



Potential Contribution of Exposure to Insecticide Used in Nigeria to the Development of Dyslipidaemia and Hormonal Disorder in Male and Female Albino Wistar Rats

Ganiyu O. Adeosun^{a*}, David D. Ajayi^b,
Thomas A. Mkwogwu^c, Oloruntoba A. Ekun^d,
Remilekun Akinrinmade^e, Magnus M. C. Anyakudo^f
and Samuel O. Oyedeji^a

^a Department of Medical Laboratory Science, University of Medical Sciences, Ondo City, Ondo, Nigeria.

^b Department of Medical Laboratory Science, Federal University, Oye Ekiti, Nigeria.

^c Department of Medical Laboratory Science, Faculty of Science, Delta State University, Abraka, Nigeria.

^d Department of Medical Laboratory Science, College of Medicine, University of Lagos, Nigeria.

^e Department of Human Nutrition and Dietetics, University of Medical Science, Ondo City, Ondo State, Nigeria.

^f Department of Physiology, Faculty of Basic Medical Sciences, University of Medical Sciences, Ondo City, Ondo, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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*Corresponding author: E-mail: gadeosun@unimeds.edu.ng;

ABSTRACT

Background: Raid is a routine liquid insecticide used frequently in the domestic homes to keep away mosquitoes from biting its human prey. The application of insecticide is not usually guided in most homes. This may portend some danger to the lipid and reproductive health of man. The study aims to evaluate the effect of the routine household insecticide used in Nigeria on the Lipids and reproductive hormones in male and female albino Wistar rat.

Methodology: Forty apparently healthy Wistar albino rats comprising 20 males and 20 females' rats were grouped in to 5 with an average weight of $110 \pm 20g$ were exposed once a day to the fume of aqueous solution of Raid in an air tight exposure chamber for 28days. Control Wista albino were kept under normal sterile environment in the animal house devoid of insecticide pollution for 28days. The animals were fed with standard feed and water. Blood specimens were harvested through cardiac puncture after overnight fast and 28 days exposure.

Triglyceride, Total cholesterol, HDL, LDL cholesterol, LH, FSH, Estrogen., Progesterone and testosterone were evaluated at the end of exposure period using a reference method.

Results: Exposure to the domestic insecticide significantly induced weight loss indicated by marked decrease in weight of both the exposed and un -exposes groups. The pyrenoid of the insecticide have no significant effect on the lipid profile indices of the exposed Wistar albino rats. Similarly, exposure of the rats to the insecticide had no significant effect on the FSH and progesterone levels. However, the LH, Oestrogen and β - HCG were significantly altered among the female experimental animals. LH only was significantly raised among the male albino rats exposed to the insecticide.

Conclusion: Acute exposure to domestic insecticide has no significant effect on the lipid profile, but alters the concentrations of LH and β - hCG hormones in the albino Wistar rats.

Keywords: Albino Wistar rat; reproductive and lipidemic effect; raid, insecticide.

1. INTRODUCTION

Excessive use of various chemicals including insecticides has become a public health concern. The Use of insecticides and other organophosphates is one of the major ways through which manufacturing workers and farmers are exposed to toxicants and this has impact on the ecosystem and public health [1]. Insecticides are natural or man-made preparation that is used to kill or otherwise control insects such as mosquitoes, cockroaches, bees, wasp, and bedbug and many other insects.

They can be can be grouped in to two major classes: systemic insecticides, which are known to have residual or long-term activity and secondly contact insecticides, that has no residual activity. They are in form of sprays, dust, gel, baits, smokers, fumigant and powders [2]. A common active chemical in insecticides are pyrethroids, derivatives of pyrethrines, natural substances obtained from the flowers of pyrethrium species [2a and 2b]. The most common active ingredient in insecticide are synergist, carbamate, whose common name is propoxur, pyrethrin (or synthetic pyrethroids), D-trans-allevrin, permethrin, tetrametrin, deltametrin, cyfluthrin, imiprothrin, chlopyritos,

Diaznon, Malathion, Silica gel, Boric acid, Arsenicals, paradichlorobenzene, Naphthalene, N,N-diethyl meta-toluamide (3Deet), Dimethylphthalate [2b] The mechanism of action of insecticide can be important in understanding whether an insecticide will be toxic to unrelated species, such as fish, birds and mammals. In terms of brand, insecticides' may be repellent or non-repellent [3]. Domestic insecticides are known to be quite different from non-insecticidal repellents, which only keep away insects.

