

HAEMATOLOGICAL EFFECT OF TOLUENE IN WISTAR RATS

Abstract

Toxicity of toluene arising from solvent abuse, occupation hazards and environmental pollution has generated a lot of concern in recent times. Young people are getting more involved in the abuse of toluene by deliberate inhalation of toluene-containing substances which may result in high level of exposure to toluene. This abuse may have adverse effect on their health. This study was therefore designed to investigate the effect of oral exposure to toluene on haematological parameters using male albino rats as model. Twenty animals were randomly divided into 4 groups of 5 rats each. Group A (Control) received 0.5 ml of olive oil (vehicle) while groups B, C and D received 31.8, 63.6 and 127.2 mg/kg respectively of toluene for 21 days by oral gavage. At the end of the treatments, the animals were anaesthetized and blood samples were collected for haematological investigations. No significant ($p > 0.05$) variation occurred in the mean values of PCV, haemoglobin concentration, RBC and platelet counts in comparison with the control. There was a significant ($p < 0.05$) increase in WBC count with no significant ($p > 0.05$) change in the differential leucocyte counts relative to the control. Oral administration of toluene as used in this study may be toxic to health depending on the dose and duration of exposure.

Keywords: toluene, blood, toxicity, solvent abuse

Introduction

In recent times, toluene has emerged as the most commonly abused solvent [1,2] and which, with increased dose and duration of exposure, can lead to toxicity. Toluene is an organic hydrocarbon used in the manufacturing of dyes, nail, shoe polish, inks and paint thinners. It is widely used in cosmetic industry and is also a component of nylon and plastic bottles [2]. Toxicity of toluene can occur from accidental or deliberate inhalation of fumes, ingestion or transdermal absorption, toluene abuse or “glue sniffing” which has become rampant, especially among young people, since it is readily available and affordable [2]. Among children and adolescents, toluene is frequently abused by dousing cloth with paint, inks and the like, and placing it over the nose and mouth for inhalation in a bid to get intoxicated and a sensation of euphoria. Toluene, when inhaled, is known to be readily absorbed into the bloodstream.

Blood, as integral component of the body system, is used to detect any disorder or anomaly arising from exposure to all forms of injuries that can adversely affect health. Haematological study is important in assessing the toxicity of drugs and pollutants in the body since blood is the major transport system of the body [3]. According to Oke *et al.* [4], anything that affects the blood will to a great extent affect the entire

36 body either adversely or moderately in terms of health, growth, maintenance and reproduction. It is against
37 this background therefore that this study was designed to investigate the effect of toluene toxicity on blood
38 parameters using male albino rats as model. This study has become necessary considering the increasing rate
39 at which young people engage in the abuse of organic solvents in order to get to a point of ecstasy.

40 **Material and Methods**

41 **Chemicals and Reagents**

42 Toluene, with CAS No: 108-88-3, was purchased from Bernaco Enterprises Nigeria as clear colourless
43 liquid with pleasant aromatic petroleum odour. The desired doses were prepared in Goya® olive oil which
44 was purchased from the supermarket.

46 **Animals and Treatment**

47 Twenty (20) mature male albino rats weighing an average of 200g, purchased from the Animal House of
48 Department of Pharmacology, College of Health Sciences, University of Port Harcourt were used for this
49 study. The rats were acclimatized for two (2) weeks before the study was commenced. They were fed *ad*
50 *libitum* with commercially sourced feed (Top Feeds Nigeria Limited) and supplied with clean drinking water
51 all through the study. After acclimatization, the animals were randomly assigned to four (4) groups – A, B, C
52 and D. Group A served as the control and was given 0.5ml of olive oil (vehicle) while the treatment groups
53 B, C and D received 31.8 mg/kg, 63.6mg/kg and 127.2 mg/kg, respectively of toluene which corresponded
54 to 1/20, 1/10 and 1/5 of the LD50 which is 636mg/kg according to Doro-on [5]. Treatments were by oral
55 gavage daily for 21 days. At the end of the treatments, the animals were anaesthetized and blood samples
56 were collected by cardiac puncture into EDTA bottles. The collected blood samples were used for the
57 estimation of haematological parameters such as packed cell volume (PCV), haemoglobin concentration
58 (HB), red blood cell count (RBC), white blood cell count (WBC), platelets count, lymphocyte and neutrophil
59 levels according to Cheesebrough [6].

