WWW.IJRPR.COM

International Journal of Research Publication and Reviews

Journal homepage: www.ijrpr.com ISSN 2582-7421

Effects of Paracetamol Toxicity on Hematological Parameters of Wistar Rats

Precious Obinna Ekeke [a*], Aondowase Paul Iorhemba [a], Chinedu Kingsley Achukee [b], Ibe Maduabuchi Valentine [c], Victoria Chioma Ayozie-Samuel [d], Emmanuella Chinenye Nwokoro [e], Abdullahi Mohammed Sani [f]

[a] Department of Biochemistry, University of Calabar, Calabar, Nigeria.
[b] Qingdao Institute of Bioenergy and Bioprocess Technology, Chinese Academy of Sciences, China.
[c] Green Researchers in Analytical Chemistry, Environment and Climate Change (GRACE & CC), Department of Chemistry, Imo State University, Owerri, Nigeria.
[d] Department of Soil Science and Environment, Faculty of Agriculture and Veterinary Medicine, Imo State University, Owerri, Nigeria.
[e] Department of Metallurgical and Materials Engineering, University of Nigeria, Nsukka, Enugu State, Nigeria.
[f] Integrated Science Department, Niger State College of Education, Minna, Niger State, Nigeria.
E-Mail: preciousekeke1@gmail.com, ORCID: https://orcid.org/0009-0004-8312-1904

ABSTRACT

Paracetamol (acetaminophen) is a widely used analgesic and antipyretic drug. However, at high doses, it induces toxicity, which may affect various physiological systems, including hematological parameters. This study investigates the effects of paracetamol toxicity on the hematological profile of Wistar rats. Fifteen female albino Wistar rats (180–200 g) were divided into three groups (n=5). Group 1 (control) received only standard rat feed, while Groups 2 and 3 were administered paracetamol at 2 g/kg body weight for 4 and 7 days, respectively. Hematological parameters, including white blood cell (WBC) count, red blood cell (RBC) count, hemoglobin (HGB) concentration, packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular hemoglobin concentration (MCHC), platelet count (PLT), lymphocytes (LYM), mid-sized cells (MID), and granulocytes (GRA), were analyzed. The results revealed significant alterations in hematological indices in the paracetamol-treated groups compared to the control group. WBC counts increased significantly, indicating leukocytosis, which is consistent with an inflammatory response. RBC counts, HGB concentration, and PCV decreased significantly, suggesting the development of anemia, likely due to oxidative stress-induced hemolysis or suppression of erythropoiesis. Platelet counts also increased, indicating thrombocytosis, which may be a compensatory response to inflammation or tissue damage. Differential leukocyte counts showed an increase in granulocytes and monocytes, further supporting the presence of systemic inflammation. These findings suggest that paracetamol toxicity induces hematological alterations that may contribute to systemic inflammation and compromised oxygen transport. Further studies are needed to explore the underlying mechanisms and potential interventions.

Keywords: Paracetamol, Hematology, Toxicity, Wistar rats, Parameters.

1.0 INTRODUCTION

Paracetamol or acetaminophen chemically named N-acetyl-p-aminophenol is a widely used over-the-counter analgesic (pain reliever) and antipyretic (fever reducer) globally. It is classified as a mild analgesic that is commonly used for the relief of headaches and other minor aches and pain and is a major ingredient in numerous cold and flu remedies (Freo *et al.*, 2021). In combination with the opiod analgesics, paracetamol can be used in the management of more severe pain such as post-surgical pain and providing palliative care in advanced cancer patients (NHS, 2008). The onset of analgesia is approximately 11 minutes after oral administration of paracetamol (Moller *et al.*, 2005) and its half-life is 1-4 hours. Though paracetamol is used to treat inflammatory pain, it is not generally classified as an NSAID (Non-Steroidal AntiInflamatory Drugs) because it exhibits only weak anti-inflammatory activity.

