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The effects of acute gasoline vapour inhalation on some haematological indices of albino Wistar rats

Chukwudi Onyeka John Okonkwo[1], Ailende Daniel Ehileboh[1], Eddy Nwobodo[1], Charles Chijioke Dike[2*]

[1]*Department of Human Physiology, Faculty of Basic Medical Sciences, College of Health Sciences, Nnamdi Azikwe University, P.M.B. 5001, Nnewi Campus, Anambra State, Nigeria*

[2]*Department of Human Biochemistry, Faculty of Basic Medical Sciences, College of Health Sciences, Nnamdi Azikwe University, P.M.B. 5001, Nnewi Campus, Anambra State, Nigeria*

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ABSTRACT

Objective: To find out if Gasoline vapour has some effects on haematological indices when inhaled by experimental rats.

Methods: The standard method for laboratory operating procedure recommended by World Health Organization was used in all the analysis done. Forty two albino Wistar rats comprising twenty one males (160–220 g) and twenty one females (140–190 g) were sampled into six groups consisting of four test groups and two control groups. The test groups were exposed to gasoline vapour for twenty one days. Test group one were exposed to gasoline for 30 min while test group two were exposed to gasoline vapour for 1 h daily. At the end of twenty one days of exposure, blood samples were collected from the rats and their haematological parameters were estimated. Statistical analysis was done using windows SPSS version 16.

Results: The results showed a significant decrease ($P < 0.05$) in the red blood count and white blood count of exposed rats and increases in packed-cell volume, haemoglobin, mean corpuscular volume, and mean corpuscular haemoglobin respectively. These increases were significant ($P < 0.05$) in test group two. There was no significant change in the mean corpuscular haemoglobin concentration of the rats in the test groups when compared to the control groups of both sexes.

Conclusions: The results obtained suggest that inhalation exposure to gasoline may result in pancytopaenia and a significant fluctuation in the red blood cell-dependent haematological indices.

1. Introduction

The use of crude oil products has increased globally due to increase in population of the world. This has resulted in increased exposure to the products. This is because man invented a lot of equipments and materials which make use of the products in order to help him better his life. In a third world country like Nigeria who depends mostly on crude oil exploration for her foreign earning, there is no serious measures being put in place by the Government to checkmate pollution of the environment by crude oil products like gasoline. This is the major cause of Niger-Delta Militancy in Nigeria today. The companies that explore crude oil in some parts of Niger-Delta, Nigeria pollute the most of the rivers and farms, thereby destroying the major source of living for the Niger-Delta people who make a living by fishing. Hence, militancy occurs in the area. There has been several cases of explosion of crude oil pipe lines due to lack of maintenance, resulting in the pollution of the immediate environment with its fractions such as gasoline. Gasoline which is one of the fractional distillation products of crude oil is an inflammable volatile mixture of toluene, xylene, benzene and over a hundred other hydrocarbons[1]. The hydrocarbons present in gasoline include a variety of branched and straight chains of saturated and unsaturated aliphatic compounds as well as aromatic compounds[2]. In general, it contains alkenes, alcohol and ether[3]. Furthermore, several additives such as antiknocks and octane enhancer are added to gasoline to enhance its performance. In unleaded gasoline,

*Corresponding author: Charles Chijioke Dike, Department of Human Biochemistry, Faculty of Basic Medical Sciences, College of Health Sciences, Nnamdi Azikwe University, P.M.B. 5001, Nnewi Campus, Anambra State, Nigeria.
Tel: +234 08038641948
E-mails: dikehcharles@yahoo.com, cc.dike@unizik.edu.ng
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these include methyl tertiary butyl ether, ethyl tertiary butyl ether, and tertiary butyl alcohol while unleaded gasoline, alkyl lead compounds such as tetramethyl lead and tetraethyl lead are used[4]. The fractional distillation products of petroleum are used for various purposes by human beings at homes for domestic purposes, in manufacturing and petrochemical industries. Gasoline which is one of crude oil products used on routine basis may have some direct medical effects on the immediate users. Gasoline is considered an environmental pollutant and toxicant which has adverse effects on some organs like kidney, lungs and liver. It is found abundant in the atmosphere as a result of its volatile nature[5,6]. Some of the ways of being exposed to gasoline include oral, skin contact and inhalation[6,7]. Gasoline vapour inhalation has been reported to change blood chemistry and induce anaemia by causing bone marrow hypoplasia in experimental animals[5]. This is due to the heavy metal contained in gasoline. It has become necessary to investigate the effects of acute gasoline exposures on some haematological indices due to its apparent health implications. This study was therefore embarked upon with the main aim of investigating the effects of gasoline vapour inhalation on haematological indices which include red blood cell (RBC), packed cell volume (PCV), haemoglobin (Hb), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular volume (MCV) and white blood cell (WBC).

2. Materials and methods

2.1. Experimental animals

Matured Wistar rats of both sexes (twenty one each) were purchased and acclimatized at the animal house of College of Health Sciences, Nnamdi Azikiwe University-Akwa, Nnewi Campus, Anambra State, Nigeria. The animals were kept in six standard animal cages. After ten days of acclimatization, the rats weighed between 140 g and 210 g. The animals were exposed to 12:12 h of light and dark periods. They were allowed for free access to feed and drinking water.

2.2. Source of gasoline

Four litres of commercially sold gasoline was purchased from Texaco Filling Station, Nnewi, Anambra State, Nigeria.

2.3. Experimental design

The animals were divided into six (6) groups of male or female rats consisting of two control groups and four test groups as follows: Group Am (male control group; $n = 7$), and Group Af (female control group; $n = 7$). Group Bm (male test group 1; $n = 7$), Group Bf (female test group 1; $n = 7$), Group Cm (male test group 2; $n = 7$) and Group Cf (male test group 3; $n = 7$).