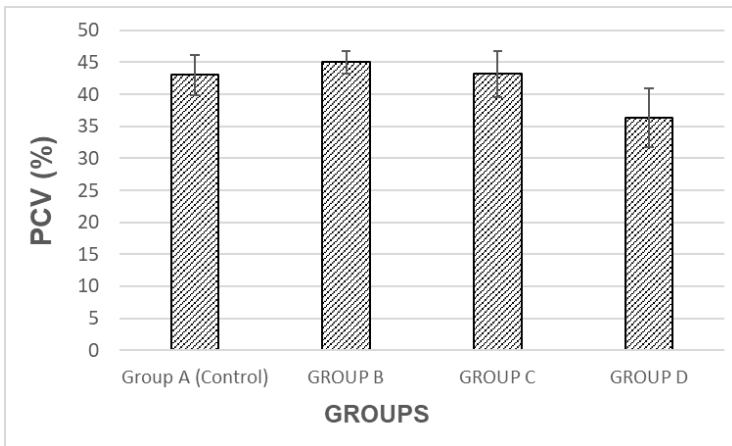
61 **Statistical Analysis**

62 Statistical analysis was done using SPSS 21. All values were expressed as mean \pm SEM and data were
63 assessed by one-way ANOVA followed by the Tukey post-test. The significance level was set at $p < 0.05$.

65 **Results**

66 The effect of different doses of toluene on various haematological parameters are summarized in figures 1 -
67 5 and Table 1. Treatment of rats for 21 days with 31.8, 63.6 and 127.2 mg/kg doses of toluene had no
68 significant ($p > 0.05$) effect on PCV, haemoglobin concentration, RBC and platelet counts relative to the
69 control as shown in figures 1 - 4.

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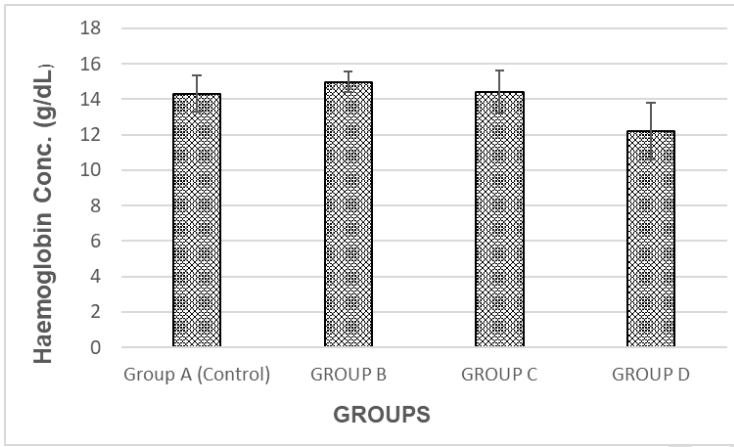


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72 **Fig. 1.** Effect of Toluene on Packed cell volume (PCV) of rats treated for 21 days. Results are given as mean
73 \pm SEM for 5 rats in each group. Experimental groups are compared with group A (control). No significant
74 difference at a 95% confidence interval ($p > 0.05$). Groups A, B, C and D represent the control (given 0.5 ml
75 olive oil), 31.8 mg/kg treated rats, 63.6mg/kg treated rats and 127.2 mg/kg treated rats, respectively.

