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Assessment of the Antioxidant Potential of *Hypoestes rosea* Leaf in Lead-acetate-induced Albino Rats

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Authors' contributions

This work was carried out in collaboration among all authors. Author ESB designed and supervised the study, author FKU wrote the protocol, the first draft of the manuscript, the literature and performed the analyses. Authors EON and FI supervised the work. All authors read and approved the final manuscript.

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ABSTRACT

Hypoestes rosea has been used as a traditional medicine in the Niger delta for dysfunction of the endocrine system. However, there has been no known study on the effects of *hypoestes rosea* on oxidative stress. In this study we evaluated the effect of aqueous extract of *Hypoestes rosea* (AEHR) leaf on oxidative stress markers of lead acetate induced male and female albino rats at acute and sub-chronic stages in pre-treatment and post-treatment phases. Animals were divided into 17 groups of five each for both sexes in the treatment groups, while the positive control group had 10 animals in each sex. 8 groups were for the acute phase of the study for 21 days in each sex, while 8 were for 35 days for the sub chronic stage of the study. Negative Control (NC) group received rat feed only, Experimental (EC) group received 100 mg/kg bwt/day for 21 days at acute and 35 days for sub chronic. Positive Control (PC) group received 60mg/kg b.wt per day of lead

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acetate for 35 days. The other 3 groups received 100 mgkg, 200 mg/kg and 300 mg/kg b. wt respectively for 14 and 28 days either as pre treatment or post treatment, for both sexes of the albino rats. Samples were taken at the end of the study period through the jugular vein under chloroform anaesthesia. Results showed lead acetate induced oxidative stress in the rats, evidenced by the significantly decreased ($p < 0.05$) Superoxide Dismutase (SOD) and Total Antioxidant Capacity (TAC) between the NC and PC groups. The plant in a dose dependent pattern was able to significantly ($p < 0.05$), reverse the effect of lead acetate in the Post and pre treatment phases. Our study also shows that dose dependent AEHR extract significantly reduced the impact of lead in oxidative stress markers. In conclusion, consumption of AEHR by albino rats could help protect against lead acetate induced oxidative stress.

Keywords: Antioxidants; Hypoestes rosea; lead-acetate; albino rats.

1. INTRODUCTION

Heavy metals toxicity is a major threat to public health throughout the world. Considerably, Lead exposure is clearly common and capable of inducing permanent damage to various organs. Several evidence has shown the risk of lead toxicity on male reproductive system [1]. Lead is one of the most hazardous metals to living matter [1]. Lead is highly toxic and can interrupt the body's biologic, neurologic and cognitive function. Children are particularly susceptible [1].

Lead acetate is a biotoxic environmental and industrial pollutant, which accumulates in almost all the body tissues such as the liver, lung, bones, kidneys, reproductive organs and immune system [2]. Studies have reported about the physiological, biochemical, and behavioral effects of this toxic lead in animal, including disorders of central and peripheral nervous systems, [3], cardiovascular system [4], kidney [5], liver [6] and reproductive system [7].

The mechanism of lead-induced testicular toxicity is the oxidative stress and it develops when there is an imbalance between the free radicals and the scavenging capacity of antioxidants in the testis [8].

Reactive oxygen species are metabolites of oxygen including superoxide anions, hydrogen peroxide, hydroxyl radical and nitric oxide. When presenting in excess they can initiate pathological damage by inducing oxidative changes to cellular lipids, proteins and deoxyribonucleic acid (DNA) [9]. Most cells possess physiological antioxidant systems that serve as defenses to these reactive oxygen species, when this system is overwhelmed cellular function is affected. In males, significant increase of superoxide anion and free radical activity has been demonstrated in some andrological conditions such as leucocyto-spermia and varicocele where great oxidative stress produce different unstable potentially toxic products [10].

Oxidative stress has been identified to play a key role in the pathogenesis of subfertility in both males and females [11]. Oxidative stress can also arise from some environmental pollutants which can destroy biomolecules and cell structures of lipids [12]. This will make the appropriate antioxidant lose its scavenging potentials to mop up the free radicals at the cellular levels [13].

Despite a lack of medical evidence to support their therapeutic efficacy and toxicological effects, the use of herbal medicine has increased considerably. According to World Health Organization (WHO), up to 80% of the world's population in underdeveloped and developing countries rely on traditional medicine practices for their primary health care needs [14]. Traditional medicines have been accorded greater acceptance in Africa because of the unavailability, unwanted side effects and high costs associated with orthodox medicines, inadequate health facilities and healthcare professionals, coupled with inadequate training of health workers [15].