In Nigeria, systemic insecticides are the most common and they include; Rambo manufactured by Gongoin and Co, Mortein manufactured by Reckitt Benckiser, Raid and Baygon both manufactured by S.C. Johnson and Co. The common household Insecticides are distinct from non-insecticidal repellents, which only repel but do not kill [4] However routine household insecticides used in Nigeria are systemic in action and these include Rambo produced by Gongoin and Co, Mortein produced by Reckitt Benckiser, Raid and Baygon both produced by S.C. Johnson Co. [4].

Mosquitoes and man cohabit in most homes in Nigeria and thus it becomes a herculean task to completely eliminate mosquitoes in many homes in our country. The resurgence of mosquitos few

minutes/hours after application of insecticide show that either the insecticide is not effective or it is a sign of resistance of the mosquitoes to the applied insecticide. This confirms that mosquitoes cannot be completely eradicated in Nigeria. This limitation of the insecticide is not known to many and can lead to abuse or indiscriminate use of insecticide in most homes without consideration of the negative effect on their health.

Lipids are class of compounds that are soluble in organic solvent and nearly insoluble in water. They have also been described as naturally occurring esters of glycerol and fatty acids which are solid at room temperature as opposed to oils which are similar esters but liquid at room temperature [5]. Structurally lipids consist of heterogeneous group of compounds soluble in non-polar solvents such as methanol, ether, chloroform and benzene. Hydrolysis of lipids yields fatty acids or complex alcohols that may combine with the fatty acids to form esters. Some lipids are more complex by the possession of other groups like phosphoric, amine, sugar, or sulphate groups which confers *amphipathic* character on them. Lipids are ubiquitous in the body tissue, playing important role in virtually all the physiological processes of the body which include, lipids serves as hormone or hormone precursors, aids digestions, facilitates energy storage, provides metabolic fuels, acts as functional and structural components in cell membrane, and also forms insulation to allow nerve conduction or prevent heat loss [6] Simple, Compound and Derived derivatives are the three majors classes of lipid fraction. Each of the fractions has a specific physiological and metabolic role in the body, the alteration of the equilibrium concentration of these lipid fraction may precipitate a pathological disorder in the body.

The reproductive hormones are essential physiological requirements for procreation and they include; Follicle stimulating hormone (FSH). Luteinizing hormone (LH) Oestrogen and Progesterone, each of them contributing specific role in human in procreation. LH and FSH are gonadotrophins synthesized by the pituitary gland [7] but released by the Gonadotropin releasing hormone (GnRH) a product of the hypothalamus. Testosterone a reproductive hormone is a product of the extra tubular Leydig cells regulated by LH, a glycoprotein synthesized by the pituitary [7]. FSH synthesized by the pituitary is another gonadotropic hormone that

controls sperm production by the regulation of the activities of the both the germinal epithelium and Sertoli cells [8]. Physiological concentration of these hormones is under the control of hypothalamic pituitary gonadal axis [9]. Testosterone improves sperm motility and epididymis function. Oestrogen and Progesterone are female sex hormone which act synergistically to regulate the female accessory sex organs and female secondary sex characteristics, the menstrual cycle, breast, uterine growth and in the maintenance of pregnancy [9].

These hormones are produced by the ovarian follicles and the corpus luteum and during pregnancy by the placenta.

Combined physiological contribution and function of FSH and LH is required for the synthesis of Testosterone in males and Ovarian hormones (oestrogen and progesterone) in female which are essential and inevitable requirements that facilitates fertility [10].

