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TOXICOLOGICAL PROFILE OF PREGNANT WISTAR RATS AFTER EXPOSURE TO SOME COMMONLY USED HOUSEHOLD AEROSOL INSECTICIDES IN NIGERIA

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ABSTRACT

This study evaluated the toxicity of selected household aerosol insecticide formulations on pregnant Wistar rats. Healthy, twenty-four pregnant Wistar rats (210.05 ± 1.13 g) were randomized into four groups. Sham control was not exposed to insecticide, while rats in groups A, B and C were exposed to household aerosol insecticide A (HAIA), B (HAIB) and C (HAIC) for 21 days. The results revealed loss of fur and significant ($P < 0.05$) reduction in total number of fetuses, weight of fetuses and number of live fetuses of the aerosol exposed pregnant rats when compared with the sham control. The HAIA, HAIB and HAIC significantly ($P < 0.05$) reduced the activities of ALP, AST and ALT in the liver with corresponding increases in the serum enzymes. The HAIA, HAIB and HAIC significantly ($P < 0.05$) reduced the serum total protein, albumin, globulin, Na^+ , K^+ and Cl^- and MCV; levels of SOD, catalase, GPX and GRed in the liver, kidney and ovary contents of the pregnant rats whereas serum total bilirubin and conjugated bilirubin contents, WBC, RBC, PVC and malondialdehyde contents in the liver, kidney, ovary and serum of pregnant rats increased significantly ($P < 0.05$). Compared with the sham control, only HAIA reduced the kidney ALP activity whereas it was only HAIC that increased the kidney ALT activity. Serum urea and creatinine levels, MCH and platelets were not significantly altered by HAIA, HAIB and HAIC. Exposure of pregnant rats to HAIA, HAIB and HAIC deranged the lobular architecture of the hepatocytes, central veins and portal tracts of the liver of the animals. Thus, the household aerosol insecticides adversely affected the functional indices of the liver and kidney of pregnant Wistar rats as well as liver histoarchitecture via oxidative mechanism.

Keywords: Aerosol insecticide, functional toxicity, oxidative stress, structural toxicity

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INTRODUCTION

Household aerosol insecticides are sprays which contain active pesticides and other inert ingredients used mainly for killing insects like resident flying and crawling insects e.g. mosquitoes and cockroaches. Household aerosol insecticides are usually pretty typical, and it is a popular mode of personal protection against household pests in all parts of the world. Different types include pyramide insecticide aerosol (potent botanical insecticide), phantom insecticide aerosol II (famous for aerosol insect pesticide), bedlam plus insecticide aerosol (kill bugs with their eggs), cb 80 insecticide aerosol (acetone free), piranha aerosol insecticide (contains ditronella scented) and alpine pt insecticide aerosol (perfect indoor insecticide) [1]. The most widely used active chemicals in insecticides are synergist, carbamate, also known by the generic version as propoxur, pyrethrin (or synthetic pyrethroids), Dtrans-alletrin, permethrin, pralletrin, tetrametrin, deltametrin, cyfluthrin imiprothrin chlopyritos, diaznon, malathion, silical gel, boric acid, arsenicals, paradichlorobenzene, naphthalene, N,N-diethyl meta-toluamide (Deet) and dimethylphthalate [1]. Pyrethrins, pyrethroids and carbamates are effective insecticides that are often used in household as sprays, insect repellent, pet shampoo, and lice treatment [1].

Some households in Nigeria prefer to use insecticides that are powdered, liquid, or aerosolized. The persistent exposure of human beings to these insecticides is currently attracting attention as it might result in insecticide toxicity as well as acute and chronic toxicities in non-target species including humans [2,3,4]. Human exposure to cypermethrin is reported to occur mainly occupationally, during application or through pyrethroid residues such as those detected in food and on the body [2,5].

Previous studies have reported adverse effects of these insecticides on different functionalities of the parts of mammals. For

instance, acute exposure to insecticide (Baygon-spray; 5ml/animal/5min) inhalation in rats did not affect the learning process but produced a significant loss of memory whereas chronic exposure on daily basis for three weeks delayed learning and memory and reduced acetylcholinesterase activity in the brain [6]. Achudume *et al.* [7] also revealed that bioaccumulation of Raid insecticide in the tissues was in the order of lipid > muscle > liver > brain, accompanied by reduction of glucose-6-phosphatase and lactic acid dehydrogenase levels in muscle and liver. Somade *et al.* [8] have also reported that prolonged and incessant exposure to Baygon insecticide aerosol over a long period of time may lead to tissue oxidative stress. Furthermore, Liu *et al.* [9] have also reported that inhalation of tetramethrin and prallethrin stimulate plasma biophysical and modify serum aminotransferases, erythrocyte membrane cholesterol and phospholipid levels, individual phospholipids and membrane fluidity of exposed rats.

Despite the foregoing studies on insecticide aerosols, there is dearth of information on the effects of commonly used insecticide aerosols in Nigeria on pregnant rats. The current study thus evaluates the toxicological profile of pregnant Wistar rats after exposure to some commonly used household insecticide aerosol formulations.