These medicines are known to be efficacious in the treatment of several ailments, ranging from minor ailments to diseases that affect the organs and systems. Their importance is based on their presumed use in folklore. Plants with natural medicinal constituents are seen as potent and safe. A medicinal plant is any plant which, in 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 [16].

Hypoestes rosea is an erect herb standing a little above 1 m high, flowering periodically, it has been in use in the Niger delta for the treatment and local management of infertility in rural areas for years. Specific physiological activity of *Hypoestes rosea* as a herbal remedy in fertility treatment in relationship to its effect on the fertility hormones and oxidative stress has not been fully studied.

Some studies have found lead to be a cause of reproductive toxicity in both men and women. In men, it causes a reduction in libido, infertility, as well as a reduction in sperm count and vigor, while in women there is an increase in the incidence of stillbirth and miscarriage [17]. The aim of this study therefore was to assess the antioxidant potential of aqueous extract of *Hypoestes rosea leave* on oxidative stress of lead-acetate induced albino rats.

2. MATERIALS AND METHODS

2.1 Plant Collection, Identification, Extraction and Preparation

Fresh leaves of *Hypoestes rosea* were collected from Sime in Tai (4°42' 59.99"N. 7°17' 60.00"E) Local Government Area of Rivers State in Nigeria in November 2018. They were deposited at the forest herbarium of the Forestry Research Institute of Nigeria, Ibadan where it was identified by Dr Osiyemi Seun as *Hypoestes rosea* Beauv, with a Herbarium number of FHI 112295.

2.1.1 Preparation of aqueous extracts *Hypoestes rosea* leaf

Extract was prepared using the method of Janardan, 2008. The identified leaves were air dried in a room away from sunlight. It was ground using a blender. The pulverized powdered material was macerated with distilled water in a maceration jar for twenty four hours. During this period of maceration, the contents are well agitated. They are subsequently filtered with whatman No 1 filter paper severally until a very clear filtrate is obtained. The filtrate is transferred to an evaporating dish, which is then poured into a tall column. Cold water is added until the powdered material is completely immersed. It is allowed to stand for 24 hrs so that water-soluble ingredients attain equilibrium in the water. The enriched aqueous extract is concentrated in multiple-effect evaporators until it becomes completely dry. The dry extract is weighed, kept in the fridge until use.

2.2 Calculation of Dose of *Hypoestes rosea* Leaf for Administration

The aqueous extract for the experimental animals was prepared according to the organiza-tion of economic corporation and developments guideline [18], using the calculations based on the method of [18].

The vehicle for the dissolution of the extract for administration was distilled water [19].

Calculations

A uniform 1 ml was used for all animals.

Dosage in mg = {Body weight of animal (g)} x dose (mg)/ 1000

For a rat of 120 g receiving 100 g/kg body weight

$$= \frac{120}{1000} \times 100 = 12 \text{ mg/ml.}$$

2.3 Reagent Acquisition and Preparation

Lead acetate 99.5% purity for this research was bought from Tianjin Kermel chemical reagent co. ltd, China – 022-28545263 through their agent in Nigeria Hysec Services. It was confirmed to be pure lead acetate by the Chemistry department of the Rivers State University.

The reagents for the analysis of the reproductive hormones were imported from Elabscience Biotechnology incorporated USA, Monobind Incorporated USA and Perfemed Incorporated USA.

2.4 Experimental Animals

A total number of one hundred and eighty albino rats made up of ninety male rats and ninety female rats with an average weight 150-180 g were procured for the research work. All animals were procured from the Animal House Physiology department of the Faculty of Basic Medical Science of the University of Port Harcourt. The animals were kept in a well-ventilated cage, where they were fed with growers mash. Rats were allowed free access to feed and water *ad libitum.* They were divided into their different groups and allowed to acclimatize for two weeks.

All animals were handled in conformity with the conditions outlined by the National Academy of Science [20-22].

2.5 Experimental Design

2.5.1 Grouping and Treatment

A total of 180 rats previously acclimatized for two weeks weighing between 150-180 g rats equal in both sexes of 90 each were divided into 17 groups comprising of 5 rats in each group except the positive control group that had 10 rats. The process of the experiment involved induction of some rats with 60 mg/kg body weight of lead acetate for 7days to alter their hormones and subsequent treatment with 3 different doses of *Hypoestes rosea* by oral gavage for acute and sub chronic stages for the pre-treatment phase, while post treatment phase had treatment with the extract for a period of time, (14 days for acute stage and 28 days for sub chronic stage), then subsequent induction with lead acetate for 7 days for both sexes.

