International Journal of Healthcare Sciences ISSN 2348-5728 (Online)
Vol. 4, Issue 2, pp: (2148-2157), Month: October 2016 - March 2017, Available at: www.researchpublish.com

Comparative Effects of *Jatropha gossypifolia* Leaf and Alpha-Lipoic Acid on The Hematological Profile of Rats Exposed to Cypermethrin

Opeyemi Faokunla[1], Oluseyi Akinloye [2], Regina Ugbaja [2], Abideen Adeogun [3]

[1]Department of Biological Sciences, Wesley University Ondo, Ondo State, Nigeria. [2]Department of Biochemistry and [3]Chemistry, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria

Abstract: ***Jatropha gossypifolia*** **has been associated with diverse medicinal characteristics. This study was aimed at investigating the hemato-protective ability of the methanolic extract of** ***Jatropha gossypifolia*** **leaf (JG) and alpha-lipoic acid (ALA) on cypermethrin-induced hematotoxicity; and to investigate the possible hematotoxic effect of the plant extract on the rats. The animals were grouped into eight groups of seven rats each and were administered as follows: Group I (control), corn oil; Group II, cypermethrin (CYP) (20mg/kg); Group III, CYP + (JG50mg/kg); Group IV, CYP + JG100mg/kg; Group V, CYP + ALA50mg/kg; Group VI, JG50mg/kg; Group VII, JG100mg/kg; and Group VIII, ALA50mg/kg. Data from this study showed that cypermethrin significantly (P<0.05) increased the amount of the red blood cells (RBC), white blood cells (WBC), lymphocytes (LYMP) and significantly (P<0.05) decreased the concentrations of the mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular volume (MCV) when compared to control. It also caused a non-significant (P>0.05) increase in hematocrit (HCT), haemoglobin (HG), granulocytes (GRAN) and platelets when compared to the control. Co-administration with the different doses of the methanolic extract of** ***Jatropha gossypifolia*** **and ALA were able to reverse these alterations. Thus, we concluded that the methanolic extract of** ***Jatropha gossypifolia*** **at the doses considered are safe and will elicit hemato-protective effect on cypermethrin-induced hematotoxicity.**

Keywords: ***Jatropha gossypifolia*****, alpha-lipoic acid, cypermethrin, hematotoxicity, methanolic extract.**

I. INTRODUCTION

The Euphorbiaceae family, which is considered one of the largest families of the angiosperms, covers about 7,800 species distributed in approximately 300 genera and 5 subfamilies worldwide (55). Jatropha have been reported for their medicinal uses, chemical constituents and biological activities such as *Jatropha curcas*, *Jatropha elliptica*, *Jatropha gossypifolia* and *Jatropha mollissima*, among others (41). From these species, *Jatropha gossypifolia* is a vegetal species widely known as "bellyache bush" and is a multipurpose medicinal plant largely used in folk medicine for the treatment of various diseases (3,41). Different parts of *J. gossypifolia* have been reportedly used in different countries in many ways. The leaves are used for intermittent fevers, carbuncles, eczema, itches, and sores on the tongues of babies, swollen mammae, stomachache and venereal disease (8). The leaf decoction is used for bathing wounds (30). The stem sap stops bleeding and itching of cuts and scratches (31,22). The bark contains the alkaloid jatrophine and a lignin (jatroiden) is found in its stem (22). *Jatropha gossypifolia* is an interesting medicinal plant with many medicinal uses. Especially its anticancer and molluscicidal activities which have been investigated with promising results, but more research is needed to confirm the activities of the individual compounds. In addition, the antiplasmodial activity, blood pressure lowering activity and coagulant or anticoagulant properties of different parts of the plant needed to be studied more into detail (33).

Alpha lipoic acid (ALA) with a molecular formula, $C_8H_{14}O_2S_2$, is a potent naturally occurring antioxidant found in many tissues of the body and synthesised by the liver in humans (13). ALA has been shown to affect cellular processes, alter redox status of cells, and interact with thiols and other antioxidants (36-37). It functions as a cofactor within pyruvate dehydrogenase and α-keto-glutarate dehydrogenase (46). Its solubility in both water and fat compartments enhanced the wide distribution of the antioxidant in plants and animals in both cellular membranes and cytosol (54). In addition, ALA and its reduced dithiol form, dihydrolipoic acid (DHLA), are powerful antioxidants, performing important roles such as: quenching of reactive oxygen species (53); regeneration of exogenous and endogenous antioxidants such as vitamins C and E, and glutathione (9); chelation of metal ions; and reparation of oxidized proteins (12,49). In most cells containing mitochondria, ALA is reduced by an NADH-dependent reaction with lipoamide dehydrogenase to form DHLA. In cells that lack mitochondria ALA can be reduced to DHLA via NADPH with glutathione and thioredoxin reductases (27).

Synthetic pesticides have become an integral component of various pest eradication programmes for modern farming, various vector borne diseases and household pests. Pyrethroid pesticides represent a major class of very effective multipurpose chemicals, accounting for about 30% of global insecticidal market. (2,6,11,18). Pyrethroids are considered as comparatively safe pesticides, but their increased utility due to enhanced toxic potential and easy biodegradability, necessitate non-target toxicity assessment (1). The lipophilic nature of pyrethroids exacerbate their toxicity by facilitating rapid access to the various tissues which in turn increase their interaction with central nervous system, for which these pesticides have high affinity (4).

Cypermethrin is a type II pyrethroid pesticide, used widely in developing and undeveloped nations for almost every aspect of pest control, either alone or in combination (50). Due to its universal usage, it has maximum chance of accumulating in various food chains and thus imparting related toxicity (11,39,32,43,57). Cypermethrin (both the cis- and trans-isomers) is metabolized via the cleavage of the ester bond to phenoxybenzoic acid and cyclopropane carbolic acid.