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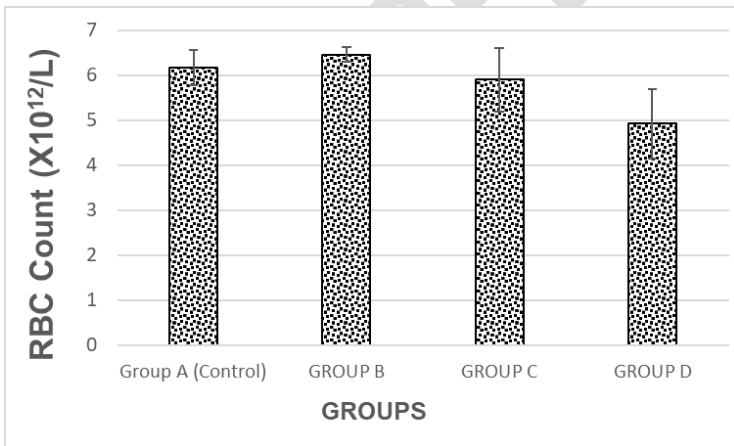
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78 **Fig. 2.** Effect of Toluene on Haemoglobin Concentration of rats treated for 21 days. Results are given as
 79 mean \pm SEM for 5 rats in each group. Experimental groups are compared with group A (control). No
 80 significant difference at a 95% confidence interval ($p > 0.05$). Groups A, B, C and D represent the control
 81 (given 0.5 ml olive oil), 31.8 mg/kg treated rats, 63.6mg/kg treated rats and 127.2 mg/kg treated rats,
 82 respectively.

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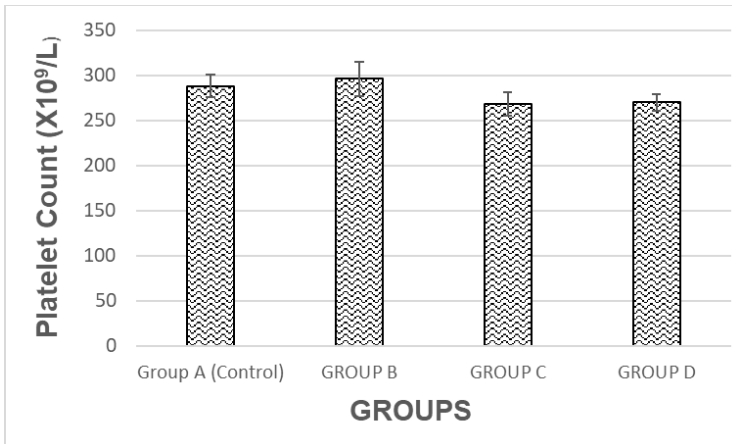


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85 **Fig. 3.** Effect of Toluene on Red Blood Cell (RBC) Count of rats treated for 21 days. Results are given as
 86 mean \pm SEM for 5 rats in each group. Experimental groups are compared with group A (control). No
 87 significant difference at a 95% confidence interval ($p > 0.05$). Groups A, B, C and D represent the control

88 (given 0.5 ml olive oil), 31.8 mg/kg treated rats, 63.6mg/kg treated rats and 127.2 mg/kg treated rats,
89 respectively.

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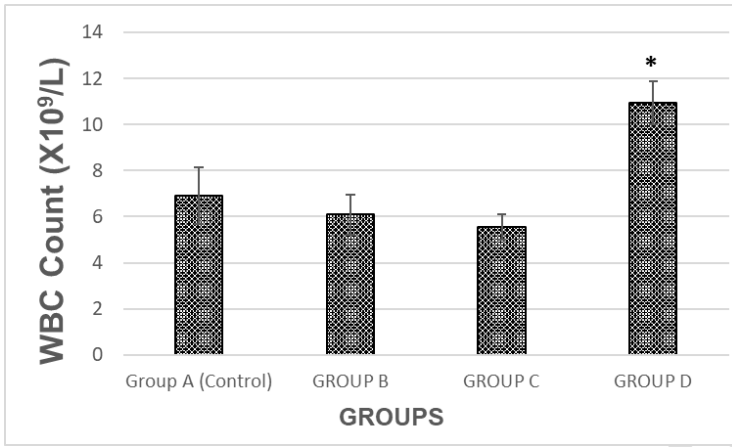


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94 **Fig. 4.** Effect of Toluene on Platelet Count of rats treated for 21 days. Results are given as mean \pm SEM for
95 5 rats in each group. Experimental groups are compared with group A (control). No significant difference at
96 a 95% confidence interval ($p > 0.05$). Groups A, B, C and D represent the control (given 0.5 ml olive oil),
97 31.8 mg/kg treated rats, 63.6mg/kg treated rats and 127.2 mg/kg treated rats, respectively.

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100 Figure 5 shows that treatment of rats for 21days with 31.8 and 63.6 mg/kg doses of toluene produced no
101 significant ($p > 0.05$) change on the WBC count in relation to the control. However, only toluene treated
102 group D (127.2 mg/kg) showed significant increase ($p < 0.05$) in the WBC count in comparison with the
103 control.