The group that was used as negative control had the normal rat feed only. Positive control received lead acetate only, extract control received 100 mg of the extract only. The 3 doses for the treatment groups were 100 mg/kg body weight, 200 mg/kg body weight and 300 mg/kg body weight of the extract respectively as pre-treatment, post-treatment for acute and sub chronic stages by oral gavage. In the two stages of the experiment the positive control rats were given 60 mg/kg body weight of lead acetate for 7 days, were fasted overnight and sacrificed on the 8th day, while all others in the pre-treatment started their different doses of extract on the 8th day and continued until the 21st day when they were fasted overnight and sacrificed on the 22nd day while the rats for the sub-chronic phase continued on the extract until the 35th day when they were fasted overnight and sacrificed on the 36th day.

The post treatment group for the acute phase had their varying doses of the extract from day 1 till day 14, when they were commenced on 60 mg/kg body weight of lead acetate up to the 21st day when they were fasted and sacrificed. The sub-chronic group continued with the extract only until the 28th day when they were treated with 60 mg/kg body weight of lead acetate until the 35th day when they were fasted overnight and sacrificed on the 36th day. Euthanasia was under diethyl ether anesthesia. On sacrifice, blood was taken from the jugular vein for oxidative stress markers into lithium heparin bottles. The blood for oxidative stress markers was spurn at 3000 rpm for 10 mins in a Wisperfuge centrifuge (Model 1384).

2.6 Experimental Analysis

2.6.1 Quantitative determination of Total Antioxidant Capacity (TAC)

Method: Colorimetric. Catalog Number: E-BC-K136.

Principle: A variety of antioxidant macro molecules, antioxidant molecules and enzymes in a system can eliminate all kinds of reactive oxygen species and prevent oxidative stress induced by reactive species. The total levels affect the total antioxidant capacity in the system. Many antioxidants in the body can reduce Fe^{3+} to Fe^{2+} can form stable complexes with phenanthroline substance. The antioxidant capacity (T-AOC) can be calculated by measuring the absorbance at 520 nm.

Calculation of Results for plasma samples: Definition: At 37°C, the OD value of the reaction system was increased 0.01 by per milliliter of serum (plasma) sample per minutes is defined as a unit of total antioxidant capacity.

T-AOC activity *(U/mL)*

$$= \frac{OD_{sample} - OD_{control}}{0.01} / 30(min) \times \frac{\text{Total volume of reaction system (mL)}}{\text{The volume of sample (mL) Before tested}} \times \text{Dilution factor of sample}$$

OD= Optical Density

2.6.2 Quantitative determination of total-Superoxide Dismutase (SOD) (Hydroxylamine method)

Method: Colorimetric. Catalog Number: E-BC-K019

Principle: The superoxide anion free radical (O_2) can be produced by xanthine and xanthine oxidase reaction system, O_2-oxidize hydroxylamine to form nitrite, it turns to purple under the reaction of

developer. When the measured samples containing SOD, the SOD can specifically inhibit superoxide anion free radical (O_2-) the inhibitory effect of SO can reduce the formation of nitrite, the absorbance value of sample tube is lower than control than control tube. Calculate the SOD of sample according to the computational formula.

Calculation of Results:

For serum (plasma), culture cell and other liquid samples:

The amount of SOD when the inhibition ratio reaches 50% in mL reaction solution is defined as 1 SOD activity unit (U).

SOD activity *(U/mL)* =

$$\frac{OD_{control}-OD_{sample}}{OD_{control}} \div 50\% \times \text{Dilution multiple of reaction system} \times \text{Dilution multiple reaction before tested}$$

OD= Optical Density

2.7 Quality Control Measures

External quality control sera were assayed along with the parameters analysed. Standard operating procedures were duly adhered to while carrying out the analysis. Good laboratory practices were observed while conducting the test. The machines were checked and calibrated before use.

2.8 Statistical Analysis

The statistical software used for the analysis and graphics presentation was the Statistical Analysis System (SAS), STAT 15.1, developed by SAS Institute, North Carolina State University, USA. Data are presented as Means ± SEM, comparison of mean values of groups that are more than two was done using analysis of variance (ANOVA), and the Turkey test of multiple comparison was used to test for variance within and across groups. Variation between two groups was done using the Student t-test analysis. P values less than 0.05 were considered statistically significant.