MATERIALS AND METHODS

Three commonly used household insecticide formulations in Nigeria were used and referred to as Household Aerosol Insecticide A (HAIA), Household Aerosol Insecticide B (HAIB) and Household Aerosol Insecticide C (HAIC). Household Insecticide A contained 0.250% of D-allethrin (pyrethroid), 0.150% of tetramethrin (pyrethroid) and 0.015% of deltamethrin (pyrethroid) whilst HAIB contained 1% of dichlovos (organophosphate) and 0.50% of permethrin (pyrethroid). Household Aerosol Insecticide

C contained 0.05% of imiprothrin (pyrethroid), 0.05% of prallethrin (pyrethroid) and 0.015% of cyfluthrin (pyrethroid). The HAIA and HAIC were products of Johnson Wax Nigeria Ltd, Isolo Industrial Estate, Isolo, Nigeria whereas HAIB was factory made by Gongoni Nigeria Ltd, Sharada Industrial Estate, Bompai, Kano State, Nigeria.

Assay kits

The assay kits for total protein, albumin, total and conjugated bilirubin, alkaline phosphatase (ALP), alanine aminotransferase (ALT), aspartate aminotransferase (AST), Na⁺, K⁺, Cl⁻, urea and creatinine were products of Randox Laboratories Ltd, Co-Antrim, UK. Superoxide dismutase (SOD), catalase, glutathione peroxidase (GPx) and reduced glutathione (GRed) were products of Fortress Diagnostics Limited, Antrim Technology, United Kingdom. All other reagents used were of analytical grade obtained from Sigma-Aldrich Ltd., Mannheim, Germany.

Haematological Analyzer

The automated hematology analyser (Sysmex KX-21N) was manufactured by Sysmex Corporation, Hyogo, Japan.

Experimental animals

Healthy, twenty-four female Wistar rats weighing (210.05 ± 1.13 g) and twenty-four healthy male Wistar rats weighing (225.14 ± 2.35 g) were obtained from the Biological Garden, College of Education, Ilorin, Nigeria. The animals were kept in stainless steel cages with a dimension of 19 x 19 x 12 cm (L x B x H), placed in well-ventilated Animal Housing of the Department of Biochemistry, University of Ilorin, Ilorin, Nigeria with the housing conditions of temperature ($28 \pm 3^\circ\text{C}$), photoperiod (12 h natural light and 12 h dark) and humidity (40-45%).

Animal pairing and confirmation of pregnancy

The procedure described by Ogunro and Yakubu [10] was adopted. Briefly, female Wistar rats were brought into artificial oestrous by sequential administration of estradiol benzoate (10µg/100 g body weight) and progesterone (0.5mg/100 g body weight, subcutaneously), 48 hours and 4-6 hours, respectively. Thereafter, a receptive female rat was placed in the cage containing a male rat (1:1). The pairing of the male rat and female rat was done overnight, beginning from 7:00 pm in a dim light and in the absence of the feed to allow for mating. Exactly 24 hours after pairing, the vagina of the female was observed for the presence of plug; vaginal smear was further prepared and observed under the light microscope for the presence of spermatozoa. The day on which spermatozoa were detected in the vaginal plug was regarded as day zero of pregnancy [11].

Animal grouping and exposure of pregnant rats to household aerosol insecticides

Twenty-four pregnant rats were randomized into 4 groups (A, B, C and D) of 6 animals each. Pregnant rats in groups A, B and C were maintained in poorly ventilated rooms and exposed to 20 ml (soaked in cotton wool, 2.5 g), each of HAIA, HAIB and HAIC respectively, for 30 minutes. The pregnant rats in group A (sham control) were placed in well ventilated room far away from the other groups. The exposure to the household aerosol insecticides commenced on Day 7 of pregnancy and lasted till Day 20 of pregnancy. While on these treatment periods, the animals were maintained on pelleted feed (Top Feed, a division of Premier Feed Mill Company Limited, Ibadan, Nigeria) and tap water, *ad libitum*. The pregnant rats in each group were observed for any clinical signs of toxicity during the exposure period. All the animals in this study were handled strictly in accordance with the Guidelines on the Care

and Use of Laboratory Animals of the National Research Council [12].

Examination of foetal outcome and preparation of serum and tissue homogenates

The procedures described by Yakubu and Bukoye [11] and Yakubu *et al.* [13] and were adopted. Under ether anaesthesia, the neck of the pregnant rats were swiftly shaved to remove the fur and to expose the jugular veins. The jugular veins after being slightly displaced from the neck were cut to allow the blood to drip into the EDTA coated sample vials and plain sample bottles. The blood contained in the EDTA sample vials were swirled gently (to prevent blood clotting) while the blood (5 ml) in the plain sample bottles were left to clot for 30 minutes. This was then centrifuged at $126 \times g$ for 15 minutes with a Uniscope Laboratory Centrifuge (model SM800B, Surgifriend Medicals, Essex, England). The serum was then aspirated with Pasteur pipettes into clean, dry, sample vials, and kept frozen overnight before being used for the enzyme assay [11]. Prior to preparing the serum, the pregnant rats were immediately laparotomised after collecting the blood. The foetal outcome of total number of foetuses, live foetuses, dead foetuses and average weight of the foetuses were determined. Thereafter, the liver, kidney and ovary were blotted with blotting paper, weighed and homogenized in 0.25M sucrose solution (1:5 w/v). The homogenates were transferred into specimen bottles and kept frozen for 24 hours before being used for the analyses.

Determination of biochemical parameters

The biochemical parameters evaluated in this study included alkaline phosphatase (ALP) [14], alanine aminotransferase and aspartate aminotransferase [15], total protein [16], total bilirubin and conjugated bilirubin [17], albumin [18], Na^+ , K^+ and Cl^- [19], urea [20] and creatinine [21]. Globulin was estimated

as the differences between total protein and albumin [19]. The procedures described by Misra [22], Hadwan [23], Mohnadas *et al.* [24] Zheng *et al.* [25] and Varshney and Kale [26] were used for the antioxidant parameters of superoxide dismutase (SOD), catalase, glutathione peroxidase (GPX), reduced glutathione (GRed) and malondialdehyde respectively.

Determination of haematological parameters

The packed cell volume (PCV), red blood cell (RBC), white blood cell (WBC), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and platelet were quantified using the automated haematology analyzer.

Histological examination

The histoarchitectural changes in the liver of pregnant rats were evaluated by adopting the procedures of slicing, fixing, dehydrating, clearing, embedding and staining (haematoxylin and eosin) as described by Wei *et al.* [27]. The photomicrographs were examined a light microscope (OLYMPUS, Model: CX21, New York Microscope Company Inc., New York) at 400. The images were captured with a Canon! Image Folio package software (Model: Powershot A2500, Japan).

Statistical analysis

Data which were mean $\pm$ SEM of 6 determinations, were subjected to One-way Analysis of variance, and followed by Tukey's Multiple Comparison Test at $P < 0.05$, using GraphPad 9.0 Software (GraphPad Software Inc., San Diego, California).

RESULTS

Exposure of the pregnant rats to HAIA and HAIC produced dull eyes with loss of fur whereas HAIB induced red eyes only (Table

1). All the household insecticides (HAIA, HAIB and HAIC) significantly ($P<0.05$) reduced the total number of foetuses and number of life foetus (HAIA, HAIB and HAIC by 30.8%, 15.4% and 32.7% respectively) with HAIC producing the most pronounced reduction (Table 1). In contrast, the HAIA, HAIB and HAIC significantly ($P<0.05$) increased the average weight of the foetuses with the most pronounced increase by HAIB. No foetal death was recorded in the pregnant rats that were exposed to HAIB and HAIC whereas HAIA produced three foetal death (Table 1).

All the insecticides (HAIA, HAIB and HAIC) significantly ($P<0.05$) reduced the activities of ALP, AST and ALT in the liver of pregnant rats with HAIB producing the most outstanding reduction in the liver ALP activity and liver AST activity whereas the most pronounced reduction in liver ALT activity was by HAIC (Table 2). Furthermore, the reduction in all the liver enzymes (ALP, ALT and AST) were accompanied by corresponding increases in the serum enzymes (ALP, ALT and AST) with HAIB producing the most remarkable increase in all the serum enzymes (Table 2). With respect to the sham control, all the HAIA, HAIB and HAIC significantly ($P<0.05$) reduced the total protein, albumin and globulin content in the serum of the pregnant rats whereas the serum total bilirubin and conjugated bilirubin contents were significantly ($P<0.05$) increased by the HAIA, HAIB and HAIC (Table 2). The reduction in total protein was most pronounced in the pregnant rats that were exposed to HAIB whereas HAIA produced the most remarkable decrease in serum albumin and globulin contents of the pregnant rats. Furthermore, the increases in total bilirubin and conjugated bilirubin levels of the pregnant rats were most remarkable in the animals exposed to HAIC (Table 2).

Compared with the sham control, only HAIA reduced the kidney ALP activity whereas it

was only HAIC that increased the kidney ALT activity (Table 3). The kidney AST activity was not significantly ($P>0.05$) altered by all the household aerosol insecticides (HAIA, HAIB and HAIC) when compared with the sham control (Table 3). Furthermore, the activities of ALP, AST and ALT in the serum of the pregnant rats were not significantly ($P>0.05$) altered by the HAIA, HAIB and HAIC when compared with the sham control (Table 3). Exposure of the pregnant rats to HAIA, HAIB and HAIC significantly ($P<0.05$) reduced the levels of Na^+ , K^+ and Cl^- in the serum of the pregnant rats whereas HAIA, HAIB and HAIC did not significantly ($P>0.05$) alter the levels of urea and creatinine in the pregnant rats (Table 3). The most remarkable reduction in the levels of Na^+ , K^+ and Cl^- were produced by pregnant rats that were exposed to HAIC, HAIA and HAIB respectively (Table 3).

Compared with the sham control, exposure of the pregnant rats to HAIA, HAIB and HAIC significantly ($P<0.05$) reduced the levels of SOD, catalase, GPX and GRed in the liver, kidney and ovary of the pregnant rats (Figures 1-4). The levels of reduction in the SOD, GPX and GRed by HAIA, HAIB and HAIC in the liver, kidney and ovary were not significantly ($P>0.05$) different from each other. In contrast, the reduction in the activity of catalase in the kidney of pregnant rats was most remarkable in the animals exposed to HAIB (Figure 3). The malondialdehyde contents in the liver, kidney, ovary and serum of pregnant rats were significantly ($P<0.05$) increased after the exposure to HAIA, HAIB and HAIC (Figure 5). However, the increase in the malondialdehyde content of each of the organs and serum were not significantly ($P>0.05$) different from each other after exposure to HAIA, HAIB and HAIC (Figure 5).

Exposure of the pregnant rats to HAIA, HAIB and HAIC significantly ($P<0.05$) increased the levels of WBC, PCV and RBC with HAIB

producing the most remarkable increase (Table 4). In contrast, the HAIA, HAIB and HAIC reduced significantly ($P < 0.05$) the level of MCV with HAIA producing the most pronounced reduction in MCV (Table 4). Furthermore, in comparison to sham control, the levels of MCH and platelets were not significantly ($P > 0.05$) altered in the pregnant rats after exposure to HAIA, HAIB and HAIC (Table 4).

The architecture of the liver of pregnant rat that constituted the sham control revealed lobular architecture, central vein and portal tracts that were within normal histology (Plate 1) whereas exposure of pregnant rats to HAIA, HAIB and HAIC deranged the lobular architecture of the hepatocytes, central veins and portal tracts of the liver of the animals (Plates 2-4).

DISCUSSION

Exposure to household aerosol insecticides has been associated with many hazardous effects [28]. The widespread uses of pyrethroids containing insecticides and the corresponding increase in human exposure have led to toxicological interest [29].

Yakubu *et al.* [30] have reported that squinted and dull eyes are useful signs of toxicity whereas erection of fur is a physical indicator of stress. Therefore, the dullness of the eyes with loss of fur exhibited by the HAIA- and HAIC-exposed pregnant rats are indications of toxicity and stress whereas the red eyes produced by the pregnant rats that were exposed to the HAIB suggests toxicity. All these further indicate that the household aerosol insecticides have toxic implications on the pregnant rats. The toxic effects on eyes caused by aerosol insecticides exposure via inhalation, ingestion, dermal contact and ocular exposure are consistent with previous reports [31].

Changes in organ body weight either absolute or relative after administration of a xenobiotic has been reported to reflect the harmful effects

of the chemical compound [32]. The increase in the average weight of the foetus in HAIA, HAIB and HAIC could be associated with the effects of the dichlovos and permethrin contents of the household aerosol insecticides whilst the reduced number of foetuses and life foetuses by HAIA (30.8%), HAIB (15.4%) and HAIC (32.7%) may be attributed to the toxic effect of pyrethroids containing insecticides. The foetal death by the HAIA suggests fetotoxicity of the household aerosol insecticide.

The determination of the activities of cellular enzymes such as alkaline phosphatase, acid phosphatase, alanine aminotransferase, aspartate aminotransferases and gamma glutamyl transferase can be used useful pointer to cellular integrity of the organ, functional status of the organ and indices of damage after the administration of chemical compounds [33]. Hepatotoxic compounds inflict damage on the hepatocytes by reducing the activity of the enzymes which eventually leaked from the cells into the serum resulting in their elevation in the biological fluids and these enzymes are leaked out into serum and shows increased activities [33,34]. In specific terms, ALP is a marker for damage to the plasma membrane whilst the aminotransferases (ALT and AST) are useful biomarkers cytolysis [35]. Therefore the reduction in the liver ALP activities which was picked up in the serum suggests disruption of the cell membrane via membrane permeability, thus accounting for the rise in the serum ALP activity. Furthermore, the loss of the aminotransferases is quite understandable since a disruption of the plasma membrane will consequentially result in leakage of the cytosolic enzymes into the serum that manifested as elevation in the activity of the aminotransferases in the serum of the pregnant rats. The implication is that all the insecticides (HAIA, HAIB and HAIC) have displayed similar pattern of disruption of the plasma membrane with HAIB being the

most toxic on the liver ALP activity and liver AST activity and HAIC most toxic on the liver ALT activity. These findings with respect to the elevation in the serum ALP, ALT and AST agree with the report by Al-sarar *et al.* [36] in pesticides-exposed workers. Studies have demonstrated that pyrethroid-containing insecticides raise the serum enzymatic activity of AST, ALT, and ALP, hence, the serum enzyme elevation may be due to pyrethroid-induced toxicity [37].

Evaluation of total protein, albumin and globulin contents of experimental animals after the administration of chemical compounds can be used to predict the effect of the compound on the synthetic and the secretory capacity of the liver whereas the assessment of the bilirubin content could be a pointer to the effects of a chemical compound on the excretory functions of the liver [33,35]. Therefore, the reduction in the concentration of total protein, albumin and globulin of the pregnant rats by the HAIA, HAIB and HAIC suggests that the household aerosol insecticides have impacted negatively on the secretory and synthetic roles of the liver as a consequence of hepatocellular injury earlier opined by the loss of the hepatic enzymes [33]. It will be interesting to note that HAIB tremendously affected the ability of the liver to synthesize proteins whereas HAIA negatively imparts on the ability of the liver to synthesize and secrete the albumin and globulin. Bilirubin is considered to be a true test or confirmatory test for hepatotoxicity, as it reflects the ability of the liver to carry out secretory role of releasing bilirubin into the bile. Therefore [38], the increase in the level of bilirubin (total and conjugated) suggests enhanced excretory function of the liver with HAIC exhibiting the most positive effect.

The reduction of ALP activity in the kidney of the pregnant rats only by the HAIA without corresponding increase in the serum enzyme suggests that HAIA might have inhibitory activity *in situ* on the kidney enzyme. In

contrast the increase in kidney ALT by HAIC might portend *de novo* synthesis of the enzyme molecule by the household aerosol insecticide. All of these are indications of differential effects on the activities of the enzymes in the kidney and serum of the animals. Creatinine and urea are the standard indices of renal function and the most reliable clinical estimates of glomerular filtration rates [39]. The absence of an effect on the creatinine and urea levels after exposure of the pregnant rats to the household aerosol insecticides might indicate that the insecticides did not adversely affected the functioning of the nephron at the glomerular levels. Since the reabsorption of electrolytes like Na^+ , K^+ and Cl^- takes place in the renal tubules, therefore, the reduction in the levels of these electrolytes after exposure of pregnant rats to HAIA, HAIB and HAIC may suggests impairment in the normal functioning at the tubular region of the kidney leading to tubular dysfunction. This reduction in the electrolytes might be due to the effect of pyrethroids as they have been implicated as electrolyte pump modulators. It appears that the household aerosol insecticides have selective effects on the kidney sparing the glomerulus and inducing tubular dysfunction. The findings with respect to the serum urea and creatinine levels in this study are similar to that of Al-Sarar *et al.* [36] who found no change in the levels of urea and creatinine of adults when exposed to insecticides containing pyrethroids.

Antioxidant enzymes are used by the organisms as natural endogenous protection against the generation of reactive oxygen species and maintain cellular homeostasis by removing reactive oxygen species [40]. The enzymes involved in antioxidant reactions are the superoxide dismutase, glutathione peroxidase and catalases [41]. Exposure to insecticides has been linked to oxidative stress as it causes lipid peroxidation and depletes a number of antioxidant enzymes, including

GRed, GPX, catalase, and SOD [42]. Therefore, the reduction in the levels of SOD, catalase, GPX and GRed in the liver, kidney and ovary of the pregnant rats in the present study may be due to accumulation of lipid peroxidation products by the pesticides. This is more evident from the elevated levels of malondialdehyde in the liver, kidney and ovary of pregnant rats in the present study. Depletion of antioxidant enzyme activity could also be due to the direct effect on the enzymes by the household aerosol insecticides and in the process inducing the generation of reactive oxygen species, or direct inhibition of the enzymes by the household aerosol insecticides [43].

Blood can act as a pathological and physiological indicator of animal health [44]. The elevated level of PCV and RBC could be attributed to the production capacity of the bone marrow that outweighs the destruction of the red blood cells by the HAIA, HAIB and HAIC. It may also be due to the one of the by-products of insecticide formulations containing pyrethroid compounds, the cyanide, which is known to cause reduction in oxygen carrying capacity of red blood cells leading to reduced metabolism. This reduction of oxygen will enhance erythropoietin which in turn will stimulate the bone marrow to produce RBC. Garba *et al.* [43] Schoeinig [45], Parker *et al.* [46] similarly observed increase in RBC and PCV in rats exposed to pyrethroids containing insecticides. Furthermore, the increase in WBC after exposure to the household aerosol insecticides may suggest leucocytosis [47]. The reduction in MCV and absence of effect on MCH and platelets suggest differential effects on the blood parameters by the household aerosol insecticides.

Tissue histological alterations are the result of a chemical, physical, mechanical, or inflammatory intrusion [48]. Therefore, the deranged lobular architecture of the hepatocytes, central veins and portal tracts of

the liver of pregnant rats after exposure to HAIA, HAIB and HAIC suggests structural toxicity. The findings here is similar to that of Armalina *et al.* [48] who reported necrosis in portal areas, hydropic degeneration of lobules, infiltration of inflammatory cells, and fibrosis in the portal area, causing a sinusoidal portal to widen in pregnant rats exposed to D-Allethrin in mosquito repellent.

CONCLUSION

The study concluded that household aerosol insecticides exhibited functional and structural toxicities in the pregnant rats via oxidative means with household aerosol insecticide B (HAIB) producing the most pronounced outcome.

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Table 1: Clinical signs of toxicity and foetal outcome of pregnant Wistar rats after exposure to some commonly used household aerosol insecticides

Parameters	Sham Control	HAIA	HAIB	HAIC
Clinical signs of toxicity	None	Dull eyes and Loss of fur	Red eyes	Dull eyes and loss of fur
Total number of foetuses	10.40 ± 0.51 ^a	7.20 ± 0.66 ^b	8.80 ± 0.37 ^c	7.00 ± 1.05 ^b

Average weight of foetuses (g)	3.49 ± 0.29 ^a	3.66 ± 0.12 ^a	4.61 ± 0.15 ^b	3.67 ± 0.17 ^a
Number of life foetuses	10.40 ± 0.51 ^a	7.20 ± 0.66 ^b	8.80 ± 0.37 ^c	7.00 ± 1.05 ^b
Number of dead foetuses	0	3	0	0

Values are mean ± SEM of six determinations. Test values carrying different superscripts from that of the sham control across the row are significantly different (p<0.05)

HAIA = Household aerosol insecticide A

HAIB = Household aerosol insecticide B

HAIC = Household aerosol insecticide C

Table 2: Liver function indices of pregnant Wistar rats after exposure to some commonly used household aerosol insecticides

Parameters	Sham Control	HAIA	HAIB	HAIC
Liver ALP (U/L)	4.23 ± 0.32 ^a	2.59 ± 0.38 ^b	2.23 ± 0.32 ^c	2.74 ± 0.21 ^b
Serum ALP (U/L)	1.21 ± 0.03 ^a	1.50 ± 0.03 ^b	1.73 ± 0.01 ^b	1.42 ± 0.05 ^b
Liver AST (U/L)	6.21 ± 0.24 ^a	4.06 ± 0.24 ^b	3.07 ± 0.14 ^c	4.92 ± 0.15 ^b
Serum AST (U/L)	0.71 ± 0.01 ^a	1.06 ± 0.02 ^b	1.09 ± 0.02 ^b	0.97 ± 0.03 ^b
Liver ALT (U/L)	6.31 ± 0.34 ^a	4.11 ± 0.2 ^b	4.07 ± 0.34 ^c	4.37 ± 0.15 ^b
Serum ALT (U/L)	0.65 ± 0.02 ^a	1.02 ± 0.01 ^b	1.09 ± 0.02 ^b	1.03 ± 0.01 ^b
Total Protein (mg/dL)	16.54 ± 0.32 ^a	11.83 ± 0.24 ^b	10.89 ± 0.22 ^b	12.82 ± 0.25 ^b
Albumin (g/L)	53.02 ± 0.32 ^a	43.87 ± 0.45 ^b	47.95 ± 0.82 ^c	45.98 ± 0.40 ^d
Globulin (g/L)	36.48 ± 0.35 ^a	32.04 ± 0.28 ^b	37.06 ± 0.98 ^b	33.06 ± 0.24 ^b
Total bilirubin (µmol/L)	45.98 ± 1.02 ^a	53.91 ± 1.28 ^b	55.52 ± 1.02 ^b	56.18 ± 0.03 ^c
Conjugated bilirubin (µmol/L)	51.10 ± 0.02 ^a	54.12 ± 0.04 ^b	53.32 ± 1.01 ^b	54.22 ± 0.05 ^b

Values are mean ± SEM of six determinations. Test values carrying different superscripts from that of the sham control across the row are significantly different (p<0.05)

HAIA = Household aerosol insecticide A; **HAIB** = Household aerosol insecticide B;

HAIC = Household aerosol insecticide C; **ALP** = Alkaline phosphatase;

AST = Aspartate aminotransferase; **ALT** = Alanine aminotransferase

Table 3: Kidney function indices of pregnant Wistar rats after exposure to some commonly used household aerosol insecticides

Parameters	Sham Control	HAIA	HAIB	HAIC
Kidney ALP (U/L)	8.05 ± 0.00 ^a	7.98 ± 0.02 ^b	7.98 ± 0.03 ^a	8.00 ± 0.01 ^a
Serum ALP (U/L)	1.23 ± 0.02 ^a	1.24 ± 0.01 ^a	1.23 ± 0.02 ^a	1.24 ± 0.02 ^a
Kidney AST (U/L)	6.21 ± 0.21 ^a	6.26 ± 0.04 ^a	6.27 ± 0.04 ^a	6.22 ± 0.05 ^a
Serum AST (U/L)	0.21 ± 0.02 ^a	0.23 ± 0.02 ^a	0.23 ± 0.01 ^a	0.22 ± 0.01 ^a
Kidney ALT (U/L)	4.10 ± 0.03 ^a	4.12 ± 0.03 ^a	4.12 ± 0.04 ^a	4.92 ± 0.15 ^b

Serum ALT (U/L)	0.31 ± 0.01 ^a	0.30 ± 0.01 ^a	0.32 ± 0.01 ^a	0.30 ± 0.01 ^a
Creatinine (mg/dL)	8.74 ± 0.24 ^a	8.70 ± 0.29 ^a	8.78 ± 0.21 ^a	8.70 ± 0.29 ^a
Urea (mg/dL)	54.14 ± 0.77 ^a	54.02 ± 0.73 ^a	54.29 ± 0.77 ^a	54.22 ± 0.72 ^a
Na⁺ (mg/dL)	1.22 ± 0.44 ^a	0.62 ± 0.01 ^b	1.04 ± 0.13 ^c	0.60 ± 0.02 ^b
K⁺ (mg/dL)	1.55 ± 0.13 ^a	0.37 ± 0.09 ^b	0.72 ± 0.09 ^c	0.38 ± 0.04 ^b
Cl⁻ (mg/dL)	1.73 ± 0.11 ^a	0.81 ± 0.08 ^b	0.77 ± 0.09 ^b	0.75 ± 0.08 ^b

Values are mean ± SEM of six determinations. Test values carrying different superscripts from that of the sham control across the row are significantly different (p<0.05)

HAIA = Household aerosol insecticide A; **HAIB** = Household aerosol insecticide B;

HAIC = Household aerosol insecticide C; **ALP** = Alkaline phosphatase;

AST = Aspartate aminotransferase; **ALT** = Alanine aminotransferase

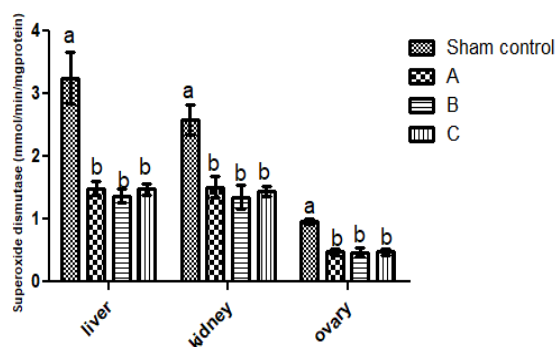


Figure 1: Superoxide dismutase activity in the liver, kidney and ovary of pregnant Wistar rats exposed to some commonly used household insecticide formulation

Values are mean ± SEM of six determinations; Bars with alphabets that are different from that of the sham control are significantly different (p<0.05)

A =HAIA=Household aerosol insecticide A;

B =HAIB=Household aerosol insecticide B;

C =HAIC = Household aerosol insecticide C

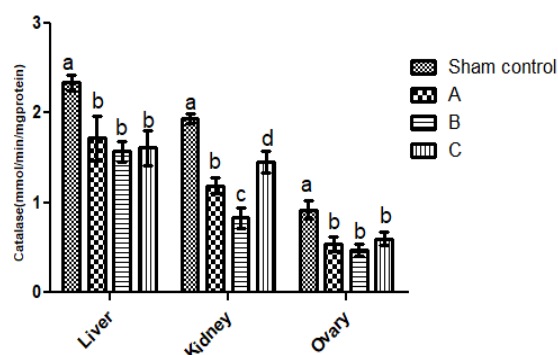


Figure 2: Catalase activity in the liver, kidney and ovary of pregnant Wistar rats exposed to some commonly used household insecticide formulation

Values are mean ± SEM of six determinations; Bars with alphabets that are different from that of the sham control are significantly different (p<0.05)

A =HAIA=Household aerosol insecticide A;

B = HAIB=Household aerosol insecticide B;

C = HAIC =Household aerosol insecticide C

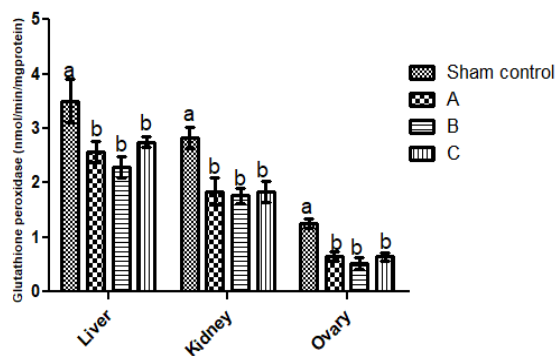


Figure 3: Glutathione peroxidase activity in the liver, kidney and ovary of pregnant Wistar rats exposed to some commonly used household insecticide formulation

Values are mean $\pm$ SEM of six determinations; Bars with alphabets that are different from that of the sham control are significantly different ($p < 0.05$)

A =HAIA=Household aerosol insecticide A;
B =HAIB =Household aerosol insecticide B;
C =HAIC = Household aerosol insecticide C

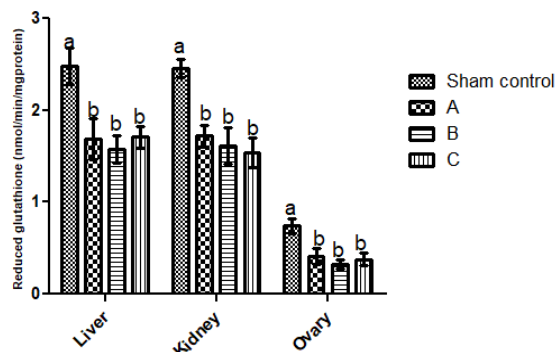


Figure 4: Reduced glutathione levels in the liver, kidney and ovary of pregnant Wistar rats exposed to some commonly used household insecticide formulation

Values are mean $\pm$ SEM of six determinations; Bars with alphabets that are different from that of the sham control are significantly different ($p < 0.05$)

A =HAIA=Household aerosol insecticide A;
B =HAIB =Household aerosol insecticide B;
C = HAIC =Household aerosol insecticide C

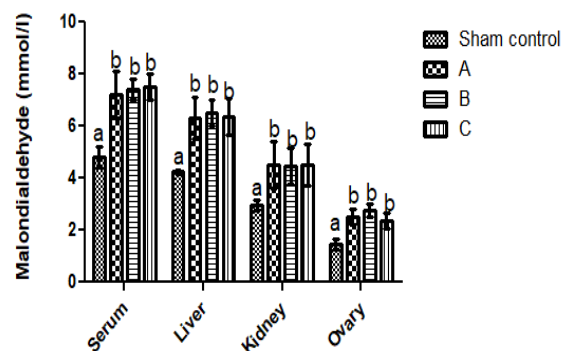


Figure 5: Malondialdehyde levels in the liver, kidney and ovary of pregnant Wistar rats exposed to some commonly used household insecticide formulation

Values are mean $\pm$ SEM of six determinations; Bars with alphabets that are different from that of the sham control are significantly different ($p < 0.05$)

A =HAIA=Household aerosol insecticide A;
B =HAIB =Household aerosol insecticide B;
C =HAIC = Household aerosol insecticide C

Table 4: Haematological parameters of pregnant Wistar rats after exposure to some commonly used household aerosol insecticides

Parameters	Sham Control	HAIA	HAIB	HAIC
WBC ($10^3/\mu\text{L}$)	13.74 ± 1.29^a	15.76 ± 1.64^b	23.74 ± 4.41^c	15.10 ± 1.72^b
PCV (%)	41.85 ± 1.93^a	47.73 ± 1.54^b	48.95 ± 3.58^a	46.60 ± 1.06^b
RBC ($10^6/\mu\text{L}$)	6.33 ± 0.20^a	7.23 ± 0.36^b	7.64 ± 0.57^b	7.50 ± 0.65^b
MCV (fg)	66.13 ± 2.14^a	60.75 ± 1.84^b	61.73 ± 0.90^b	60.90 ± 1.23^b
MCH (pg)	19.05 ± 0.62^a	18.93 ± 0.67^a	18.93 ± 0.22^a	18.83 ± 0.41^a
Platelets ($10^6/\mu\text{L}$)	0.77 ± 0.15^a	0.85 ± 0.06^a	0.70 ± 0.17^a	1.01 ± 0.11^a

Values are mean $\pm$ SEM of six determinations. Test values carrying different superscripts from that of the sham control across the row are significantly different ($p < 0.05$)

HAIA = Household aerosol insecticide A; **HAIB** = Household aerosol insecticide B;
HAIC = Household aerosol insecticide C; **WBC** = White blood cell;
PCV = Packed cell volume; **RBC** = Red blood cell; **MCV** = Mean corpuscular volume;
MCH = Mean corpuscular haemoglobin



Plate 1: Cross section of the liver of pregnant Wistar rat that was exposed to air (H&E; x400)

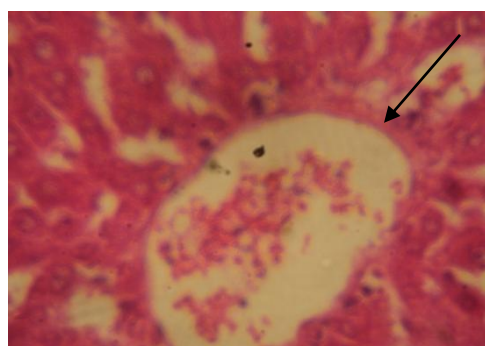


Plate 2: Cross section of the liver of pregnant Wistar rat exposed to commonly used household aerosol insecticide A (HAIA) (H&E; x400)

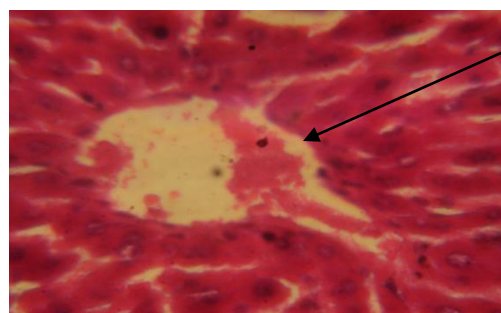


Plate 3: Cross section of the liver of pregnant Wistar rat exposed to commonly used household aerosol insecticide B (HAIB) (H&E; x400)



Plate 2: Cross section of the liver of pregnant Wistar rat exposed to commonly used household aerosol insecticide C (HAIC) (H&E; x400)