2.4. Exposure to gasoline

The test groups were exposed to gasoline vapour for twenty one days. Group B males and Group B females were exposed to gasoline vapour for 30 min daily while Group C males and Group C females were exposed to gasoline vapour for 1 h daily. Exposure was achieved by soaking 100 g of cotton wool in 25 mL of commercially procured gasoline. The gasoline-soaked cotton wools were placed on the roofs of the rat cages. The vapour was allowed to mix with the ambient air of the cages. The control groups consisting of Group A male and Group A female were not exposed to gasoline vapour. After twenty one days, the animals were euthanized and blood was collected by cardiac puncture using a 2 mL syringe. The blood was immediately transferred into an EDTA anticoagulant coated blood bottles.

The haematological parameters of the blood were then analysed. The PCV was determined by the use of micro-haematocrit method. The total RBC and WBC were counted using the improved Neubauer counting chamber under a light binocular electric microscope *i.e.*, estimation by haemocytometric method.

2.5. Statistical analysis

Statistical analysis of the data was done using Windows SPSS version 16 software. The results were expressed as mean ± SE.

3. Results

The results obtained as shown in Table 1 in which the mean Hb, PCV, RBC, WBC, MCH, MCV and MCHC of the control group were compared with the means of the test groups for males. Table 2 compares the control group with the test groups for females. The mean results for the control male group for RBC, WBC, PCV, Hb, MCV, MCH and MCHC were (8.54 ± 0.25) × 10^6/mm^3, (37.67 ± 0.25) × 10^3/mm^3, (34.71 ± 0.01)%, (11.29 ± 0.42) g/dL, (40.29 ± 2.48) fL, (13.20 ± 0.81) pg, and (33.20 ± 0.14) g/dL, respectively. The mean results for male test group one RBC, WBC, PCV, Hb, MCV, MCH and MCHC were (8.89 ± 0.31) × 10^6/mm^3, (32.60 ± 1.08) × 10^3/mm^3, (39.20 ± 0.01)%, (12.71 ± 0.29) g/dL, (43.57 ± 1.97) fL, (14.00 ± 0.53) pg, and (33.00 ± 0.00) g/dL, respectively while the mean results for male test group two are also presented in Table 1. There were significant decrease ($P < 0.05$) in the RBC and WBC of test group two. There was, however, a non-significant increase and decrease in the RBC and WBC respectively when compared to the control. A significant increase in the PCV and Hb values was observed in both test groups. A significant increase ($P < 0.05$) was observed in the MCV and MCH of test group two while non-significant increase was observed in test group one. However, the increases observed in the MCHC of both test groups were not significant. The mean results for the female control group for RBC, WBC, PCV, Hb, MCV, MCH and MCHC were (6.16 ± 0.31) × 10^6/mm^3,

Table 1
Effects of gasoline vapour inhalation on some haematological parameters of male Wistar rats.

Parameters	Group Am	Group Bm	Group Cm
RBC (× 10^6/mm^3)	8.54 ± 0.25	8.89 ± 0.31	5.0 ± 0.20
PCV (%)	34.71 ± 0.01	39.20 ± 0.01	38.71 ± 0.01
Hb (g/dL)	11.29 ± 0.42	12.71 ± 0.29	12.57 ± 0.30
MCV (fL)	40.29 ± 2.48	43.57 ± 1.97	77.43 ± 2.71
MCH (pg)	13.20 ± 0.81	14.00 ± 0.53	25.43 ± 1.15
MCHC (g/dL)	33.20 ± 0.14	33.00 ± 0.00	33.14 ± 0.88
WBC (× 10^3/mm^3)	37.67 ± 0.25	32.60 ± 1.08	27.71 ± 2.83

Results are expressed as mean ± SE for seven rats in each group.

Table 2
Effects of gasoline vapour inhalation on some haematological parameters of female Wistar rats.

	Group Af	Group Bf	Group Cf
RBC ($\times 10^6$/mm^3)	6.16 ± 0.31	7.43 ± 0.22	3.67 ± 0.22
PCV (%)	36.29 ± 0.04	34.29 ± 0.01	35.00 ± 0.01
Hb (g/dL)	11.71 ± 0.18	11.00 ± 0.38	11.14 ± 0.26
MCV (fL)	58.86 ± 3.18	45.71 ± 1.97	96.43 ± 6.99
MCH (pg)	19.29 ± 1.06	14.86 ± 0.70	32.00 ± 2.37
MCHC (g/dL)	33.14 ± 0.14	33.00 ± 0.00	33.00 ± 0.00
WBC ($\times 10^3$/mm^3)	35.29 ± 1.29	33.00 ± 0.65	23.29 ± 0.97

Results are expressed as mean ± SE for seven rats in each group.

$(35.29 \pm 1.29) \times 10^3$/mm^3, (36.29 ± 0.04)%, (11.71 ± 0.18) g/dL, (58.86 ± 3.18) fL, (19.29 ± 1.06) pg, and (33.14 ± 0.14) g/dL. The mean results for the female test group one on RBC, WBC, PCV, Hb, MCV, MCH and MCHC were $(7.43 \pm 0.22) \times 10^6$/mm^3, $(33.00 \pm 0.65) \times 10^3$/mm^3, (34.29 ± 0.01)%, (11.00 ± 0.38) g/dL, (45.71 ± 1.97) fL, (14.86 ± 0.70) pg, and (33.00 ± 0.00) g/dL respectively while the mean results for the female test group two can also be found in Table 2. