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HEMATOLOGICAL EVALUATION OF ABINO RATS ORALLY ADMINISTERED PYRETHROIDS FORMULATION

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Abstract

Aim: This study was carried out to determine the effect of Mortein on some hematological parameters.

Methods: Fifty adult Albino rats (150- 200g) were acquired from the animal house of the College of Health Sciences, Delta State University Abraka and placed into four groups. Groups 1- 3 contained 15 rats each, while 4 contained 5 rats. Groups 1-3 were further sub-divided into 3 sub-groups of 5 rats each. The rats in Groups 1-3 were treated with 3 different doses of M for 7, 21 and 40 days respectively while Group 4 was given water as placebo. The rats were later sacrificed and blood samples were collected for hematological analysis. The data generated were presented as mean and standard error of mean. Significant differences were obtained by One Way Analysis of Variance at ($p \leq 0.05$) with the aid of SPSS version 21.

Results: Significant decrease in total white blood cell count was observed in rats treated with M in dose and time-dependent manner when compared to rats of the control group. There was also a consequent time-dependent significant decrease ($p \leq 0.05$) of lymphocyte, significant increase ($p \leq 0.05$) of neutrophil and slight increase of monocyte when compared to the control. Subsequently, PCV significantly ($p \leq 0.05$) reduced in treated rats compared to the control.

Conclusion: Over exposure to the insecticide, Mortein reduces total white blood cells and packed cell volume.

Keywords: Mortein, Albino Rats, Hematological, Blood, Pyrethroids

INTRODUCTION

Pyrethroids are chemically synthesized with synergist to potentiate their pesticidal effects. They are synthetic derivatives of the naturally occurring pyrethrins found as insecticidal components gotten as pyrethrum extracts of chrysanthemum flower. Presently, they are among the most modern groups of pesticides most frequently used domestically in Nigeria. Some studies have previously reported that blood is one of the major sites of Pyrethroids metabolism in laboratory animals (Anandonet al., 2006; Mirfazaelianet al., 2006) as serum of rats contained carboxylesterase which has the ability of breaking down Pyrethroids. Since this enzyme was found only in the serum, the possibility of other organs of this system to suffer from toxicity may arise. Alteration in erythrocytic indices had been reported in fishes subjected to environmental stressors like pesticides and heavy metals (Bhatia et al., 2004; Johal and Grewal, 2004; Gill and Pant, 1987). Several studies have also documented a reduced RBC, PCV and Hemoglobin content in rats exposed to pyrethroids insecticide (Kamal et al., 2007; Yousefet al., 2003;Shakooriet al., 1988). In one study by Elbendary et al., (2004) which involved the exposure of male mice to two different pyrethroids pesticides, they observed

disorganization and depletion of lymphocyte and hemorrhages in the mice spleen. Some other studies have demonstrated that many of the commonly used pesticides can suppress the immune system, thereby leading to increase toll of infectious agents. (M) Pesticide is composed of the following Pyrethroids: d-trans Allethrin (0.10%w/w), Imiprothrin (0.02%w/w) and d-phenothrin (0.03%w/w) as active ingredients. Although there is a common perception by users about the general safety of these chemicals, several studies have reported their toxic effects (Sanghaet al., 2011; Inayatet al., 2007; Kamal et al., 2007). It is also believed that insecticides generally have adverse effects with prolonged exposure mostly when they are mixed in a way to complement their mode of action such as observed in the pesticide of choice. According to Gosselinet al., (1984), some of these agents are often formulated with oils or petroleum distillates and packaged in combination with synergists such as piperonylbutoxide (PBO) and n-octylbicyclo heptene dicarboximide thereby rendering them more toxic and prone to causing diseases. This study was consequently carried out to determine the hematological effects of M pesticide on PCV, WBC and RBC counts in adult albino rats.

MATERIALS AND METHODS

Animals Selection

Fifty adult Albino rats of both sexes were acquired from the animal house of the Faculty of Basic Medical Sciences, Delta State University Abraka. The rats were healthy and weighed 150-250g prior to the start of the treatment and were allowed to acclimatize for two weeks under favorable housing conditions and fed with rat chaw and water ad libitum.

Test Compound Selection

The chemical product of pyrethroids composed of Allethrin, Imiprothrin and Phenothrin was manufactured by Rekitt Benckiser Nigeria Limited, Agbara Ogun State and registered with National Agency for Food and Drug Administration Control (NAFDAC) with number 048724.

Pesticide Preparation

The can of the insecticide was placed in cold flax containing ice blocks for few hours thereby allowing the gaseous content to be liquefied and thereafter spewed into a measuring cylinder.The insecticide was subsequently diluted with Olive oil to give different grades (25, 50%, and 75%) of a stock solution of the insecticide and three different doses of the chemical were calculated and administered orally to the animals as shown in Table 1.

Grouping and Treatment of Rats

The fifty rats were randomly divided into four groups and were housed in compartmented metallic cages in a room, where temperature was satisfactory. Groups' I-III (treatment groups) containing 15 rats each were further subdivided into three sub-groups each of five animals respectively. Prior to the time of administration of the drug, the male rats were kept in different compartments from those of the females in order to avoid mating. The rats of normal control group (IV) were given tap water as placebo; whereas the three calculated doses of the insecticide were administered to rats of Groups I-III via oral route by the use of improvised cannula.

Table 1: Summary of treatment of the animals

Groups Treatment groups		No. of Animals	Treatment (3ml/Kg/Bw)	Dosage (Mg/Kg/Bw)	Duration In Days
1	D	5	75% of M solution	2250	7
	E	5	50% of M solution	1500	7
	F	5	25% of M solution	750	7
2	X	5	75% of M solution	2250	21
	Y	5	50% of M solution	1500	21
	Z	5	25% of M solution	750	21
3	A	5	75% of M solution	2250	40
	B	5	50% of M solution	1500	40
	C	5	25% of M solution	750	40
4	Ctrl	5	Tap water	3	40

Euthenization and Collection of Blood Samples

At the end of the respective duration of treatment for each group (7, 21 and 40 days), the rats in each group were euthanized by cervical dislocation, and blood was collected from the inferior venae cava with 5ml syringe into EDTA bottles for PCV, TWBC, and differential WBC using standard manual hematological techniques.

RESULTS

Table 2: Full blood count

Gp	Sub-Gp	Statistics	TWBC (mm3)	Neutrophil (%)	Lymphocyte (%)	Eosinophil (%)	Monocyte (%)	PCV (mm3)
1	D	Mean ±SD	$^{b}1 \times 10^{3} \pm 5 \times 10^{2\downarrow}$	$^{b}48.67 \pm 1.15^{\uparrow}$	$^{b}51.33 \pm 1.16^{\downarrow}$	$^{a}0.0 \pm 0.0$	$^{a}0.0 \pm 0.0$	$^{b}46 \pm 2$
	E		$^{c}7.5 \times 10^{3} \pm 5 \times 10^{2\downarrow}$	$^{c}65.33 \pm 5.03^{\uparrow}$	$^{c}34.67 \pm 5.03^{\downarrow}$	$^{a}0.0 \pm 0.0$	$^{a}0.0 \pm 0.0$	$^{b}43 \pm 1.7^{\downarrow}$
	F		$^{d}8 \times 10^{3} \pm 5 \times 10^{2\downarrow}$	$^{b}48.67 \pm 1.15^{\uparrow}$	$^{b}51.33 \pm 1.16^{\downarrow}$	$^{a}0.0 \pm 0.0$	$^{a}0.0 \pm 0.0$	$^{b}45 \pm 2^{\downarrow}$
2	X	Mean ± SD	$^{d}8 \times 10^{3} \pm 2 \times 10^{3\downarrow}$	$^{a}40.67 \pm 20.03$	$^{a}59.33 \pm 20.03$	$^{a}0.0 \pm 0.0$	$^{a}0.0 \pm 0.0$	$^{b}43 \pm 3.61^{\downarrow}$
	Y		$^{e}1.4 \times 10^{3} \pm 3.2 \times 10^{3\downarrow}$	$^{d}38 \pm 3.46^{\downarrow}$	$^{a}61.33 \pm 2.31$	$^{a}0.0 \pm 0.0$	$^{b}0.67 \pm 1.16^{\uparrow}$	$^{a}48 \pm 2^{\downarrow}$
	Z		$^{a}1 \times 10^{4} \pm 1 \times 10^{3}$	$^{e}43.33 \pm 8.33^{\uparrow}$	$^{b}51.33 \pm 11.37^{\downarrow}$	$^{a}0.0 \pm 0.0$	$^{c}4.67 \pm 6.43^{\uparrow}$	$^{b}44.33 \pm 1.53^{\downarrow}$
3	A	Mean ± SD	$^{c}7.3 \times 10^{3} \pm 1.5 \times 10^{3\downarrow}$	$^{a}41.33 \pm 17.01$	$^{a}58.67 \pm 17.01$	$^{a}0.0 \pm 0.0$	$^{a}0.0 \pm 0.0$	$^{b}44 \pm 4^{\downarrow}$
	B		$^{e}1.4 \times 10^{3} \pm 2.3 \times 10^{3\downarrow}$	$^{a}39.33 \pm 3.06$	$^{a}60 \pm 2$	$^{a}0.0 \pm 0.0$	$^{b}0.67 \pm 1.16^{\uparrow}$	$^{b}44.33 \pm 0.58^{\downarrow}$
	C		$^{f}9.7 \times 10^{3} \pm 2.1 \times 10^{3\downarrow}$	$^{e}43.33 \pm 3.06^{\uparrow}$	$^{b}53.33 \pm 11.37^{\downarrow}$	$^{b}0.67 \pm 1.16^{\uparrow}$	$^{c}3.33 \pm 4.16$	$^{b}43.67 \pm 1.53^{\downarrow}$
4	control	Mean ± SD	$^{a}1.03 \times 10^{4} \pm 3.8 \times 10^{2}$	$^{a}40.67 \pm 1.15$	$^{a}59.33 \pm 1.16$	$^{a}0.0 \pm 0.0$	$^{a}0.0 \pm 0.0$	$^{a}49.33 \pm 1.16$