Lots of work has been done on cypermethrin toxicity, cutting across the histopathological, toxicological and the biochemical alterations in different species of animals (23) but very few was centred on the amelioration of these toxicities induced by this pyrethroid. Thus, the aim of this work was to examine the efficacy of the methanolic extract of *Jatropha gossypifolia* leaf at different doses and alpha-lipoic acid (ALA) in combating cypermethrin-induced alterations in the haematological profile of male wistar rats.

II. MATERIALS AND METHODS

Laboratory animals: Fifty-six adult male wistar rats weighing between 160-200g were purchased from the animal house, University of Ibadan, Nigeria. The Animals were acclimatized in the animal house of the Federal University of Agriculture, Abeokuta (FUNAAB), for a period of two weeks under standard environmental conditions, with an approximately 12 hours light/dark cycle before the commencement of the experiment. The animals were fed with standard laboratory diet and water *ad libitum*. After 2 weeks of acclimatization, the animals were subjected to various treatments for a period of 4 weeks. All experiments were carried out in accordance with guidelines of the Experimental Animal Ethic's Committee. Cypermethrin (10% EC) (trade name: Cyperforce) was obtained from an agricultural store in Abeokuta, Nigeria while the alpha lipoic acid was gotten from Hanzhou Zhenghan Biological Technology Co., Ltd., China. Appropriate doses of each based on the weight of the rats were measured and dissolved in corn oil.

Plant Material: Fresh leaves of *Jatropha gossypifolia* were plucked from Bashiri area of Ado-Ekiti and authenticated at the Plant Science department of the Ekiti State University, Ado-Ekiti by the plant taxonomist, Mr. F. O. Omotayo, with an electronic herbarium number UHAE163. The leaves of the plant were gently and thoroughly washed with clean water to avoid any form of contamination, then air-dried under shade for about two weeks and mechanically pulverized into a fine texture which was subjected to cold extraction.

Extraction of *Jatropha gossypifolia*: 500g of the powdered form were soaked in 3.3L of methanol for 72hrs. The solvent was decanted and filtered using whatman no 1 filter paper. The extract was then concentrated with the aid of a rotary evaporator at 40^0C. The percent yield of the methanolic extract was 9.8% w/w. The concentrated methanolic extract obtained was stored at -4^{o}C until use.

Treatment: The animals were divided into eight (8) groups of seven (7) animals each based on their weight ranges. The first group is the control and were given corn oil, the second group was administered with cypermethrin (20mg/kg), the third, fourth and fifth groups were co-administered with cypermethrin (20mg/kg) and methanolic extract of *Jatropha gossypifolia* (50mg/kg), cypermethrin (20mg/kg) and methanolic extract of *Jatropha gossypifolia* (100mg/kg), and,

cypermethrin (20mg/kg) and alpha lipoic acid (50mg/kg) respectively; while the sixth, seventh and eighth groups took only the methanolic extract of *Jatropha gossypifolia* (50mg/kg), methanolic extract of *Jatropha gossypifolia* (100mg/kg), and alpha lipoic acid (50mg/kg) respectively. Administration was carried out once daily using orogastric canula for a period of 28 days. After the experiment, blood was collected through ocular puncture into an ethylene diamine tetrachloro acetic acid (EDTA) bottle for the estimation of hematological parameters.

Hematological studies: Blood collected in EDTA tubes was analyzed for white blood cells (WBC), red blood cells (RBC), Mean corpuscular haemoglobin (MCH), Mean corpuscular haemoglobin concentration (MCHC), Mean corpuscular volume (MCV), hemoglobin (HG), hematocrit (HCT), platelets (PLT), lymphocytes (LYMP) and granulocytes (GRAN) using automated cell counter.

Statistical analysis: Results are expressed as the Mean ± SD. Statistical significance was evaluated by one-way analysis of variance (ANOVA) using SPSS version 17.0. and the individual comparisons were obtained by the Duncan multiple range test (DMRT) (Duncan, 1957). A value of $p<0.05$ was considered to indicate a significant difference between groups.

III. RESULTS

The effects of the methanolic leaf extract of *Jatropha gossypifolia* and alpha-lipoic acid on the haematological parameters of rats exposed to sub-chronic administration of cypermethrin were shown in the figures below. From this present study, cypermethrin caused alterations in the haematological profile of rats.

In the present study, figure 1 below showed that cypermethrin exposure caused an increase in RBC (106.07%), HCT (12.04%) and HG (8.83%) concentrations compared to the control but the increase was only significant ($P<0.05$) in the RBC. Supplementation with the methanolic leaf extract of *Jatropha gossypifolia* at both 50mg/kg and 100mg/kg body weight, and alpha-lipoic acid (50mg/kg body weight) was able to lower the increase in RBC (38.30%, 28.79% and 42.02% respectively), HCT (9.11%, 11.24% and 13.10% respectively) and HG (12.55%, 12.86% and 13.94% respectively). The decrease was only significant ($P<0.05$) in haemoglobin. Compare to the control, the groups administered with only 50mg/kg and 100mg/kg body weight of the methanolic extract of *Jatropha gossypifolia* leaf (JG50mg and JG100mg respectively), and alpha-lipoic acid (ALA50mg) showed no significant ($P>0.05$) difference.