While it is generally safe for use at recommended doses (1000mg per single dose and up to 3000mg per day for adults human), acute overdoses of paracetamol can cause potentially fatal kidney, brain and liver damage, and in rare individuals, a normal dose can do the same; the risk is heightened by alcohol consumption. In the western world, paracetamol toxicity accounts for most causes of acute liver failure and for most drug overdoses (Daly *et al.*, 2008; Khashab *et al.*, 2007). Paracetamol toxicity occurs when its primary metabolic pathways become saturated, leading to the excessive formation of the reactive metabolite N-acetyl-p-benzoquinone imine (NAPQI). Under normal conditions, NAPQI is detoxified by conjugation with glutathione. However, in cases of overdose, glutathione stores become depleted, allowing NAPQI to bind to cellular macromolecules, resulting in oxidative stress and

tissue damage (James *et al*., 2003). While the hepatotoxic effects of paracetamol are well-documented, its impact on hematological parameters remains an area of ongoing investigation.

Acetaminophen is safe and well tolerated when taken in the usual therapeutic dose. However, overdose of paracetamol is fairly common and often associated with hepatic and renal damage in both humans and experimental animals. In large dosages, acetaminophen produces acute liver and kidney necrosis in most mammalian species. Paracetamol-induced liver necrosis has been studied extensively. Acute renal failure is not uncommon and occurs in approximately 1-2% of patients with acetaminophen overdose. Most instances of paracetamol toxicity resulted from large, single overdose (Seham *et al.,* 2004). Paracetamol toxicity occurs in 3 phases; Phase I - begins within hours of overdose Phase II- begins within 24hours-72 hours of overdose Phase III- begins within 3- 5 days of overdose (McMurtry *et al.,* 1978).

Hematological parameters, including red blood cell (RBC) count, white blood cell (WBC) count, hemoglobin concentration, platelet count, etc, serve as essential indicators of physiological and pathological changes in the body. They provide valuable insights into the effects of toxic substances on erythropoiesis, leukopoiesis, and thrombopoiesis (Dacie and Lewis, 2011). Alterations in these parameters can provide insights into the systemic effects of toxic substances, including drug-induced toxicity (Adebayo *et al.*, 2015). Previous studies have reported alterations in red blood cell (RBC) count, hemoglobin concentration, white blood cell (WBC) count, and platelet levels following paracetamol overdose, suggesting possible bone marrow suppression or inflammatory responses (Olaleye *et al.*, 2014).

Wistar rats are commonly used as experimental models for toxicological studies due to their genetic stability and physiological similarities to humans (Festing and Fisher, 2000). Investigating the effects of paracetamol toxicity on the hematological parameters of Wistar rats can provide insights into the systemic impact of the drug and contribute to understanding its toxicological profile.

Previous studies have demonstrated that paracetamol toxicity can induce oxidative stress and inflammation, which may indirectly affect hematopoiesis and blood cell integrity (Yuan *et al.*, 2019). However, the specific effects of paracetamol overdose on hematological parameters in Wistar rats remain underexplored, necessitating further investigation.

This study aims to evaluate the effects of paracetamol toxicity on hematological parameters in Wistar rats, providing valuable insights into the systemic impact of paracetamol overdose. By examining changes in RBC, WBC, hemoglobin, and platelet counts, as well as other hematological indices. Understanding these changes can provide critical information for clinical management and therapeutic interventions in cases of paracetamol overdose. This research seeks to contribute to the growing body of knowledge on paracetamol-induced toxicity and its broader implications for human health.

2.0 EXPERIMENTAL DESIGN

2.1 Materials

All chemicals used in this study were of analytical grade. The following chemicals were used; Paracetamol syrup (Emzor Pharmaceuticals Limited, Lagos, Nigeria), Chloroform (Sigma Chemicals Co., USA), Distilled water, Ethanol (70%).

2.2 Equipment

All the equipments used in this study were of laboratory standard. The following equipment and instruments were used in the study: Animal cages, digital weighing scale, Syringes (2 mL and 5 mL), EDTA-coated blood sample containers, Oral Gavage Cannula, Surgical Gloves, Dissecting Set, dissecting board, cotton wool, Automated Hematology Analyzer, Microhematocrit centrifuge.

2.3 Methodology

2.3.1 Animal Management

Fifteen (15) healthy female albino Wistar rats, weighing between 180g and 200g, were obtained from the College of Basic Medical Science Animal House, University of Calabar, Calabar. The rats were acclimatized for 14 days under standard laboratory conditions. The animals were housed in clean plastic cages, lined with sawdust that was changed every three days to maintain hygiene. During the acclimatization period, the rats were fed with grower's mash pellets and provided with drinking water *ad libitum*. All procedures adhered to the guidelines of the National Institute of Health for the care and use of laboratory animals (NIH Publication No. 18-23, 1985).