3. RESULTS AND DISCUSSION

The analysis of the acute effect of various concentrations of *hypoestes rosea* on the oxidative stress parameters superoxide dismutase (SOD) and Total antioxidant capacity (TAC) in the Albino rats by sex, treatment phases, and experimental groups are shown Tables 1-8.

3.1 Effect of Aqueous Extract of *Hypoestes rosea* on Oxidative Stress Markers

The parameters used in the assessment of the effect of the aqueous extract of *hypoestes rosea* leaf on the reproductive hormones are superoxide dismutase and total antioxidant capacity. These parameters were used to analyze the oxidative stress level in all the sexes, phases and stages of this study. Oxidative stress occurs when the production of potentially destructive reactive oxygen species (ROS) exceeds the body's own natural antioxidant defenses, resulting in cellular damage [23].

Total antioxidant capacity and superoxide dismutase are key measurements of cellular injury causing generation of reactive oxygen species.

3.2 Total Antioxidant Capacity

Total antioxidant capacity provides an estimation of antioxidant capacity which includes those antioxidants not yet recognized or not easily measured and represents the overall free radical scavenging ability of various antioxidants [24]. It is a useful measurement for investigating oxidative stress, which has been implicated in the pathological mechanisms of many diseases [25]. It has the advantage of measuring not just the antioxidant capacity of a single compound, but the antioxidant capacity of all antioxidants in a biological sample [26].

The result of analysis of total antioxidant capacity of the albino rats in this study shows that the administration of lead acetate on the albino rats in sexes, treatment phases and stages of the study caused a significant drop in the total antioxidant capacity of the albino rats. The PC group had a very low mean level of total antioxidant capacity. Lead is reputed to bring about damage in tissues by inducing oxidative stress, thereby raising the reactive oxygen species in the body and also depleting anti-oxidant enzymes [27]. It has the

ability of increasing the production of reactive oxygen species (ROS) [7,28]. This finding also agrees with [29,30]. The ability of lead to increase lipid peroxidation causes the generation of ROS, and inhibits the activity of antioxidants such as glutathione peroxidase (GPx), superoxide dismutase (SOD) and catalase. The depletion of these collective antioxidants as a result of the increase lipid peroxidation causes a significant decrease in the total antioxidant capacity.

The Acute pre-treatment phase of the study had a remarkable raised level of total antioxidant capacity of all rats that were treated with aqueous extract of *hypoestes rosea* leaf after initial induction with lead acetate. The values of all the treatment groups (100 mg/kg, 200 mg/kg and 300 mg/kg) increased significantly above the Positive control group and the normal control group. In this study we noticed despite the duration of the acute exposure, the aqueous leaf extract was able to reverse the cellular injury caused by the introduction of lead acetate, through its activity of lipid peroxidation. This is confirmed as the rise in the total antioxidant capacity, showing a reversal in the antioxidant status. The same result showed in the acute post treatment stage of the study as the treatment with aqueous extract of *hypoestes rosea* leaf is able to protect the albino rats from oxidative damage. This is in agreement with earlier findings of Van and Hekimi [31] who in their findings showed the seeds of *Garcinia kola* possess antilipoperoxidative effect, a proof of its antioxidative property inhibiting lipid peroxidation as seen in the acute pre-treatment and post-treatment groups.

Pre-treatment and Post-treatment of the albino rats with lead acetate and aqueous extract *hypoestes rosea* leaf (100 mg/kg, 200 mg/kg, 300 mg/kg) for both sexes in the sub-chronic phase of the study caused a mark increase in the total antioxidant capacity, which is significantly different PC group whose total antioxidant capacity was quite low, the results showed that the treated albino rats even had higher values than the NC group. The values of the total antioxidant capacity also increased as the concentration of the aqueous leaf extract increased. This suggests that the leave extract of *hypoestes rosea* may have protective effect against lead acetate induced oxidative stress as seen in the AEHR 100, AEHR 200 and AEHR 300 groups. Antioxidants form protection in the body against lipid perodixation, this action subsequently protects the body cells and tissues against dysfunction that could lead to disease [26]. The increase in antioxidant activity in the cell as a result of release of free radicals causes a rise in the total antioxidant capacity, which is a reliable biomarker for diagnostic and prognostic evaluations [26]. The increase in the total antioxidant capacity shows an ability of the aqueous extract of *hypoestes rosea* leaf to prevent the oxidation of other molecule in a tissue, scavenge free radicals and attenuate deleterious effects. T his finding is in agreement with previous studies which shows medicinal plants have antioxidant properties in reducing free radicals. Our findings in this study is also in agreement with [27] which shows that treatment with *Curcuma* which is a member of the family *Zingiberaceae* protects against lead induced toxicity, by minimizing the toxic effect of lead acetate via its antioxidant activity. The antioxidant protective mechanism decreases the oxidative stress and scavenges the free radical responsible for cellular damage and thus inhibits the lipid peroxidation.