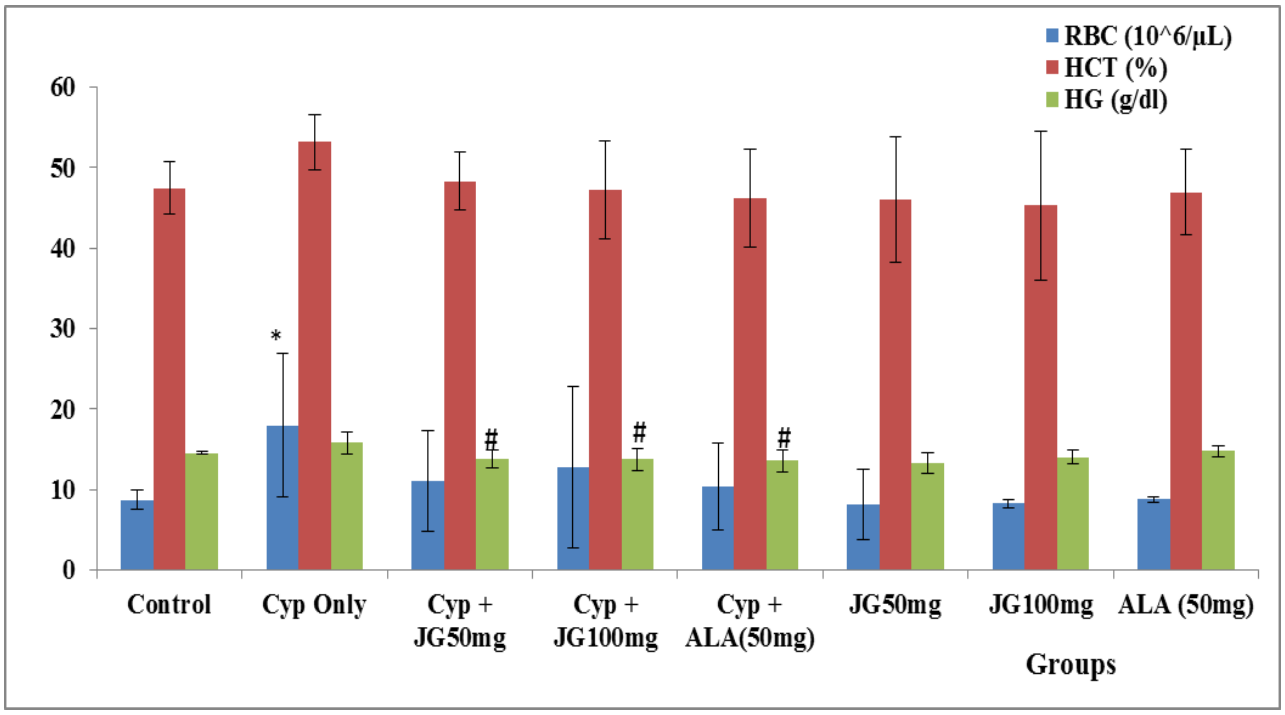

Fig 1: Effect of the different doses of the methanolic extract of *Jatropha gossypifolia* (JG50mg and JG100mg) and alpha lipoic acid (ALA) on the red blood cells (RBC), hematocrit (HCT) and hemoglobin (HG) in rats orally administered with cypermethrin. The bars represent mean±SD (n = 7); *: significantly different ($p<0.05$) compared with control group; # : significantly different ($p<0.05$) compared with CYP Only group.

Cypermethrin exposure caused a significant (P<0.05) increase of 76.82% and 56.01% in WBC and LYMP respectively; and a non-significant (P>0.05) increase of 93.48% in GRAN when compared to control (figure 2). Co-administration of cypermethrin with the methanolic leaf extract of *Jatropha gossypifolia* (both 50mg/kg and 100mg/kg) and alpha-lipoic acid were able to lower these increases. Co-administration with JG50mg significantly (P<0.05) reduced WBC by 32.44% and showed a non-significant (P>0.05) reduction of 33.06% and 37.98% in LYMP and GRAN respectively. Co-administration with JG100mg showed a significant (P<0.05) decrease of 37.55% and 47.06% in WBC and LYMP respectively and a non-significant (P>0.05) decrease of 10.79% in GRAN. Also, co-administration with ALA50mg resulted in a significant (P<0.05) decrease of 60.67% and 51.12% in WBC and LYMP respectively and a non-significant (P>0.05) decrease of 37.30% in GRAN as shown in figure 2. Moreover, the result further showed that the groups that took only the plant extracts (JG50mg and JG100mg) and the group that took only the alpha-lipoic acid showed no significant (P>0.05) difference when compared to the control: JG50mg caused an insignificant (P>0.05) reduction of 29.64% and 26.90% in WBC and LYMP respectively, and an insignificant (P>0.05) increase of 5.22% in GRAN; JG100mg caused an insignificant decrease of 23.84% in WBC and an insignificant (P>0.05) increase of 2.53% and 21.74% in LYMP and GRAN respectively; while ALA50mg caused an insignificant (P>0.05) decrease of 40.98% in WBC, an insignificant (P>0.05) increase of 9.57% in GRAN and has no change on the LYMP concentration

