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Effect of Exposure to Lead and Cadmium in Drinking Water Sourced from Nigeria's Niger Delta Region on Serum Enzymes and Eletrolytes of Albino Wistar Rats

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Abstract
Drinking water randomly collected from various sources in Nigeria's Niger Delta region were assessed for lead (Pd) and cadmium (Cd) contents. Groups of albino Wistar rats were exposed to these water samples *ad libitum* as drinking water for a duration of 28 days, following which levels of three(3) serum enzymes (aspartate aminotransferase- **AST**, alanine aminotransferase- **ALT** and alkaline phosphatase- **ALP**) and electrolytes (Na^+, K^+ and CI) were investigated using standard analytical methods. The results indicated that Pd (0.105-0.142 mg/L) and Cd (0.015-0.024mg/L) detected in the water samples caused significant increases ($P<0.05$) in AST (102.92 $\pm$ 0.23 U/L), ALT (96.63$\pm$ 0.20 U/L) and ALP (83.86$\pm$ 0.35 U/L), but there were significant decrease ($P<0.05$) in Na^+ (102.72 $\pm$ 0.75 mmol/L), CI^- (39.38 + 0.55 mmol/L) and increase ($P<0.05$ in K^+ (5.18 $\pm$ 0.40 mmol/L) in the albino Wistar rats. These results suggest possible toxicity on the liver and kidney.
Keywords: Lead, cadmium, drinking water, serum enzymes and electrolytes

Introduction
The road to achieving the Millennium Development Goal (MDG) for water (to reduce to half by 2015 the number of people globally who do not enjoy sustainable access to safe drinking water), can only be built on improvement to water access (WHO, 2011a). The health benefits of drinking safe, clean water to meet the requirements in human, especially to maintain the proper functioning of the body system, cannot be over-emphasized. Heavy metals like Pd and Cd in drinking water are known to cause liver and kidney damage (WHO, 2011b). Chronic exposure to contaminated drinking water with these heavy metals can result in the development of various toxic effects and adverse human health (Mudgal et al; 2010). Cadmium poisoning has been associated with kidney disease, hypertension, and deformation of reproductive tract as well as endocrine system (Jomova and Valko, 2010). Long term exposure to lead in drinking water may lead to morphopathological changes in the kidney (Szyczewski et al; 2009). Cadmium may interfere with metallothionein's ability to regulate zn and Cu concentration in the body that some patients showed some elevation in Zn in their urine samples. Metallothionein is the protein that binds to excess essential metals to render them unavailable (Hanaa et al; 2000).When Cd induces metallothionein inactivity, it binds to Cu and Zn disrupting the homeostasis level (Hanaa et al; 2000). This research was undertaken to assess the effect of exposure to Pd and Cd in drinking water sourced from Nigeria's Niger Delta region on serum enzymes and electrolytes of albino Wistar rats.

Materials and methods
Collection of water samples
Borehole and surface water samples were obtained from selected zones in five states, namely, Akwa Ibom, Bayelsa, Cross River, Delta and Rivers. Random collection covered industrial zones waste dump sites, Villages, urban and semi-urban centers. Polyethylene terephthalate (PET) sterile bottles of 150ml volume capacity were used for sample collection. The PET bottles were sterilized by soaking them in 10% w/v chlorine solution for 34 hours and pre-numbered for easy sample collection. Water from borehole was allowed to run for a while before filling the pre-coded sterile 150ml sample bottles. The bottles filled with water samples were well-stoppered and put in a cooler for easy transportation from the collection site to the laboratory for analysis. Pd and Cd concentration were determined by the method of spectrophotometry as described by Tietz (2008).

Animals, grouping and experimental protocol
Adult albino Wistar rats of both sexes (210-240g) were obtained from the disease –free stock of the animal house of Biochemistry department, University of Calabar, Nigeria. The animals were housed in standard cages, placed in a well-ventilated animal room under standard conditions of temperature (28 ± 2^oC) and relative humidity ($46\pm5\%$), and were acclimatized for two weeks. the animals were maintained on rat chow (obtained from Pfizer Livestock Feeds, Lagos, Nigeria), which was provided with water *ad libitum* throughout the duration of the work, permission for use of animals and animal protocols in the present study was obtained from the

College of Medical Sciences Animal Ethics Committee, University of Calabar prior to experimentation.
The animals were assigned into three study groups of twelve rats each and exposed directly to the Pd and Cd containing water samples in drinking water bottles as follows:
Group 1 (control): Animals were allowed access to distilled water and feed only.
Group 11: Animals were exposed to Pd-contaminated water as obtained from the natural source along with feed
Group 111: Animals were exposed to Cd-contaminated water as obtained from the natural source along with feed.
The exposure to water lasted for a 28-day period.

