of 12.5–50.0 ppm dispersed crude oil in solution prevented the development of high mucosal transfer rates in the ducklings given hypertonic saline drinking water (Crocketer et al. 1974). Alkindi et al. (1996), showed that water soluble fraction of Omani crude oil resulted in a progressive increase in plasma cortisol concentrations from 3 h onwards (rising from 18 to 51 ng ml^{-1} after 48-h exposure), increased plasma glucose concentration, did not affect plasma osmolality, sodium and chloride concentration but caused decrease in plasma potassium concentration in Flounders Pleuronectes flesus.

Analysis of osmo-regulation of the resident estuarine fish Atherinella brasiliensis 1^{st} month, 4^{th} month, and 7^{th} month after oil spill in Paranaguá Bay, Brazil, showed an increase in plasma osmolality (reaching ~525 mOsm/kg H_{2}O, or ~70% above values in reference fish) and chloride (reaching 214 mM or ~51% above values in reference fish) were detected 4 months after the spill; Plasma cortisol concentration increased progressively in samples from fish obtained 4^{th} month (462 ng/ml) and 7^{th} month (564 to 650 ng/ml) after the spill, compared to values in reference fish (192 ng/ml) (Souza-Bastos and Freire, 2011). Alonso-Alvarez et al. (2007), reported that gulls (laridae) fed with prestige fuel oil showed reduced glucose, inorganic phosphorus levels in plasma, and significantly reduced creatinine values. Glucose concentration was inversely related to Total Polycyclic Aromatic Hydrocarbon (TPAH) levels. Males gulls fed with fuel oil showed higher plasma activity of aspartate amino transferase (AST) than controls, and plasma activity of gamma-glutamyl transferase (GGT) was reduced. Shakirov, (2001).

Crude oil contains chemicals that readily penetrate cell walls, damage DNA, and alter the functions of the cells of any organ in the body (CDC, 1999; Achuba and Osakwe, 2004; EPA, 2010). The aims of this study were to examine the effects of oral administration of crude oil on plasma electrolytes, packed cell volume and plasma lipid profile in male R. norvegicus.

**MATERIALS AND METHODS**

Twenty eight male R. norvegicus of average weight of 150 g bought from animal farm from Ibadan were used for this experiment. They were housed in rat cages and acclimatized for two weeks before the commencement of the experiment. They were fed with commercially prepared rat diet and allowed access to water ad libitum throughout the period of the study. The rats were randomly divided to four groups (n=7). Crude oil was not administered to the control group but was given water as placebo. Rats in test received Nigerian Eket crude oil orally every day for two weeks: 5, 10 and 20 ml/kg respectively were given per rat in group I, group II and group III. Body weights were measured at the end of two weeks before collecting blood sample through peri-orbital sinus. Samples were collected into lithium heparinised sample bottles. They were centrifuged for lipid profile and electrolyte analysis. EDTA sample bottles were used to collect samples for packed cell volume analysis.

**Electrolytes analysis**

The electrolytes were analyzed for plasma calcium, potassium, and sodium concentration using Atomic absorption Spectrophotometer (Walsh, 1955).

**Digestion of the sample**

10 ml of Aqua radial (H_{2}SO_{4}/ HCL ratio 1:3) (H_{2}SO_{4}) was added to the sample inside 250 ml conical flask, the mixture was evaporated inside fume cupboard until brown fume disappear leaving white fumes. Then it was distilled and water was added up to 50 ml mark and filtered into sample bottle for analysis.

**Standard preparation**

The standard of calcium, sodium and potassium were prepared by dividing the molar mass of the compound of the elements by the molar mass of the elements, the product of the division is dissolved in 1litre of 5% nitric oxide, and this is stock standard (1000ppm). Serial dilution was used to calibrate the instrument. C1V1=C2V2.

**PCV analysis**

Packed cell volume was determined by micro-heamatocrit centrifugation (Jain, 1986).

**Lipid profile analysis**

Total cholesterol, HDL-cholesterol and triglyceride was determined through spectrophotometric method using , BIOLABO SA CHOD-PAP reagents (Ref 80106), BIOLABO S-HDL-CHOLESTEROL (PTA) and BIOLAB reagents – GPO respectively while LDL-cholesterol was calculated using this formular; (using the fried Waldes formula)

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LDL \text{ cholesterol} = \text{Total cholesterol} - \frac{\text{TG}}{2.2} + \text{HDL-cholesterol (mmol/L)}
\]

The analysis of variance (one way ANOVA) test was carried out using SPSS 15.0 to analyse the level of significant between the control group and treated groups. The level of significance for the ANOVA test was taken at \( P < 0.05 \).