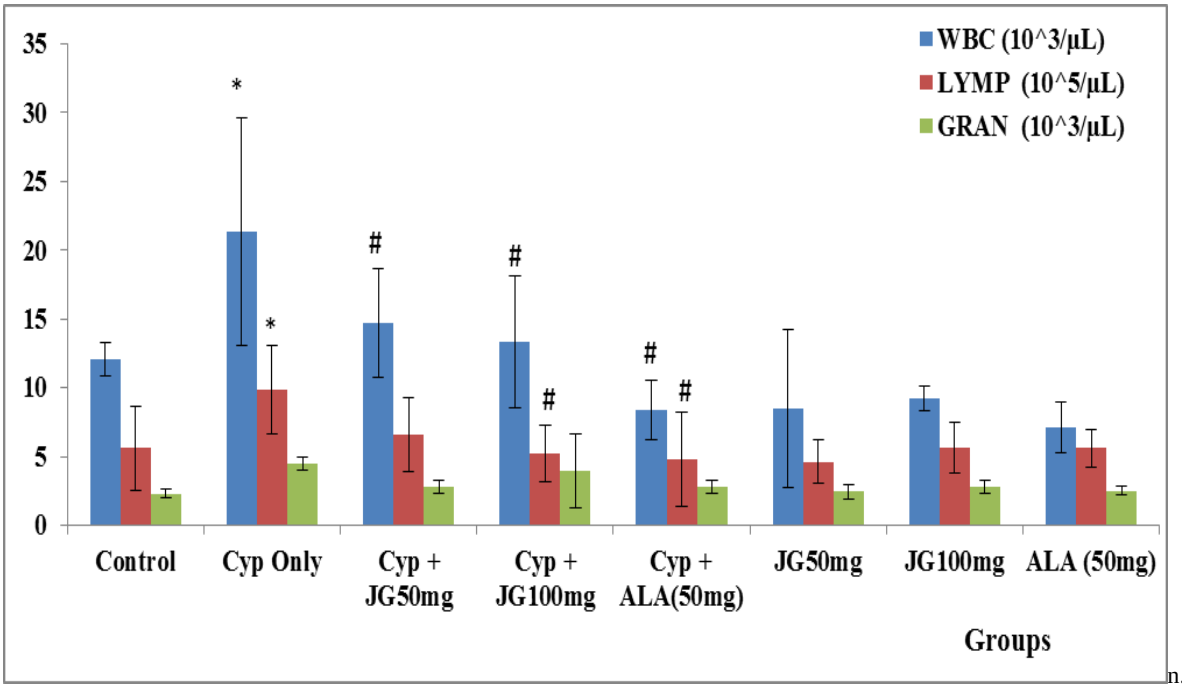

Fig 2: Effect of the different doses of the methanolic extract of *Jatropha gossypifolia* (JG50mg and JG100mg) and alpha lipoic acid (ALA) on the white blood cells (WBC), lymphocytes (LYMP) and granulocytes (GRAN) in rats orally administered with cypermethrin.

The bars represent mean±STD (n = 7); *: significantly different (p<0.05) compared with control group; # : significantly different (p<0.05) compared with CYP Only group.

All the red cell indices were significantly (P<0.05) decreased (MCH by 50.64%, MCHC by 14.34% and MCV by 51.72%) after cypermethrin exposure when compared with the control as shown in figure 3. Co-administration with the methanolic leaf extract of *Jatropha gossypifolia* at 50mg/kg and 100mg/kg doses and alpha-lipoic acid (50mg/kg) were able to raise the levels of all the red blood indices although at different proportions when compared to the group that was administered with cypermethrin only: MCH was significantly (P<0.05) increased by 69.49%, 90.40% and 74.92% respectively; MCV was also significantly (P<0.05) increased by 67.77%, 93.64% and 85.22% respectively; while MCHC was insignificantly increased at 50mg/kg and 100mg/kg doses of the plant extract by 12.76% and 12.43% respectively whereas alpha-lipoic acid caused a significant (P<0.05) increase of 14.26%. Compare to the control, the groups administered with only 50mg/kg and 100mg/kg body weight of the methanolic extract of *Jatropha gossypifolia* leaf, and alpha-lipoic acid (50mg/kg) insignificantly (P>0.05) reduced the levels of MCH by 0.17%, 3.35% and 5.08% respectively; MCHC by 6.15%, 3.94% and 2.28% respectively; and MCV by 9.13%, 10.80% and 2.96% respectively.

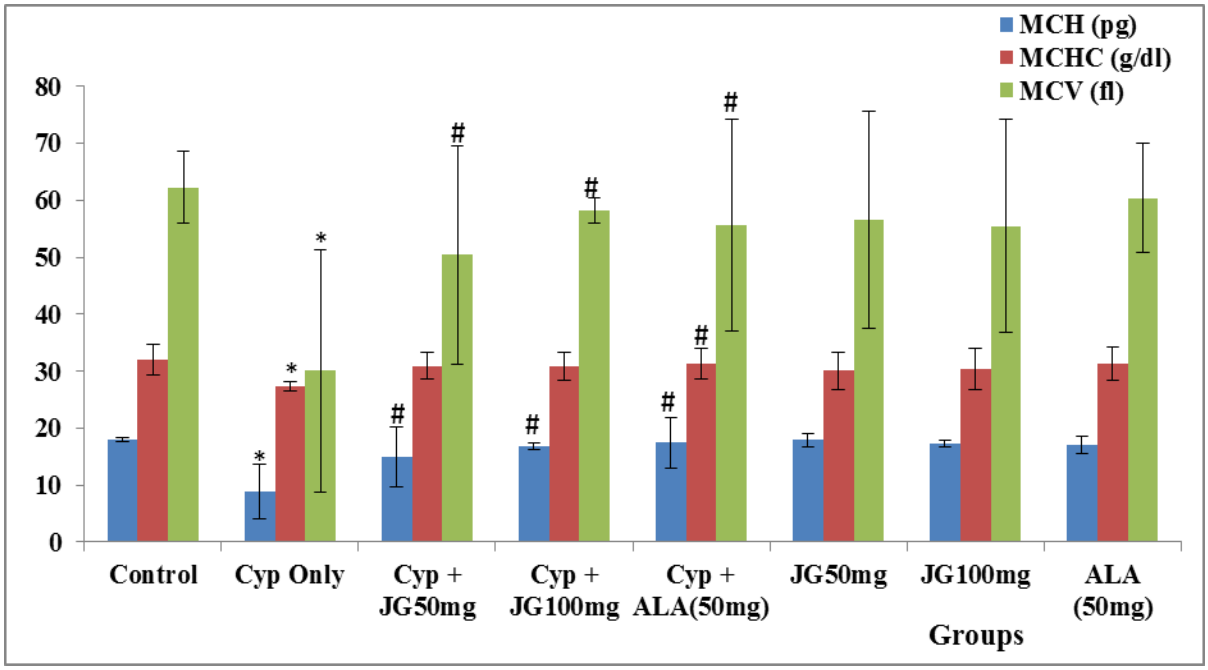

Fig 3: Effect of the different doses of the methanolic extract of *Jatropha gossypifolia* (JG50mg and JG100mg) and alpha lipoic acid (ALA) on the mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular volume (MCV) in rats orally administered with cypermethrin.

The bars represent mean±STD (n = 7); *: significantly different ($p<0.05$) compared with control group;
: significantly different ($p<0.05$) compared with CYP Only group.

Sub-acute administration of cypermethrin does not cause a significant ($P>0.05$) increase in platelets compared to control (figure 4), however, an increase of 33.06% was observed. Supplementation with the plant extract at JG50mg and JG100mg, and ALA50mg were able to lower this increase by 22.16%, 16.70% and 21.17% respectively. The groups administered with only the plant extract and ALA slightly raised the amount of the platelet counts by 5.82% (JG50mg), 22.57% (JG100mg) and 19,29% (ALA50mg) respectively. These increases were not significant ($P>0.05$) compared to control.

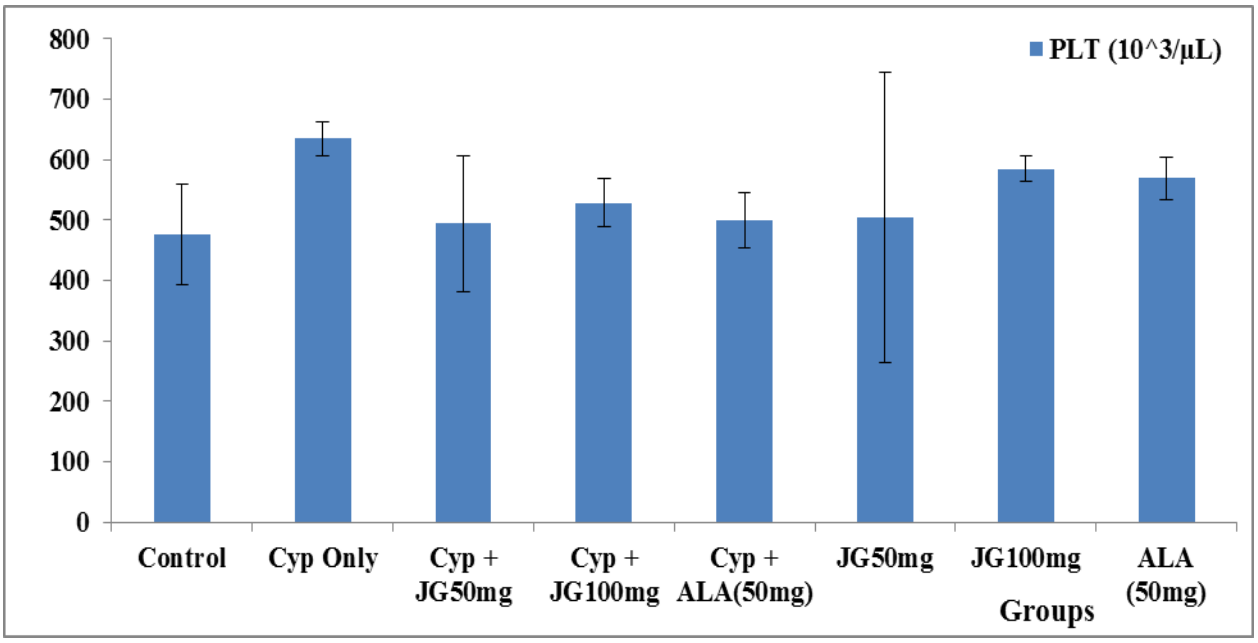

Figure 4: Effect of the different doses of the methanolic extract of *Jatropha gossypifolia* (JG50mg and JG100mg) and alpha lipoic acid (ALA) on platelets (PLT) in rats orally administered with cypermethrin.

The bars represent mean±SD (n = 7).

IV. DISCUSSION

Hematology is defined as the branch of biology, which deals with the morphology of blood and blood forming organs. Haematological profile assessment plays a critical role in appraising or evaluating the toxicity of any exogenous compound in the system since blood is an easily available fluid used as an important diagnostic tool to evaluate any xenobiotic toxicity including pyrethroid pesticides (19,51). It can thus provide important information about the internal environment of the organism (24). It is known that blood accounts for the largest percentage of the total body fluids, thus serving as the commonest route for the transportation of various substances such as oxygen, food and drugs. Bioaccumulation of compounds in an excessive or lower than normal level in the blood is a pointer to a clinic-pathological condition (14). The toxic effect of a xenobiotic is usually expressed as an increase or decrease in one or more serum or plasma markers, an enzyme, a hormone or any endogenous compound. In assessing haematotoxicity, increase or decrease in blood variables indicates excess or suppression of blood cells production respectively or an imbalance between blood cells production and destruction. Such analysis is relevant to risk evaluation as the changes in the haematological system have higher predictive value for human toxicity, when the data are translated from animal studies to humans (35).

Information regarding hematological alterations following exposure to cypermethrin is inconsistent. It might be partially due to various non-specific features influencing hematological parameters such as alterations in circulations, rate of food consumption, fluid and salt balance, food utilization and feeding pattern, venipuncture and blood sampling, and experimental variables (20).

In the present study, the effect of cypermethrin, a synthetic pyrethroid, was investigated on the haematological parameters of male albino rats; and the ameliorative effects of alpha-lipoic acid and the methanolic extract of *Jatropha gossypifolia* leaf were also determined. Exposure to cypermethrin caused polycythaemia/erythrocytosis as it significantly increased RBC; it also increased the concentrations of HCT and HG insignificantly when compared to the control. Abnormally high levels of RBC, hemoglobin and hematocrit are indicative of dehydration, a congenital heart disease, kidney tumor, pulmonary fibrosis or cigarette smoking. These changes might have been brought about by an increased synthesis of hemoglobin and RBC number in the hemopoietic organs. Tissue hypoxia leads to elevated levels of erythropoietin, which in turn leads to stimulation of erythropoeisis and increase in numbers of circulating normal RBCs. This result was similar to the work of Suleiman *et al.,* (52) who reported that there was a significant elevation of PCV, Hb concentration and RBC counts in mice administered chlorpyrifos (CPF) only. Luty et al. (28) also reported that deltamethrin and fenvalerate regardless of doses enthused erythropoiesis and synthesis of hemoglobin in male Swiss mice. However, in female mice, Luty *et al.,* (28) reported that deltamethrin led to anemia which indicated conquest of erythropoiesis and hemoglobin synthesis. It was thus hypothesized that female mice could be more sensitive to lower doses of pyrethroids for longer time, which could be related with depressant effect of synthetic pyrethroids on erythropoietin (EPO) hormone which controls erythropoiesis. Also at a dose of 40mg/kb body weight, Remya et al. (40), observed an increase in the packed cell volume, total erythrocyte count and haemoglobin while at higher doses of 80mg/kg and 120mg/kg, these parameters were decreased. Experiments conducted by Ishmael and Litchfield (25) where permethrin was fed to mice revealed non-significant effect on hematological values. Co-administration of the methanolic extract of *Jatropha gossypifolia* leaf at 50mg/kg and 100mg/kg body weight, as well as alpha-lipoic acid were able to alleviate the negative effect of cypermethrin (figure 1). Same result was reported by Suleiman et al. (52) when he was able to show that vitamins C and E pretreatment significantly suppressed the adverse hematological effect by CPF.

Increase in total leukocyte count has been suggested to be due to stimulated lymphopoiesis and/or enhanced release of lymphocytes from lymph myeloid tissue (15). Such lymphocyte response might be due to the presence of toxic substances which may be associated with the pollutant induced tissue damage and severe disturbance of the non-specific immune system leading to increased production of leukocytes. Several authors have noticed an increase in WBC in animals repeatedly treated with sublethal doses of insecticides. In this present study, cypermethrin significantly increased the white blood cells and lymphocytes when compared to control. Although not significant, this study also showed that cypermethrin increased the level of granulocytes compared to the control (figure 2). Similar results have been reported by many authours. Leukocytosis has been documented after cypermethrin or other pyrethroids treatment in mammals and rabbits (48). Increased WBC was also observed in rats treated with diodine (26), aldrin (5), novel phosphorothionate (38) and, lindane and endosulfan (7). El-Saeed and Hassan (17) also reported the relationship between chronic lymphocytic leukemia and pesticide exposure among Egyptian farm workers in which he reported high lymphocytes, WBC and platelet counts. Haratym-Maj (21) suggested that an increase in the number of WBC in the blood of animals might result from the

mobilization of the immunological system and/or a shift in the leukocytic pool from the spleen to peripheral blood. However, in combination with alpha-lipoic acid and the methanolic extracts of *Jatropha gossypifolia*, these increases were returned to normal.

MCH, MCHC and MCV were all significantly decreased after cypermethrin exposure as shown in this study (figure 3). This implies microcytic hypochromic anemia or hemoglobinopathy (44) usually as a result of iron deficiency. Haemoglobin in this condition are abnormally shaped thus making it unavailable to carry oxygen through the blood efficiently. This result is in accordance with the work of Dhembare and Pandhe, (16) where they observed a decrease in MCV, MCH and MCHC in rats treated with various insecticides such as endosulfan, malathion, methyl parathion, phosphomidon, monocrotophos and fenvalerate. However, significant increase in MCV and MCH after pyrethroid treatment was reported in various animals at different doses by Matsushima et al. (29), Sayim et al. (45), and Shah et al. (47). Sayim et al. (45) and Shah et al. (47) reported no effect of pyrethroid treatmenton MCHC. Supplementation with alpha-lipoic acid and the methanolic extracts of *Jatropha gossypifolia* leaf was able to significantly raise the levels of these red cell indices.

Platelets are blood cells involced in coagulation and this requires that the platelets be in sufficient size, number and function (56). Cypermethrin exposure in this study (figure 4) showed an insignificant increase in platelet count compared to the control. It has been shown that mild elevation of platelet count (thrombocytosis) can be a sign of chronic infection. This result is similar to the report of Saka *et al.*, (42) but at variance with that of Sayim et al. (45) that reported a decrease in platelet count in rats exposed to pyrethroids. Co-administration with alpha-lipoic acid and the methanolic extract of *Jatropha gossypifolia* leaf were able to lower this increase.

Alpha-lipoic acid and the methanolic extract of *Jatropha gossypifolia* leaf at both 50mg/kg and 100mg/kg body weight caused no significant changes on all the haematological parameters assayed for in this study when compared with the control. Oduola *et al.* (34) obtained similar result in his work when he observed that *Jatropha gossypifolia* stem latex had no significant difference on haematological parameters. This shows that both the alpha-lipoic acid and the plant extract exhibited no toxic effect at these doses. Also the ameliorative work done by alpha-lipoic acid and the plant extract when used in combination with cypermethrin could also be linked to the antioxidant ability of alpha-lipoic acid and that of the plant.

V. CONCLUSION

The results from the present investigation showed that sub-acute exposure to cypermethrin induced a level of toxicity on the haematological profile of rats. The use of alpha-lipoic acid and the methanolic extracts of *Jatropha gossypifolia* in combination with cypermethrin was able to effectively ameliorate the deleterious effects of cypermethrin on the haematological parameters. Furthermore, the two doses of methanolic extract of *Jatropha gossypifolia* leaf (50mg/kg and 100mg/kg), and the alpha-lipoic acid employed in this study showed no sign of toxicity on the rats' haematological profile. However, minimal exposure to cypermethrin during domestic, veterinary, agricultural or industrial use is recommended.

REFERENCES

[1] Addy-Orduna LM, Zaccagnini ME, Canavelli SB, Mineau P et al (2011) Formulated beta-cyfluthrin shows wide divergence in toxicity among bird species. J Toxicol.

[2] Adjrah Y, Karou SD, Agbonon A, Ameyapoh Y, de Souza C, Gbeassor M et al (2013) Effect of cypermethrin-treated lettuce (Lactuca sativa) on wistar rat liver. J Appl Pharm Sci 3: 128–132

[3] Agra MDF, Silva KN, Basílio IJLD, De Freitas PF, Barbosa-Filho JM (2008) Survey of medicinal plants used in the region Northeast of Brazil. Braz J Pharmacog 18(3): 472–508.