Collection of blood samples and biochemical analysis
Twenty for (24) hours after expiration of the time of exposure, the experimental animals were fasted overnight, anaesthetized under chloroform vapour and dissected. Blood samples were collected by cardiac puncture using syringes and needles transferred into plain sample bottle and allowed to clot for one hour at room temperature. Blood serum was prepared by centrifuging at 2000 rpm for 30 minutes. The blood serum obtained was used for biochemical assays.
Serum enzymes (AST and ALT) were assayed by the method of Rietman and Frankel (1957) as employed in the Agappe Diagnostic kit, while ALP activity was determined using the kinetic colorimetric method of optimized Deutxch Gesellschaft fu Klinische Chemie (DGKC, 1972).
Serum electrolytes were determined by the colorimetric method by Trinder (1951) for sodium, turbidimetric method for potassium and modified thiocyanate method for chloride as employed by Agappe Daignostic kit method.

Result and Discussion
The effect of exposure of the albino Wistar rats to Pd and Cd in drinking water on serum enzymes and electrolytes are as presented in Table 1.

Table 1: Effect of exposure to Pd and Cd in drinking water on serum enzymes and electrolytes

Group	AST (U/L)	ALT (U/L)	Alp (U/L)	Na^+ (U/L)	K^+ (mmol/L)	CT (mmol/L)
control	98.77±0.13	73.82±0.19	75.74±0.26	149.59±0.58	4.64±0.26	93.39±0.47
Pd-treated	102.92±0.25*	96.63±0.20*	83.86±0.35*	102.72±0.75	5.18±0.40	39.38±0.55*
Cd-treated	102.32±0.33*	109.33±0.29*	132.28±0.28*	114.11±0.37*	4.92±0.25*	67.46±0.38*

Values are expressed as the mean± SEM: n=3
*Significantly different from control at P<0.05

There was significant (P<0.05) increase in serum AST and ALP activities following exposure of albino Wistar rats to Pd and Cd in drinking water, while the serum electrolyte (mmol/L) levels showed significant (P<0.05) increase in AST and ALT activities. This may be an indication of hepatotoxicity (Goel and sood, 1989). Increased activity of serum ALP is usually related to mild biliary obstruction and is a primary indicator of space occupying hepatic lesions. Most enzymes reside and function within the cell. The presence of enzymes under normal circumstances in the blood is the result of turnover of cells or leakage of enzymes from the cells.The observed significant increase in liver and kidney weights (see Table 11), from the albino Wistar rats exposed to Pd and Cd in drinking water further confirm toxicity, since increase or decrease in relative weight of an organ is an indication of toxicity (Meisenberg and Simmons, 1998.). Both liver and kidney weights were significantly (P<0.05) increased,

Table 11: Relative organ weights of albino Wistar rats exposed to Pd and Cd in drinking water

Group	Liver (g)	Kidney (g)
Control	8.19±0.02	1.28±0.01
Pd-treated	10.35±0.15*	1.78±0.03*
Cd-treated	13.15±0.51*	2.85±0.04*

Values are expressed as the mean ± SEM: n=3
* Significant different from control at P<0.05

The increase in weight of liver and kidney were indication of toxicity. Exposure of albino Wistar rats to Pd and Cd in drinking water also resulted in the impairment of renal functions as evidenced by the significant reduction in serum Na^+ and CI^- levels, while there was an increase in K^+ levels. The kidneys are involved in excretion, homeostatic and endocrine regulations. They integrate the internal milieu (Stacey and Mitchell, 2008). The major extracellular ions are Na^+, CI^- and $HCO3^-$. In intracellular fluids, the main ions are k+, $Mg2^+$, organic

phosphates and proteins (Stacey and Mitchell, 2008). The unequal distribution of ions is the result of an active transport of Na+ from the inside to the outside of the cell against an electrochemical gradient. The process requires energy supplied by the glycolytic processes in the cell. The estimation of CI- is important in the differential diagnosis of acid-base disturbances. Na^+, K^+ and CI^- are common chemicals found in small quantities in most waters, and these element play a key role (as electrically charged minerals) in body metabolism by helping to move nutrients into and waste out of the body's cells, maintain healthy water balance and stabilize the body acid level (Stacey and Mitchell, 2008).

Conclusion

The presence of high levels of Pd and Cd in the drinking water samples which lead to the elevated levels of serum enzymes (AST, ALT and ALP) in albino Wistar rats, with increased levels of serum electrolytes like K^+ and decreased levels of Na^+ and CI^-, all of which suggest possible toxicity on the kidney, render the samples unsuitable for human consumption.

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