**RESULTS**

Plasma electrolytes showed inconsistent variation in treated groups. There was significant increase in plasma Na⁺ in treated...
Figure 1. Changes in plasma electrolytes due to oral administration of crude oil. Asterisk (*) indicates significant difference that is P-value is <0.05.

Figure 2. Effects of oral administration of crude oil on packed cell volume (*) indicates significant difference that is P-value is <0.05.

groups compared to control (22.486±2.983) (P < 0.05) while there was no significant difference in plasma K⁺ and Ca²⁺ in treated groups compared to control as shown (Figure 1). The packed cell volume was significantly lower in all treated groups compared to control (37.143±0.857%) (Figure 2). Table 1 shows the effects of oral administration of crude oil on plasma lipid profile. The results show that there was no significant difference between the treated groups and control.

DISCUSSION

Exposure to hydrocarbon may vary from negligible persistent unconscious exposure to considerably large single doses (as in drinking as remedy). Effects of crude oil on plasma electrolytes, packed cell volume and plasma lipids profile were examined in this study. This study shows a consistent increase in plasma sodium level relative to the increase in dosage in treated groups while there was no significant difference in plasma level of calcium and potassium. Different authors have observed that crude oil alters plasma constituents, including plasma enzymes, electrolytes and amino acids. Findings in this study contradicted the observation of Alkindi et al. (1996), who showed that water soluble fraction of Omani crude oil did not affect plasma
osmolality, sodium and chloride concentration but caused decrease in plasma potassium concentration in *Flounders Pleuronectesflesus*. Gad (2011), observed an initial decrease in serum potassium and sodium and subsequent increase in potassium and sodium in *oreochromis niloticus* exposed to crude oil pollution.

Increase in plasma osmolality (reaching ~525 mOsm/kg H$_2$O, or ~70% above values in reference fish) and chloride (reaching 214 mM or ~51% above values in reference fish) were detected 4 months after crude oil spill in Paranaçu Bay, Brazil by Souza-Bastos and Freire, (2011). Alonso-Alvarez et al. (2007), fed gulls with Prestige oil; they reported that gulls fed with fuel oil showed reduction in glucose and inorganic phosphorous levels in plasma, as well as a trend to significantly reduced creatinine values. Shakirov (2001), reported decreases in the levels of K, Mg, and Ca and increases in the concentrations of Na and P in the red blood cells and in those of K, Mg, and P in the plasma while plasma Na and Ca were reduced in workers exposed constantly to hydrocarbon inpetroleum-refining industry for 3 to 5 years. Alteration in plasma electrolytes level is related to the development of hypertension (Moore, 1989).

The Packed cell volume (PCV) gave indication on haematopoietic activity taking place in the bone marrow. This study revealed significant reduction in packed cell volume of the treated groups, thus corroborating the previous findings on the effects of hydrocarbon on erythropoietin. It has been shown that hydrocarbon causes bone marrow hypoplasia, consequently reducing red blood cell and white blood cell formation (Eyong et al, 2004, Cody et al., 1981). Leighton (1990), reported haemolytic anaemia in mice given crude oil. Cody et al. (1981), reported heamatoletic disorder evidenced by a decrease in PCV and red blood count in rat exposed to 1, 3 dinitrobenzene. Hydrocarbon has been reported to contribute to hemolytic anemia by Eyong et al. (2004).

Though several authors have shown that crude oil ingestion causes abnormality in lipid profile, the results of this study shows that oral administration of crude oil did not alter lipid profile significantly. Achuba (2005) discovered reciprocal relationship between HDL-cholesterol and LDL-cholesterol in the plasma of rabbit fed with crude petroleum contaminated diet. Anigbogu and Ojo (2009), observed alteration in lipid profile of rats given different fractions of petroleum products. Epidemiological study has also shown that elevated concentrations of total cholesterol and LDL-cholesterol in the blood are powerful factor for coronary heart disease (Lawn, 1999). Dyslipidemia is known to be co-risk factor for development of hypertension along with obesity and diabetes, by causing endothelia dysfunction; a dysfunctional endothelium will express impaired nitric oxide production activity as well as alteration in endothelin I and endothelin A and B receptors expression (Ruben et al., 2006).

### Conflict of interests

The authors did not declare any conflict of interest.

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