Human Chorionic gonadotropin (hCG) synthesized in the syntiotrophoblast cells of the placenta is a glycoprotein containing a protein core with branched carbohydrate side chains which usually terminate with sialic acid. hCG is composed of two non-identical, non-covalently bound glycoprotein subunits, the alpha (α) and the beta (β) subunits [11]. The chorion of the developing placenta begins to secrete hCG shortly after implantation of the fertilized egg and its concentration rises steadily in plasma and urine from the first few days after conception until the tenth or twelfth week of pregnancy [12,13] [12,13]. The detection of hCG in urine or serum is a biochemical confirmation of establishing pregnancy as early as 7-to-10-day post conception [13]. The action of hCG is similar to that of LH because it also stimulates the corpus luteum to produce progesterone. The aim of this study was to evaluate the effect of the routine household insecticide used in Nigeria on the Lipids and reproductive hormones in male and female albino Wistar rat.

2. MATERIALS AND METHODS

2.1 Collection of Insecticides

Insecticide (Liquid brand) insecticide was purchased directly from a super market in Ile-Ife, Nigeria It was preserved at room temperature for use during the experiment.

2.2 Acquisition of Lipid and Hormonal Kits and Equipment

Lipid profile, FSH, LH and Testosterone kits were purchased from a reliable reagent supplier based in Lagos, Nigeria. The equipment; Bench centrifuge, Spectrophotometer, ELISA machine and reader were available and accessible in the research laboratory where the assay was done.

2.3 Animal Selection

Healthy male and female albino rats of the Wistar strain (*Rattus norvegicus*), with an average weight of 110 ± 20 g, obtained from the Animal Breeding House of the Obafemi Awolowo University (OAU), Ile-Ife were acclimatized for one week prior to the commencement of the project. The rats were restricted to a clean quiet, well ventilated and temperature controlled experimental animal cages for 12/12 hours except during the period of exposure to the insecticide. The experimental work was performed in agreement with the guidance for care and use of laboratory animals. The median lethal dose (LD50; 25mg/kg body weight) for commercial formulations of Raid, (a domestic insecticide) on Wister albino rats from previous research was adopted for this study. They were randomly divided into five groups of six rats, each and kept in different rooms.

Twenty male and twenty female Wistar rats weighing between 109 and 133 kg were used for this study. They were randomly divided into five groups and each group contained eight animals acclimatized for seven (7) days to laboratory conditions before the commencement of the experiment. Prior to and during the experiment, the animals were acclimatized and the male were separated from the females and both were fed *ad libitum* with standard feed and drinking water in the clean cages placed in well-ventilated housing conditions (under humid tropical conditions) throughout the experiment. The animals were given humane care according to the criteria outlined in the 'Guide for the Care and Use of Laboratory Animals' prepared by the National Academy of Science and published by the National Institute of Health.

The rats in group 1 were exposed once to the insecticide and tagged acute group. The rats in group 2 were exposed trice to the insecticide at interval of 48 hours and was tagged sub- chronic group. The third groups of the rats were exposed four times to the insecticide at interval of 48 hours and was labelled the chronic group. The

rats in the fourth group were exposed four times to the insecticide at the interval of 48 hours but were left for two weeks undisturbed except for feeding. This group was labelled post exposure group and the animals in the group were sacrificed for sample collection 7 days post exposure. While the rats in group 5 were not exposed at all. (The control group). All animals received humane care in compliance with the guidelines of the Obafemi Awolowo University Ile-Ife Animal Care and Use Research Ethics Committee (ACUREC). The exposure was done twice daily via inhalation route. During the duration of the experiment the albino rats were fed ad libitum with standard feed and hygienic water. the albino rats were fed ad libitum with standard feed and hygienic water. The rats in each group were sacrificed under diethyl ether as anaesthesia after an overnight fast

Blood was collected from each rat in the group through cardiac puncture with the aid of a needle and syringe. Two milliliters (2ml) of blood were dispensed into K-EDTA anticoagulant bottle for lipid estimation while 3ml was dispensed in to plain bottle for the hormonal assay.

2.4 Assay of Reproductive Hormones

The serum progesterone, testosterone beta HCG, FSH, and LH and were measured with the aid of enzyme-linked immunosorbent assay (ELISA) according to the methods described by [14] Hidayat and Patricia, [15].

2.5 Assay of Lipid Profile

Plasma cholesterol and triglyceride were assayed using Enzymatic method described by [16] Li-Hua, 2019 and [15] while the plasma HDL, LDL and VLDL- cholesterol were measured by [17,18,19].