2.3.2 Experimental Groups

The rats were randomly divided into three experimental groups of five rats each (n=5 per group):

- **Group 1 (Normal Control):** This group received only standard rat feed and water for the duration of the experiment.
- **Group 2 (Paracetamol 2g/kg for 4 days):** This group was administered paracetamol at a dose of 2g/kg body weight orally for four consecutive days.

- **Group 3 (Paracetamol 2g/kg for 7 days):** This group received paracetamol at the same dose (2g/kg body weight) orally for seven consecutive days.

2.3.3 Sample Collection and Hematological Analysis

At the end of the experimental period, all animals were anesthetized using diethyl ether, and blood samples were collected via cardiac puncture into ethylenediaminetetraacetic acid (EDTA)-coated tubes for hematological analysis. Hematological parameters, including white blood cell (WBC) count, red blood cell (RBC) count, hemoglobin (HGB) concentration, packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular hemoglobin concentration (MCHC), platelet count (PLT), and differential leukocyte counts (lymphocytes, monocytes, and granulocytes), were analyzed using an automated hematology analyzer, at Hilltop health care center Calabar and values were recorded.

2.4 Statistical Analysis

Data were expressed as mean ± standard error of the mean (SEM) and analyzed using one-way analysis of variance (ANOVA). A significance level of $p < 0.05$ was considered statistically significant.

Parameters	**Units**	**Normal Control**	**Paracetamol 2g/kg B. W. for 4days**	**Paracetamol 2g/kg B. W. for 7days**
WBC	10^9 cells/L	6.25±0.63	9.96±2.99*	13.40±1.61*,a
RBC	10^{12} cells/L	8.67±0.67	7.21±0.27*	5.51±0.38*,a
HGB	g/dL	15.48±0.65	14.20±0.36	12.90±0.50*
PCV	%	55.57±1.48	52.87±1.19	49.61±0.40*
MCV	Fl	63.60±1.29	64.33±0.88	63.00±3.06
MCHC	g/dL	26.96±0.37	26.87±0.24	26.70±0.97
PLT	10^9 cells/L	393.60±64.79	397.00±48.03	474.33±81.02
LYM	%	67.66±3.63	72.00±2.63	75.70±2.10
MID	%	10.90±2.28	15.60±2.57	17.70±4.69*
GRA	%	10.46±1.22	12.43±1.02	21.57±4.52*,a

3.0 RESULT

Table 1: Results for the Hematological indices of the experimental groups

Values are expressed as mean ± SEM, n = 5.

*significantly different from normal control at $p<0.05$;

a = significantly different from paracetamol control at $p<0.05$.

Table 1 showing comparison of concentrations of different hematological parameters in the different experimental groups.

3.1 Effect of Paracetamol on White Blood Cells (WBC)

The WBC values increased from 6.25± 0.63 for the control group to 9.96± 2.99 for group 2 and 13.40 ± 1.61 for group 3. There was a significant difference between the control group and paracetamol treated groups (See Table).