Table 1. Antioxidant parameter (SOD and TAC) activities of lead acetate induced female albino rats post-treated with *Hypoestes rosea* in the acute phase

Experimental Group	SOD (U/ml)	TAC (U/ml)
	Mean ± SEM	
EC	318.58±0.065#	5.526±0.218#
NC	266.14±0.081#	2.070±0.143
PC	125.38±0.085	1.200±0.172
AEHR (100 mg/kg)	300.92±0.062#	2.470±0.181#
AEHR (200 mg/kg)	320.46±0.101#	4.970±0.295#
AEHR (300 mg/kg)	329.98±0.047#	5.210±0.161#
P-Value	<0.001	<0.001
F-Value	84.738	86.566

 # - significant at p < 0.05 when compared with PC

Table 2. Antioxidant parameter (SOD and TAC) activities of lead acetate induced female albino rats pre-treated with *Hypoestes rosea* in the acute phase

Experimental Group	SOD (U/ml)	TAC (U/ml)
	Mean ± SEM	
EC	318.59±0.065#	5.526±0.218#
NC	266.14±0.081#	2.070±0.143
PC	125.38±0.085	1.200±0.172
AEHR (100 mg/kg)	289.26±0.031#	3.746±0.222#
AEHR (200 mg/kg)	295.86±0.056#	5.502±0.401#
AEHR (300 mg/kg)	315.46±0.050#	7.022±0.383#
P-Value	<0.001	<0.001
F-Value	67.78	63.554

 # - significant at p < 0.05 when compared with PC

Table 3. Antioxidant parameter (SOD and TAC) activities of lead acetate induced male albino rats post-treated with *Hypoestes rosea* in the acute phase

Experimental Group	SOD (U/ml)	TAC (U/ml)
	Mean ± SEM	
EC	328.52±0.014#	9.980±0.570#
NC	277.78±0.017#	6.620±0.426#
PC	135.22±0.046	2.072±0.288
AEHR (100 mg/kg)	310.66±0.012#	5.140±0.326
AEHR (200 mg/kg)	328.48±0.019#	8.280±0.620#
AEHR (300 mg/kg)	346.22±0.040#	9.272±0.832#
P-Value	<0.001	<0.001
F-Value	95.073	25.453

 # - significant at p < 0.05 when compared with PC

Table 4. Antioxidant parameter (SOD and TAC) activities of lead acetate induced male albino rats Pre-treated with *Hypoestes rosea* in the acute phase

Experimental Group	SOD (U/ml)	TAC (U/ml)
	Mean ± SEM	
EC	328.52±0.014#	9.980±0.570#
NC	277.78±0.017#	6.620±0.426#
PC	135.22±0.046	2.072±0.288
AEHR (100 mg/kg)	299.10±0.032#	7.320±0.647#
AEHR (200 mg/kg)	305.44±0.040#	10.530±0.737#
AEHR (300 mg/kg)	325.48±0.017#	11.434±0.231#
P-Value	<0.001	<0.001
F-Value	67.781	44.618

 # - significant at p < 0.05 when compared with PC

Table 5. Antioxidant parameter (SOD and TAC) activities of lead acetate induced female albino rats post-treated with *Hypoestes rosea* in the sub-chronic phase

Experimental Group	SOD (U/ml)	TAC (U/ml)
	Mean ± SEM	
EC	340.24±0.020	8.540±0.461
NC	283.42±0.040	4.640±0.335
PC	125.38±0.085	1.200±0.172
AEHR (100 mg/kg)	322.46±0.067	9.330±0.846
AEHR (200 mg/kg)	342.70±0.032	10.920±1.060
AEHR (300 mg/kg)	354.52±0.041	11.060±0.592
P-value	<0.001	<0.001
F-value	129.431	36.129

Table 6. Antioxidant parameter (SOD and TAC) activities of lead acetate induced female albino rats pre-treated with *Hypoestes rosea* in the sub-chronic phase

Experimental Group	SOD (U/ml)	TAC (U/ml)
	Mean ± SEM	
EC	340.24±0.020	8.540±0.461
NC	283.42±0.040	4.640±0.335
PC	125.38±0.085	1.200±0.172
AEHR (100 mg/kg)	306.92±0.067	10.080±0.511
AEHR (200 mg/kg)	316.94±0.032	12.064±0.385
AEHR (300 mg/kg)	341.22±0.041	12.940±0.435
P-value	<0.001	<0.001
F-value	17.718	129.489

Table 7. Antioxidant parameter (SOD and TAC) activities of lead acetate induced male albino rats post-treated with *Hypoestes rosea* in the sub-chronic phase

Experimental Group	SOD (U/ml)	TAC (U/ml)
	Mean ± SEM	
EC	350.36±0.019	11.170±0.374
NC	293.12±0.032	7.130±0.487
PC	135.22±0.046	2.072±0.288
AEHR (100 mg/kg)	331.66±0.007	6.270±0.348
AEHR (200 mg/kg)	352.66±0.022	8.670±0.438
AEHR (300 mg/kg)	364.44±0.030	12.610±0.505
P-value	<0.001	<0.001
F-value	65.149	82.411

Table 8. Antioxidant parameter (SOD and TAC) activities of lead acetate induced male albino rats pre-treated with *Hypoestes rosea* in the sub-chronic phase

Experimental Group	SOD (U/ml)	TAC (U/ml)
	Mean ± SEM	
EC	350.36±0.019	11.170±0.374
NC	293.12±0.032	7.130±0.487
PC	135.22±0.046	2.072±0.288
AEHR (100 mg/kg)	316.96±0.032	6.270±0.348
AEHR (200 mg/kg)	324.78±0.039	11.380±0.435
AEHR (300 mg/kg)	351.64±0.018	13.370±0.306
P-value	<0.001	<0.001
F-value	56.949	109.035

3.3 Superoxide Dismutase (SOD)

Superoxide dismutase is the only eukaryotic enzyme capable of detoxifying superoxide radical, an initiator of radical chain reaction [31]. It catalyzes the conversion of superoxide radical to hydrogen peroxide, which may be subsequently converted to water by catalase or peroxiredoxin [23]. The fact that all aerobic organisms, and even some anaerobic ones, express superoxide dismutase underscores the importance of this enzyme [23].

Exposure of the albino rats to lead acetate caused a decline in the SOD value as seen in the PC group. Lead causes oxidative damage to various tissues via lipid peroxidation which leads to loss of membrane function [28]. The resultant activity causes inhibiting of SOD is one of main enzymes involved in the anti-oxidant defense mechanism in our body. SOD works against superoxide anions. There are different types of SODs, depending on the metals that co-factor with the protein, including Cu, Zn, Fe, Mn and Ni. Lead basically displaces metals in the functional group of SOD which accounts for the acute depletion of the SOD value of the PC rats. Lead is bivalently charged when ionized, hence its radical ability with bivalent ions such as Zn^{2+}, Cu^{2+} and Fe^{2+} [32] rendering the

resultant enzyme useless in catalyzing the redox reaction.

The finding of our study is in agreement with Sudjarwo et al. [28]. In their researches, they found out the administration of lead acetate caused a reduction in SOD value of albino rats. In another study by Kashyap et al. [25] rats were administered lead nitrate for 7 days. Bio-chemical tests afterwards showed a significant decrease in SOD level. The results of this study are in accordance with our study.

In our study, acute pre-treatment of both sex of albino rats with lead acetate prior to administration of the various doses of aqueous extract of *hypoestes rosea* leaf showed the SOD level of all treated albino rats significantly increased compared to the PC group. This confirms the plant enhances the production of SOD, considering the effect of pre exposure of the animals to lead acetate and subsequent generation of reactive oxygen species as a result of oxidative stress caused by administration of lead acetate. The result of our study showed that with the administration of the varying doses, the SOD value of all the animals despite the pre administration of lead was significantly higher than that of the normal control also. This further confirms that the ingestion of the plant increases antioxidant capacity and can ameliorate the effect of lead acetate on the animals.