3. RESULTS

Table 1. Explains the distribution of weight change among the Albino rats used for this experimental research before and after exposure to the domestic insecticide is shown in Table 1. Among the exposed group, the sub chronic had the highest weight loss (122%) while the acute exposed group had the lowest weight loss (52.1%) when compared with the unexposed control group. The unexposed control group gained weight significantly when compared to all the exposed groups. The comparative loss of

Table 1. Mean Initial and Final Weight of the all the Exposed and Unexposed Albino rats to Insecticide (Groups 1-5)

Variables	Grp 1 (Acute exposure)	Grp 2 (Sub-chronic exposure)	Grp 3 (Chronic Exposure)	Grp 4 (Post chronic Exposure)	Grp 5 (Control)	p-value
Initial weight (Kg)	109.53±5.4	135. ±895.2	92.20±6.0	136.75±4.5	133.55±5.2	P<0.05
Final Weight (Kg)	166.60±8.8	300.86±5.8	168.24±7.9	214.01±9.7	525.20±4.20	P<0.05
Change in weight (Kg)	57.07±3.5	164.97±4.7	76.04±4.9	77.26± 4.8	392.05±7.5	P<0.05
% Chang in weight (Kg)	52.10	122.20	82.47	56.50	293.56	P<0.05

Table 2. Lipid profile in the different groups of exposed and non –exposed albino rats to insecticide

Variables	Grp 1 (Acute exposure)	Grp 2 (Sub-chronic exposure)	Grp 3 (Chronic Exposure)	Grp 4 (Post chronic Exposure)	Grp 5 (Control)	p-value
TG (mmol/L)	1.35±0.13	0.93±0.10	1.13±0.10	1.20±0.22	1.15±0.13	P>0.05
TCHOL (mmol/L)	2.35±0.16	2.58±0.17	3.01±0.13 *	2.8±0.12	2.65±0.12	P>.05
LDLC (mmol/L)	0.70±0.08	1.18±0.35	0.78±0.33	0.41±0.17	0.80±0.23	P>0.05
HDLC (mmol/L)	1.20±0.07	0.83±0.10*	1.78±0.25 *	1.9±0.24	1.33±0.15	P>0.05

Table 3. Reproductive hormones profile in the different groups of exposed and non –exposed female albino rats

Variables	Grp 1 (Acute exposure) (Mean ±SD)	Grp 2 (Sub-chronic exposure) (Mean ±SD)	Grp 3 (Chronic Exposure) (Mean ±SD)	Grp 4 (Post chronic Exposure) (Mean ±SD)	Grp 5 (Control) (Mean ±SD)	p-value
LH	1.2±0.10	0.83±0.10	1.56±0.12	1.7±0.82	1.62±0.10	P<0.05
FSH	0.51±0.10	0.52±0.01	0.54±0.01	0.53±0.82	0.51±0.10	p>0.05
PROG	23.3±0.32	9.03±0.17	16.25±0.96	5.00±0.82	4.48±0.10	p>0.05
OEST	40.6±0.36	42.702±0.27	32.50±1.29	47.30±0.90	62.72±0.82	P<0.001
β- hCG	6.5±0.08	1.302±0.19	2.48±0.17	2.75±0.06	4.12±0.10	P<0.001

Table 4. Reproductive hormone level profile in exposed and non- exposed male albino rats to insecticide

Variables	Grp 1 (Acute exposure) (Mean ±SD)	Grp2(Sub-chronic exposure) (Mean ±SD)	Grp3 (Chronic Exposure) (Mean ±SD)	Grp4 (Post chronic Exposure) (Mean ±SD)	Grp 5 (Control) (Mean ±SD)	p-value
LH	1.71±0.08	1.70±0.08	1.55±0.13	1.15±0.13	1.90±0.08	P<0.05
FSH	0.60±0.07	0.56±0.01	0.48±0.01	0.54±0.02	0.56±0.08	p>0.05
Testosterone	0.30±0.08	2.15±0.13	0.58±0.01	9.68±0.17	8.1±0.08	p>0.05

weight by the exposed albino rats might be due to the combined negative effects of the insecticide which might have been precipitated through insecticide inhalation causing irritation. loss of appetite, distortion and probably inactivation of the digestive system.