Means bearing same alphabetical superscript does not differ significantly ($p \leq 0.05$). Downward arrow (↓) means a decrease, while an upward arrow (↑) is an increase.

Key: Gp- Group

DISCUSSION

Result from statistical analysis revealed a significant ($p \leq 0.05$) decrease in total white blood (TWBC) count (Leucopenia) of the treated animals when compared to that of the control group. These decreases observed, were both dose and time dependent. The resultant leucopenia observed in the study is mainly a reflection of the significant decrease of lymphocyte number (lymphocytopenia) which may have led to a consequent opportunistic infections as indicated by the significant increase (neutrophilia) later observed from our differential white blood analysis . Our result differs from that of Sakaet al.,

(2011) which reported that Pyrethroids did not influence TWBC in rats. This could be as a result of the inhalation-based treatment or the small doses used in their experiment. Pyrethroids absorption in oral route administration has been observed to be more effective and systemic toxicity high compared to both inhalation and dermal route (Reigartet al., 1999). The increase in oral systemic toxicity could be as a result of the lipophilic nature of Pyrethroids enhanced by bile secretion into the small intestine. Our finding is consistent with that of Tulinska et al., (1995) as well as Blaylock et al., (1995) which reported depressed cellular immune response respectively. The decreased TWBC count was reflected in turn by time-dependent decrease percentages of lymphocyte in the treated rats compared to those of the control groups. Lymphocytes are known to help build immunity by preventing the development of harmful cells; therefore, decreased percentage of lymphocyte in the treated rats may be indicating that their resistance to infection is low. This suggests that Pyrethroids may predispose rats to opportunistic infections by suppressing their immune system (Tulinskaet al., 1995). The percentage of neutrophil consequently increased as a result of sudden invasion by pathogens (Sembulingam and Sembulingam, 2006) or as a result of possible inflammations caused by active mast cells (Guyton and Hall, 2011). The later has been observed in kidneys of rats treated with same agent (Odokuma and Iteire, 2015). Furthermore, eosinophils were almost absent in both treated and control rats, while percentage of monocyte slightly increased in the rats of both acute and chronic groups but not in the sub-acute and control groups. The differences in the percentage monocyte may be due to a complimentary response to possible chronic infection likely to be suffered by rats treated with Pyrethroids acutely and chronically. Therefore we can infer that there was a general immune response by the rats to this agent. Subsequently, the packed cell volume (PCV) of the treated rats was significantly ($p \leq 0.05$) reduced in all the treatment groups when compared to that of the control group. The decreased PCV of the treated rats could have resulted from a likely bone marrow aplasia reported to have been caused by high doses of some chemicals such as insecticides (Guyton and Hall, 2011). Also, it is worthy of note that the decreased PCV was time dependent suggesting that longer treatment than that of ours could result to the rats developing aplastic anemia.

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