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106 **Fig. 5.** Effect of Toluene on White Blood Cell (WBC) count of rats treated for 21 days. Results are given as
 107 mean \pm SEM for 5 rats in each group. Experimental groups are compared with group A (control). * indicates
 108 a significant difference at $p < 0.05$. Groups A, B, C and D represent the control (given 0.5 ml olive oil), 31.8
 109 mg/kg treated rats, 63.6mg/kg treated rats and 127.2 mg/kg treated rats, respectively.

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111 The differential leucocyte count which involves the neutrophil, eosinophil, lymphocyte and monocyte counts
 112 did not change markedly ($p > 0.05$) with toluene treatment (Table 1).

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Table 1: Effect of Toluene on Differential Leucocyte Count of rats exposed for 21 days

PARAMETERS GROUPS	Neutrophils (%)	Lymphocytes (%)	Eosinophils (%)	Monocytes (%)
A	30.60 \pm 2.79	67.40 \pm 2.79	0.60 \pm 0.40	1.40 \pm 0.60
B	36.6 \pm 3.33	62.60 \pm 3.50	0.00 \pm 0.00	0.80 \pm 0.49
C	26.40 \pm 1.36	70.40 \pm 1.63	1.20 \pm 0.37	2.00 \pm 0.55
D	34.60 \pm 1.72	63.60 \pm 1.57	0.40 \pm 0.40	1.40 \pm 0.87

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115 Results are given as mean \pm SEM for 5 rats in each group. Experimental groups are compared with group A
116 (control). No significant difference at a 95% confidence interval ($p > 0.05$). Groups A, B, C and D represent
117 the control (given 0.5 ml olive oil), 31.8 mg/kg treated rats, 63.6mg/kg treated rats and 127.2 mg/kg treated
118 rats, respectively.

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120 Discussion

121 Haematological parameters are health markers which aids diagnoses and the evaluation of physiological and
122 pathological status of animals. According to Olafedehan *et al.* [7], blood acts as a pathological reflector of
123 the status of exposed animals to toxicant and other conditions. From this study, oral ingestion of toluene by
124 albino rats produced no significant effect on their PCV, RBC count, haemoglobin concentration and platelet
125 count. Although there was a significant increase in the WBC count at the highest dose of 127.2mg/kg, the
126 differential leucocyte count was not affected. This increase in mean WBC count could be attributed to the
127 presence of toluene in the body.

128

129 Higher WBC count is usually associated with prevalence of stress, inflammation, infection, allergy or certain
130 diseases [8]. Similarly, animals with high WBC counts produce antibodies during the process of combating
131 the causative agents through phagocytosis [9]. The elevated WBC count triggered by the presence of
132 causative agent ranging from biological, physical, chemical or thermal agents, in turn, leads to immunity to
133 diseases, infection or allergy as a result of the generated antibodies in the body. This finding is in line with
134 the work done by Ita and Udofia [10], who reported that the increase in WBC count of rats that orally
135 ingested gasoline for 21 days could be a defensive mechanism developed by the body against toxicity of the
136 gasoline constituents, which include toluene.

137

138 The non-significant change recorded in the differential leucocyte count suggests that the immune responses
139 of the body to infection may not have been compromised. [11]. This finding is similar to that of Obinna *et al.*
140 [12] who reported that rats treated with polar leaf extracts of *Portulaca oleracea* for 60 days, showed a
141 significant increase in the WBC count, with a non-significant variation in the differential leucocytes count.

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143 Conclusion

144 The present study, which provided insight into the toxicity of subacute exposure to toluene, concludes that
145 oral administration of toluene as used in this study may be toxic to blood in particular and the entire body

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146 system in general, depending on the dose and duration of exposure. Further study on the effect of toluene in
147 experimental animal for a longer duration of exposure is recommended.

148 **Competing Interests**

149 Authors have declared that no competing interests exist.

150 **Ethical Approval**

151 All authors hereby declare that "Principles of laboratory animal care" (NIH publication No. 85-23, revised
152 1985) were followed, as well as specific national laws where applicable. All experiments have been
153 examined and approved by the appropriate ethics committee

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