In the acute post treatment group, the prior administration of the plant extract on the albino rats before the lead showed from the SOD results that the plant was able to protect the exposed animals significantly against the tissues damage resultant from oxidative stress generated by reactive oxygen species as a result of lipid peroxidation from lead acetate. Our result shows that the mean values of the SOD of the pre-treatment phase of the animals are significantly higher than the post-treatment phase and moreso as the varying doses increased, the mean SOD vale increased also.

In our study, the sub chronic exposure of the albino rats in the pre-treatment and post treatment phase for both male and female albino rats and subsequent administration for a longer period showed a more increased mean value of SOD for all groups of treated albino rats. The significant was relative to the dose of administration in the different groups as increased doses caused more significant increase in the SOD value of the albino rats.

This suggests aqueous extract of hypoestes rosea leaf at acute exposure can conveniently reverse the effect of lead acetate, even as it can protect the system against the effect of lead acetate poisoning in both sex. The findings of our study here suggest the ability of plant extract to boost the production of antioxidants that mop up free radicals in the system. Our findings is in agreement with the findings of Wegwu and Didia [33] and Waribo et al. [32] which saw the seeds of *Garcinia kola* possessing natural antioxidants that protects the cell against free radical damage. In the study they discovered that pretreatment with Gercinia kola was able to salvage the cells from the activities of free radicals.

4. CONCLUSION

This study showed that lead acetate induced oxidative stress in the rats, evidenced by the drop in anti-oxidant parameters (SOD and TAC). This study also found that the administration of aqueous extract of *Hypoestes rosea* at a concentration of 100, 200 and 300 mg/kg rat body weight reversed the effect of lead acetate induction in a dose dependent manner and could also protect the albino rats against subsequent toxicity against lead acetate.

CONSENT

It is not applicable.

ETHICAL APPROVAL

Animal ethic Committee approval has been collected and preserved by the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Musa AS, Iliyasu MO, Hamman WO, Ibegbu AO, Umana UE. Preventive activity of ascorbic acid on lead acetate induced cerebellar damage in adult wistar rats. Medical and Health Science Journal. 2012;13:99-104.
2. Flora G, Gupta D, Tiwari A. Toxicity of lead. A review with recent updates. Interdisciplinary Toxicology. 2012;5(2): 47-48.