[4] Anadon A, Martinez-Larranaga MR, Diaz MJ, Bringas P, Fernadez MC, Martinez MA, Fernadez-cruz ML et al (1996) Pharmacokinetics of amoxicillin in broiler chickens. Avian Pathol 25: 449 – 458.

[5] Anil DM, John DJ, Sudhee K et al (1996) Haematological changes in albino rats under aldrin intoxication. Indian J Comp Anim Physiol 14(1):63-66.

[6] Assayed ME, Khalaf AA, Salem HA et al (2010) Protective effects of garlic extract and vitamin C against in vivocypermethrin-induced teratogenic effect in rat offspring. Food Chem Toxicol 48: 3153–3158.

[7] Azhar MD (2007). Pesticidal residue analysis of organochlorine residues in different milk samples from Chittoor district in Andhra Pradesh, India. Final report of UGC Minor Research Project during the period from 2004-2006.

[8] Balee RE, Ikuebe OM, Ihimire IG et al (1994) Afr J Med Med Sci 25(4): 373 –375.

[9] Bast A and Haenen GR (2003) Lipoic acid, a multifunctional antioxidant. Biofactors 17: 207-213

[10] Bhushan B, Saxena N, Saxena PN et al (2010) Beta-cyfluthrin induced histochemical alteration in the liver of the albino rat. Scand J Lab Anim Sci 37: 61–66.

[11] Bhushan B, Saxena PN, Saxena N et al (2013) Biochemical and histological changes in rat liver caused by cypermethrin and beta-cyfluthrin. Arch Ind Hyg and Toxicol 64: 57–67.

[12] Biewenga GP, Haenen GR, Bast A et al (1997) The pharmacology of the antioxidant lipoic acid. Gen Pharmacol 29: 315–331.

[13] Busby RW, Schelvis JPM, Yu DS, Babcock GT, Marletta MA (1999) Lipoic acid biosynthesis: LipA is an iron-sulphur protein. J Am Chem Soc 121: 4706–4707.

[14] Calabrese AL, Thurberg FP, Dawson MA, Wenzl DR et al (1975) Sublethal physiological stress induced by cadmium and mercury in winter flounder *Pseudopleuronectes americanus*. In: sublethal effect of toxic chemicals (eds.), Koeman JH and Strik JJ. Elsevier, Scientific Co. Amsterdam, Netherland. pp. 15-21.

[15] Das BK and Mukherjee SC (2003) Toxicity of cypermethrin in Labeo rohita fingerlings: biochemical, enzymatic and hematological consequences. Comp Biochem and Physiol C 134:109-121.

[16] Dhembare AJ and Pondhe GM (2000) Haematological changes in fish, Punctivs sophoreexposed to some insecticides. J Exp Zool India 3(1):41-44.

[17] El.Saeed WY and Hassan MH (2000) Chronic lymphocytic leukemia in Egyptian farm workers exposed to pesticides. East Mediterr H J 5: 960 – 966.

[18] El-Magd SAA, Sabik LME, Shoukry A et al (2011) Pyrethroid toxic effects on some hormonal profile and biochemical markers among workers in pyrethroid insecticide company. Life Sci J 8: 311–322.

[19] Fetoui H, Garoui EM, Zeghal N et al (2009) Lambda-cyhalothrin-included biochemical and histopathological changes in the liver of rats: Ameliorative effect of ascorbic acid. Exp Toxicol Pathol 61: 189–196.

[20] P. Greaves, "Histopathology of Pre-clinical Toxicity Studies: Interpretation and Relevance in Drug Safety Evaluation", 3rd ed, Elsevier: UK 2007, pp. 102

[21] Haratym-Maj A (2002) Hematological alternations after pyrethroids poisoning in mice. Annu Agric Environ Med 9: 199–206

[22] Horsten SFAJ, Van den Berg AJJ, Kettenes-van den Bosch JJ, Leeflang BR, Labadie RP (1996) Cyclogossine A: a novel cyclic heptapeptide isolated from the latex of *Jatropha gossypifolia*. Planta Med 62(1): 46-50.

[23] Hussain J, Khan AL, Rehman N, Hussain ST, Khan F, Shinwari ZK et al (2009) Proximate and nutrient analysis of selected medicinal plant species of Pakistan. Pak J Nutr 8(1): 620 – 624.

[24] Hymavathi V and Rao LM (2000) Effect of sublethal concentrations of lead on the haematology and biochemical constituents of Channa punctatus, Bull Pure and Appl Sci 19A (1): 1-5.

[25] Ishmael I and Litchfield MH (1988) Chronic toxicity and carcinogenic evaluation of permethrin in rats and mice. Fundam Appl Toxicol 11: 308–322

[26] Jabeen M, Rahaman MF, Narasaiah J, Mustafa M (1991) Heamatological and Biochemical alterations in rats subjected to Dermal application of diodine. Indian J Comp Anim Physiol 9 (1): 27 – 31.

[27] Jones W, Li X, Qu Z, Perriott L, Whiteshell RR, May JM et al (2002) Uptake, recycling, and antioxidant actions of-lipoic acid in endothelial cells. Free Radic Biol Med 33: 83 – 93

[28] Luty SA Haratym-Maj J, Latuszynska DB, Rodak MK et al (2001) Oral toxicity of deltamethrin and fenvalerate in swiss mice. Annu Agric Environ Med 8: 245–254.

[29] Matsushima YO, Uchida M, Saitoh Y, Kawasaki K, Isama M, Kaniwa T, Kanno J et al (2003) Twenty-eight day repeated dose oral toxicity test of synergist of a pyrethroid insecticide, 2,3,3,3,2',3',3',3'-Octachlorodipropyl ether (S-421) in rats. Kokuritsu Iyakuhin Shokuhin Eisei Kenkyusho Hokoku, 121: 40–47

[30] Morton JF (1968) A survey of medicinal plants of Curacao. Econ. Bot. 22: 87 – 102.