Table 2 shows the MEan \pm S.E.M plasma lipid profile in both the exposed and non –exposed Albino rats to the domestic insecticide.

All the lipid profile indices in the exposed group (groups 1to 4) were not statistically different ($P>0.05$) when compared to the unexposed groups (group5). Comparison of the lipid profile indices among different groups of exposed albino rats (groups 1 to 4) showed no statistically significant difference ($P>0.05$).

The hormonal profiles in the different groups of the exposed and non- exposed female albino rats are shown in Table 3.

There was no statistically significant difference ($P>0.05$) in the FSH and progesterone concentrations values between the control and exposed groups. However, there was statistically significant difference ($P<0.05$) in LH, Oestrogen and β - hCG values between the control and the exposed groups. Comparison of the reproductive hormones among different groups of exposed female albino rats (1 to 4) showed no statistically significant difference ($P>0.05$) for all the reproductive hormones.

The mean \pm S.E.M. of the reproductive hormones in the exposed male group only were presented in Table 4. Comparison of the reproductive hormones among different groups of exposed male albino rats (groups 1 to 5) showed no statistically significant difference ($P>0.05$) for all the reproductive hormones except for the LH which was significantly reduced ($P>0.05$) when compared with the non-exposed control group.

4. DISCUSSION

Exposure to different insecticide has been reported to cause adverse effects on the physiological equilibrium of the biochemical and the biological systems of different tissues [20] Exposure to Raid, a domestic insecticide significantly induced weight loss in the exposed rats relative to the un-exposed group. (Table 1). The weight loss by the exposed rats might be due to the combined negative effects of the insecticide which might have been precipitated through insecticide inhalation causing irritation

loss of appetite and probably inactivation of the digestive system.

The holistic values obtained for the lipid and reproductive hormones suggests that exposure to the insecticide has no significant adverse effect on the hormonal and lipid indices among the exposed albino rats (Tables 2 & 4). Our study showed no statistically significant difference ($P>0.05$) between the values obtained for the lipid indices among the exposed groups compared to the controls therefore this study shows no correlation between the duration of exposure and the lipid indices. However, this is in contrast with the reports of [21 and 22] who reported significant ($p<0.05$) increase in the levels of lipids in the exposed albino Wistar rats in relation to the duration of exposure to the raid insecticide. We observed significant decrease ($P<0.05$) in the values of LH, Oestrogen and β -hCG in the the female exposed groups only than the in the unexposed female rats but the values were within the reference range. This result agrees with [23] who reported decreased levels of the reproductive hormones among exposed Wistar albino rats.

In contrast, the values obtained for the FSH and progesterone concentrations among the female rats in this study showed no significant difference ($P>0.05$) between the exposed Wistar rats and the control group and they are within the reference values. This result is at variance with the findings of [24] who reported significant concurrent reduction in the gonadotropic hormone, oestradiol, and progesterone levels in insecticide treated rats.

In this study, we also obtained a significantly ($P<0.05$) reduced LH and a non-significant difference ($p>0.05$) in the testosterone value among the male albino rats compared to the control's male controls, however the values were within the reference range [25]. This is in contrast with [26] who reported significant decrease in LH, FSH and testosterone levels between the exposed and non-exposed control male albino rats [27,28].

5. CONCLUSION

We conclude from the findings of this study that exposure to insecticides have no significant effect on the concentration of lipids and reproductive hormones in both female and male Wistar rats. The results of this research further indicate that exposure to routine domestic

insecticides applied in Nigeria homes is not linked with dyslipidemia and hormonal disorder. However, cohabitation with the aerosol of the insecticides in a fumigated room should be prevented.

CONSENT

It is not applicable.

ETHICAL APPROVAL

Ethical approval was obtained from Animal Care and Use Research Ethics Committee (ACUREC), of Obafemi Awolowo University Ile-Ife. (OAU),

COMPETING INTERESTS

Authors have declared that no competing interests exist

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