3. Ani M, Moshjaghi HA, Aghadavood M. Protective effects of selenium and zinc on the brain acetyl cholinesterase activity in lead intoxified rats. Research Pharmaceutical Science. 2006;2:80-84.
4. Vaziri ND, Gonick HC. Cardiolvascular Effects of lead exposure. Indian Journal of Medical Research. 2008;128(4):426-435.
5. Sudjarwo SA, Koerniasari SN. Protective effects of ethanol extract of Mangosteen (*Garcinia Mangostana* L.) Pericarp against lead acetate-induced nephrotoxicity in mice. Global Journal of Pharmacology. 2015;9(4):385–391.
6. Koerniasari S, Ngadino R, Sudjarwo SA. Protective effect of ethanol extract of Mangosteen (*Garcinia* Mangostana *L*.) pericarp against lead acetate induced hepatotoxicity in mice. International Journal of Current Research. 2015;7(2): 12518–12522.
7. Adhikari N, Sinha N, Marayan R, Saxena DK. Lead induced cell death in testes of young rats. Journal of Applied Toxicology. 2001;21(4):275-277.
8. Owolabi JO, Ghazal OK, Williams FE, Ayodele EO. Effect of *Moringa Oleifera* (Drumstick) leaf extracts on lead induced testicular toxicity in adult wistar rats (*Rattus Novargiens*). International Journal of Biotechnology & Biomedical Research. 2012;2(12):4003-4009.
9. Griveau JF, Le Lannou D. Reactive oxygen species and human spermatozoa: Physiology and pathology. International Journal of Andrology. 1997;20(2):61-69.
10. Koksal I, Usta F, Orhan I, Abbasoglu S, Kadioglu A. Potential role of reactive oxygen species on testicular pathology associated with infertility. Asian Journal of Andrology. 2003;5(2):95-99.
11. Agarwal BB, Ichikawa H, Garodia P, Weerasinghe P, Sethi G. From traditional ayurvedic medicine to modern medicine: Identification of therapeutic targets for suppression of inflammation and cancer. Expert Opinion on Therapeutic Targets. 2006;10:87-118.
12. Geetha A, Lakshmi PM, Javachristy A, Swendran R. Level of oxidation stress in the red blood cells of patients with liver cirrhosis. Indian Journal of Medical Research. 2007;126:204-210.
13. Neupane DP, Majhi S, Chaudra L, Rijal S, Baral N. Erythrocyte glutathione status in human visceral leishmaniasis. Indian Journal of Clinical Biochemistry. 2008;23: 95-97.
14. World Health Organization. WHO traditional medicine strategy 2002-2005.
15. Piero NM, Eliud NN, Susan KN, George OO, David NJMM. *In Vivo* Antidiabetic activity and safety In rats of *Cissampelos Pareira* traditionally used In the management of diabetes mellitus In Embu County, Kenya. Journal of Drug Metabolism &Toxicology. 2015;6:184-188.
16. Sofowora A, Eyitope O, Adedeji O. The role and place of medicinal plants in the strategies for disease prevention. African Journal of Traditional, Complimentary Medicine. 2013;10(50): 210-219.
17. Levin SM, Goldberg M. Clinica evaluation and management of lead exposed; 2000.
18. Organization for Economic Cooperation Development OECD. Guidance document on acute oral toxicity. Environmental Health and Safety Monograph Series on Testing and Assessment, No.24. of Clinical Nutrition. 1999;53:319-327.
19. Karl-Heinz, D., Robin, H., David, M., Rudolf P, Yvon R, David S, Jean-Marc V, Cor V. A good practice guide to the administration of substances and removal of blood, Including Routes and Volumes. Journal of Applied Toxicology. 2001;21: 15–23.
20. Office of Laboratory Animal Welfare [OLAW]. Institutional Animal Care and Use Committee Guidebook. (2nd edition). Bethesda: NIH Publication; 2002.
21. Institute for Laboratory Animal Research [ILAR] Guideline for the Care and Use of Laboratory Animal.8th edition, Washington D.C: National academic press; 2011.
22. Public Health Service [PHS]. Public health Service policy on humane care and use of laboratory animals. Publication of the Department of Health and Human Services. National Institute of Health. Office of Laboratory Animal Welfare; 2015.
23. Barbosa KBF, Volp ACP, Rocha JLM, Ribeiro SMR, Navarro-Blasco I, Zulet MÁ, Martinez JA, Bressan J. A low caloric and carbohydrate intake is associated with higher total antioxidant capacity in apparently Healthy Adults. Nutrition. 2014; 30(11-12):1349 – 1354.
24. Erel OA. New automated colorimetric method for measuring total oxidant status.

Clinical Biochemistry. 2005;38(12):1103-1111.

25. Kashyap MK, Yadav V, Sherawat BS, Jain S, Kumari S, Khullar M, Sharma PC, Nath R. Different antioxidants status, total antioxidant power and free radicals in essential hypertension Molecular and Cellular Biochemistry. 2005;277(1-2):89-99.
26. Kusano C, Ferrari B. Total antioxidant capacity: A biomarker in biomedical and nutritional studies). Journal of Cellular and Molecular Biology. 2008;7(1):1-15.
27. Birben E, Sahiner UM, Sackesen C, Erzurum S, Kalayci, O. Oxidative stress and antioxidant defense. World Allergy Organization Journal. 2012;5(1): 9–19.
28. Sudjarwo SA, Sudjarwo GW, Koerniasari I. Protective effect of curcumin on lead acetate-induced testicular toxicity in wistar rats. Journal Of Research In Pharmaceutical Science. 2017;12(5):381-390.
29. Malecka A, Jarmuszkiewicz W, Tomaszewska B. Antioxidant defense to lead stress in subcellular compartments of pea root cells. Acta Biochemical Polonica, 2001;48(3):687-698.
30. Adaramoye OA, Farombi EO, Emerole GO. Comparative study of the antioxidant properties of flavonoids of *Garcinia Kola* seeds. Pakistan Journal of Medical Science. 2005;21(3):331-339.
31. Van Raamsdonk JM, Hekimi S. Superoxide dismutase is dispensable for normal animal lifespan. Proceedings of the National Academy of Sciences of the United States of America. 2012;109(15): 5785–5790.
32. Waribo HA, Bartimaeus ES, Nwanjo HU. *Gercinia Kola* seed and vitamin E ameliorates acetaminophen induced oxidative stress in albino rats. European Journal of Pharmaceutical and Biomedical Research. 2017;4(11):130-136.
33. Wegwu MO, Didia BC. Hepatoprotective effect of *Garcinia Kola* seed against hepatotoxicity induced by carbon tetrachloride in rats. Biochemistry. 2007; 19(1):17-21.

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