[31] Morton JF (1981). Atlas of medicinal plants of Middle America: Bahamas to Yucatan. J Ethnopharmacol 75(1): 25 – 31

[32] Muthuviveganandavel V, Muthuraman P, Muthu S, Srikumar KS et al (2011). Individual and combined biochemical and histological effect of cypermethrin and carbendazim in male albino rats. J Appl Pharm Sci 1: 121–129.

[33] Oduola T, Avwioro OG, Ayaniyi TB et al (2005) Suitability of the leaf extract of *Jatropha gossypifolia* as an anticoagulant for biochemical and haematological analyses. African J Biotechnol 7: 679 – 681.

[34] Oduola T, Popoola GB, Avwioro OG, Oduola TA, Ademosun AA, Lawal MO et al (2007) Use of *Jatropha gossypifolia* stem latex as a haemostatic agent: how safe is it? J Med Plants Res 1 (1): 014 – 017.

[35] Olson H, Betton G, Robinson D, Thomas K., Monro A et al (2000) Concordance of toxicity of pharmaceuticals in human and in animals. Regul Toxicol Pharmacol 32: 56 – 67.

[36] Packer LK and Kraemer G (2001) Molecular aspects of lipoic acid in the prevention of diabetes complications. Nutr 17: 888 – 895

[37] Packer L, Witt EH, Tritschler HJ et al (1995) Alpha-lipoic acid as a biological antioxidant. Free Radic Biol Med 19: 227–250.

[38] Rahman MF and Siddiqui MKJ (2006) Hematological and clinical chemistry changes induced by subchronic dosing of a novel phosphorothionate (RPR-V) in wistar male and female rats. Drug Chem Toxicol 29: 95 – 110.

[39] Rana N, Saxena N, Sharma HN, Saxena PN et al (2008). Comparative genotoxicity of alpha-cyano pyrethroids on Drosophila melanogaster. Entamon 33: 135–138.

[40] Remya R, Mammen J, Abraham ND, Nair CR, Lalithakunjamma CM (2010) Hemtological and biochemical profile in sub lethal toxicity of cypermethrin in rats. Int J Biol Med Res 1(4): 211-214.

[41] Sabandar CW, Ahmat N, Jaafar FM, Sahidin I et al (2013) Medicinal property, phytochemistry and pharmacology of several Jatropha species (Euphorbiaceae). Phytochem 85: 7–29.

[42] Saka WA, Akhigbe RE, Azeez OM, Babatunde TR (2011) Effect of pyrethroids insecticide exposure on haematological and haemostatic profiles in rats. Pak J Biol Sci 14(22): 1024-1027

[43] Sangha GK, Kaur K, Khera KS et al (2013). Cypermethrin induced pathological and biochemical changes in reproductive organs of female rats. J Enviro Biol 34: 99–105.

[44] Savithri, Y.; Ravi, P.; Jacob doss, P. (2010). Changes in hematological profiles of albino rats under chlorpyrifos Toxicity. Int J Pharmacol Biol Sci. 1(3): 1 – 7

[45] Sayim F, Yavasolglu NUK, Uyanikgil Y, Aktug H, Yavasolglu A, Turgut M et al (2005) Neurotoxic effects of cypermethrinin Wistar rats: a hematological, biochemical and histopathological study. J Health Sci 51: 300–307

[46] Schmidt AM, Hori O, Brett J et al (1994) Cellular receptor for ad-vanced glycation end products: implications for induction of oxidative stress and cellular dysfunction in the pathogenesis of vascular lesions. Atheroscler Thromb 14: 1521–1528.

[47] Shah MKA, Khan F, Rizvi M, Siddique S et al (2007). Effect of cypermethrin on clinico-haematological parameters in rabbits. Pak Vet J 27: 171–175

[48] Shakoori AR, Aslam F, Sabir M (1992). Effectof prolonged administration of insecticide (cyhalothrin/karate) on the blood and liver of rabbits. Folia Biol 40: 91 – 99

[49] Shay KP, Moreau RF, Smith EJ, Smith AR, Hagen TM et al (2009) Alpha-lipoic acid as a dietary supplement: Molecular mechanisms and therapeutic potential. Biochim Biophys Acta 1790: 1149–1160.

[50] Singh VK and Saxena PN (2001) Effect of Cybil (cypermethrin 25EC) and Cybil-sevin (carbaryl 50EC) combination on liver and serum phosphates in wistar albino rats. J Ecophysiol Occup Health 1: 229–234.

[51] Stonard MD and Evans GO (1995) Clinical Chemistry. In: General and Applied Toxicology. Ballantyne B, Marrs T, Turner P (eds). Macmillan Press, London. UK. pp. 247.

[52] Suleiman F, Ambali D, Olufemi A, Mufta'u S, AbdulGaniyu G, Olushola OO, Joseph OA (2010). Chlorpyrifos-Induced Clinical, Hematological and Biochemical Changes in Swiss Albino Mice- Mitigating effect by co-administration of vitamins C and E. Life Sci J 7(3): 37 – 44.

[53] Suzuki YJ, Tsuchiya M, Packer L et al (1991) Thioctic acid and dihdrolipoic acid are novel antioxidants which interact with reactive oxygen species. Free Radic Res Comm 15: 255 – 263

[54] Wada H, Shintani D, Ohlrogge J (1997) Why do mitochondria synthesize fatty acids? Evidence for involvement in lipoic acid production. Proc of Natl Acad Sci. U.S.A. 94: 1591–1596.

[55] Webster GL (1994) Classification of the euphorbiaceae. Ann Missouri Bot Gard 81: 3–143.

[56] Yakubu MT and Afolayan AJ (2009). Effect of aqueous extract of Bulbine natalensis (Baker) stem on the sexual behaviour of male rats. Int J Androl 32: 629-636.

[57] Yavasoglu A, Sayim F, Uyanikgil Y, Turgut M, Yavasoglu NUK (2006). The pyrethroid cypermethrin induced biochemical and histological alterations in rat liver. J Health Sci 52: 774–780.