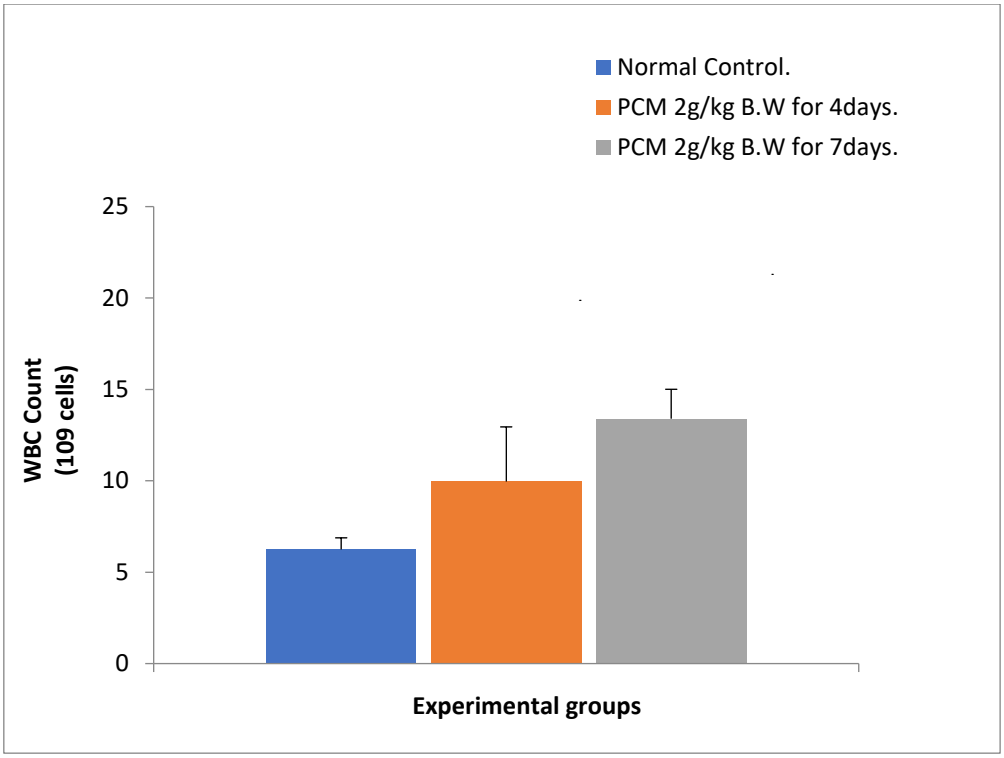

Figure 1: Comparison of White Blood Cell count in the different experimental groups.

Values are expressed as mean + SEM, n = 5.

*significantly different from Normal Control at p<0.05;

a = significantly different from Paracetamol Control at p<0.05.

3.2 Effect of Paracetamol on Red Blood Cells (RBC)

The RBC values decreased from 8.67± 0.27 for the control group to 7.21± 0.27 for group 2 and 5.51± 0.38 for group 3. There was a significant difference between the control group and the paracetamol treated groups (See Table).

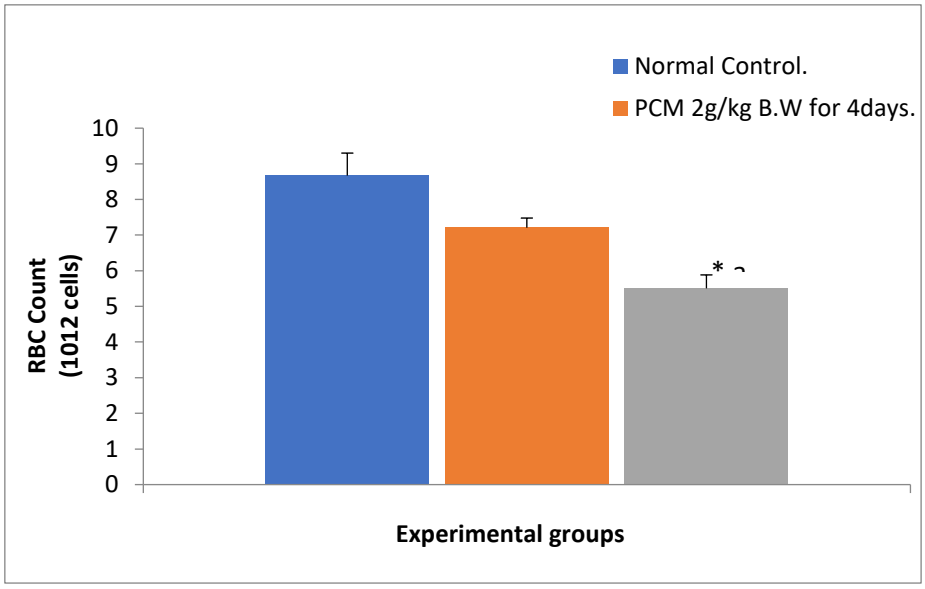

Figure 2: Comparison of Red Blood Cell count in the different experimental groups.

Values are expressed as mean + SEM, n = 5.

*significantly different from Normal Control at p<0.05;

a = significantly different from Paracetamol Control at p<0.05.

3.3 Effect of Paracetamol on Hemoglobin (HGB)

The hemoglobin concentration decreased from 15.48± 0.65 for the control group to 14.20± 0.36 and 12.90± 0.50 for group 2 and group 3 respectively. (See Table)

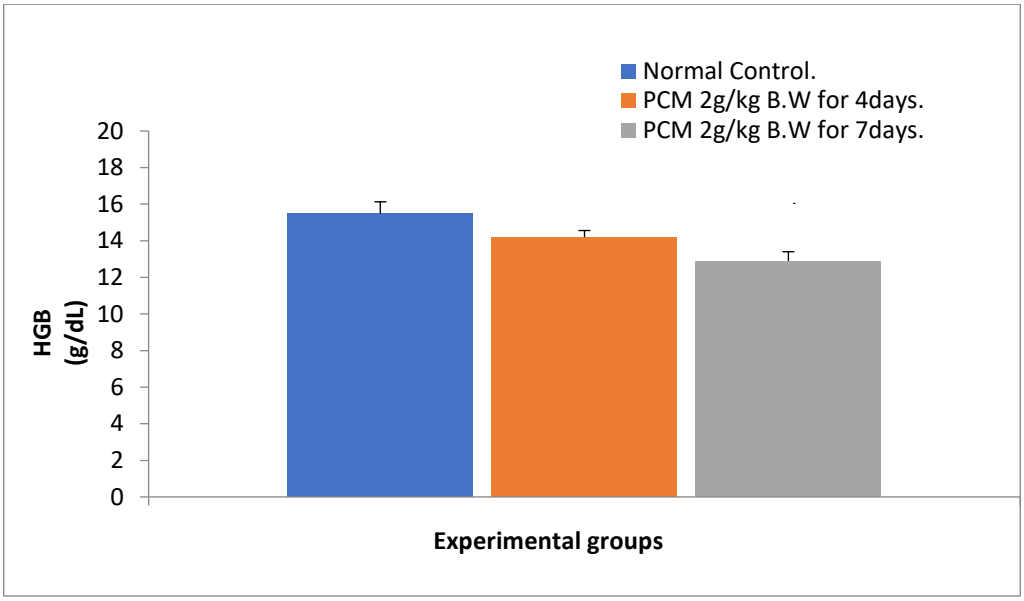

Figure 3: Comparison of hemoglobin concentration in the different experimental groups.

Values are expressed as mean + SEM, n = 5.

*significantly different from Normal Control at p<0.05

3.4 Effect of Paracetamol on Packed Cell Volume (PCV)

The PCV values decreased from 55.57± 1.48 for the control group to 52.87± 1.19 and 49.61± 0.40 for group 2 and group 3 respectively. (See Table)

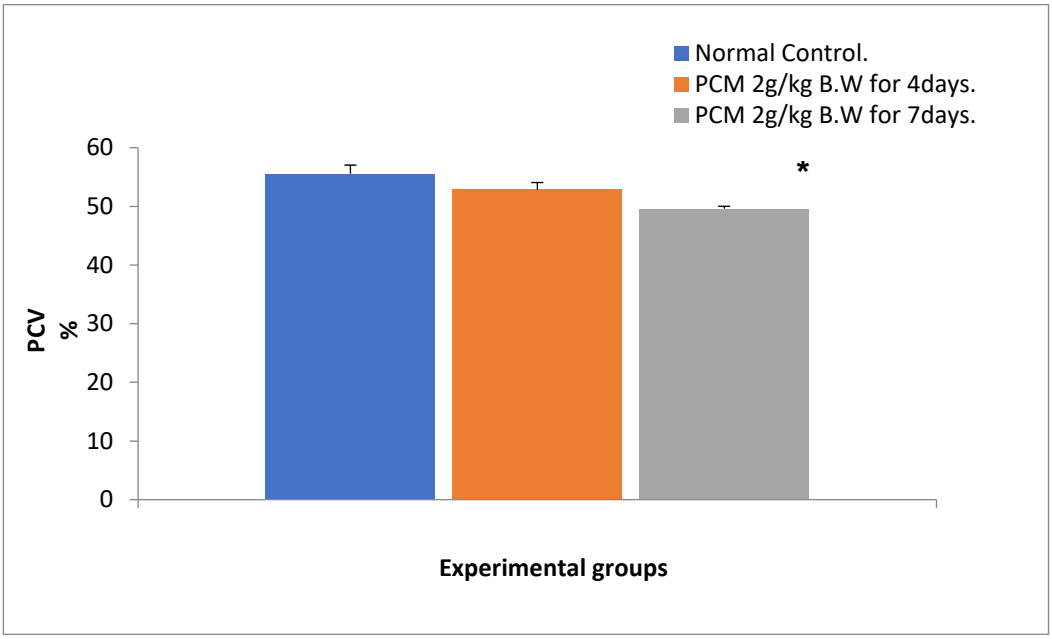

Figure 4: Comparison of packed cell volume percentages in the different experimental groups.

Values are expressed as mean + SEM, n = 5.

*significantly different from Normal Control at p<0.05

3.5 Effect of Paracetamol on Platelet Count (PLT)

The platelets count increased from 393.60± 64.79 for the control group to 597.00± 48.03 and 474.33± 81.02 for group 2 and group 3 respectively. (See Table)

4.0 Discussion and Conclusion

4.1 Discussion

The results of this study demonstrate that paracetamol toxicity significantly alters hematological parameters in Wistar rats, indicating systemic effects beyond its well-documented hepatotoxicity. The observed changes in hematological indices suggest that paracetamol overdose induces inflammation, oxidative stress, and potential bone marrow suppression, which are reflected in the blood parameters.

The results showed a significant increase in WBC count in the paracetamol-treated groups compared to the control. The WBC count increased from 6.25 ± 0.63 in the control group to 9.96 ± 2.99 in the 4-day paracetamol group and 13.40 ± 1.61 in the 7-day group. This elevation suggests an immune response to oxidative stress or tissue damage induced by paracetamol toxicity. Paracetamol toxicity is known to induce oxidative stress and inflammation, which can stimulate the release of pro-inflammatory cytokines and lead to an increase in WBC count (Yuan *et al*., 2019). The higher WBC count in the 7-day treatment group compared to the 4-day group suggests a dose-dependent effect.

A significant reduction in RBC count was observed following paracetamol administration. The RBC count decreased from 8.67 ± 0.67 in the control group to 7.21 ± 0.27 in the 4-day treatment group and 5.51 ± 0.38 in the 7-day group. This suggests that prolonged paracetamol exposure may lead to erythropoietic suppression or increased hemolysis, possibly due to oxidative damage to red blood cells. The depletion of glutathione, a key antioxidant, in red blood cells may lead to increased susceptibility to oxidative damage, resulting in hemolysis (Adebayo *et al*., 2015). Hemoglobin concentration and PCV levels also showed a decreasing trend. HGB levels decreased from 15.48 ± 0.65 in the control group to 14.20 ± 0.36 (4-day group) and 12.90 ± 0.50 (7-day group). Similarly, PCV levels dropped from 55.57 ± 1.48 in the control to 52.87 ± 1.19 and 49.61 ± 0.40 in the 4-day and 7-day groups, respectively. These reductions indicate anemia, which could be due to impaired erythropoiesis, increased RBC destruction, or hemorrhagic effects of paracetamol toxicity.

Platelet count was slightly elevated in paracetamol-treated groups, increasing from 393.60 ± 64.79 in controls to 397.00 ± 48.03 (4-day group) and 474.33 ± 81.02 (7-day group). Paracetamol-induced oxidative stress and inflammation can stimulate thrombopoiesis, leading to an increase in platelet production (Jaeschke *et al*., 2012). Lymphocyte (LYM) and granulocyte (GRA) percentages increased in the treated groups. LYM increased from 67.66% (control) to 75.70% (7-day group), while GRA levels increased significantly from 10.46% (control) to 21.57% (7-day group). The elevation in GRA suggests enhanced neutrophil recruitment, likely as a response to tissue injury and oxidative stress. The increase in monocytes (MID) also suggests an ongoing inflammatory process, as monocytes are involved in the immune response to tissue injury and infection.

4.2 Conclusion

The findings of this study demonstrate that paracetamol toxicity significantly affects hematological parameters in Wistar rats. The observed leukocytosis and increased platelet counts suggest an inflammatory response, while the reduction in RBC, hemoglobin, and PCV indicates possible anemia. he findings highlight the systemic effects of paracetamol overdose and underscore the importance of monitoring hematological parameters in cases of paracetamol toxicity. Further studies are needed to explore the mechanisms underlying these hematological alterations and their implications for human health.

Acknowledgements

- The University of Calabar Medical College and Biochemistry Departments, for their Technical assistance.
- Professor Uboh Friday Effiong for his professional directive and advice.
- My team for their dedication.

Compliance with Ethical Standards

Conflict of Interest: The authors declare no conflict of interest.

A. Statement of human rights

Ethical approval: For this type of study formal consent is not required.

B. Statement on the welfare of animals

Ethical approval: This study was conducted in strict compliance with ethical guidelines for the care and use of laboratory animals. The experimental protocol was reviewed and approved by the Institutional Animal Ethics Committee (IAEC) of the University of Calabar, ensuring adherence to internationally accepted ethical standards.

REFERENCES

Adebayo, O. L., Sandhir, R. and Adenuga, G. A. (2015). Hepatoprotective and hematoprotective effects of selenium in paracetamol-induced toxicity in rats. *Journal of Biochemical and Molecular Toxicology, 29*(9), 398-405.

Dacie, J. V. and Lewis, S. M. (2011). Practical Haematology. *Churchill Livingstone*.

Daly, F. F., Fountain, J. S., Murray, L., Graudins, A. and Buckley, N. A. (2008). Guidelines for the management of paracetamol poisoning in Australia and New Zealand—explanation and elaboration. A consensus statement from clinical toxicologists consulting to the Australasian poisons information centres. *Medical Journal of Australia*. Vol.188 (5):296–301.

Festing, M. F. W. and Fisher, E. M. C. (2000). *Animal models of human disease: Laboratory animals and genetics*. ILAR Journal, 41(2), 62–65.

Freo, U., Ruocco, C., Valerio, A., Scagnol, I. and Nisoli, E. (2021). Paracetamol: a review of guideline recommendations. *Journal of clinical medicine, 10*(15), 3420.

Jaeschke, H., McGill, M. R. and Ramachandran, A. (2012). Oxidant stress, mitochondria, and cell death mechanisms in drug-induced liver injury: Lessons learned from acetaminophen hepatotoxicity. *Drug Metabolism Reviews, 44*(1), 88-106.

James, L. P., Mayeux, P. R. and Hinson, J. A. (2003). Acetaminophen-induced hepatotoxicity. Drug Metabolism and Disposition, 31(12), 1499-1506.

Khashab, M., Tector, A. J. and Kwo, P. Y. (2007). Epidemiology of acute liver failure. *Curr. Gastroenterol. Rep*. Vol 9 (1): 66–73.

McMurtry, R. J., Snodgras, W. R. and Mitchell, J. R. (1978). Renal necrosis, glutathione depletion, and covalent binding after acetaminophen. *Toxicol. Appl. Pharmacol*. Vol 46: 87-100.

Moller, P., Sindet-Pedersen, S., Petersen, C., Juhl, G., Dillenschneider, A. and Skoglund, L. (2005). Onset of acetaminophen analgesia: comparison of oral and intravenous routes after third molar surgery. *British journal of anaesthesia*. Vol 94 (5): 642–648.

National Health Service (NHS). (2008). Control of pain in adults with cancer. Scottish Intercollegiate Guidelines Network (SIGN). Scotland: National Health Service (NHS)."6.1 and 7.1.1". Guideline, 2008; 106.

Olaleye, M. T., Rocha, J. B. T., Kade, I. J. and Adeogun, A. I. (2014). *Paracetamol-induced toxicity and its hematological effects in animal models: A review*. African Journal of Pharmacy and Pharmacology, 8(46), 1190-1195.

Seham, A., Abd, E., Begonia, M., Morales, A., Sara, K., Daniel, L. S. and Lucy, F. D. (2004). Localisation of cyclooxygenase-3 in rat central nervous system, *University of Cambridge Journal Physiology*. 555P, C156.

Yuan, L., Kaplowitz, N. and Li, J. (2019). Glutathione in liver diseases and hepatotoxicity. *Molecular Aspects of Medicine, 30*(1-2), 29-41.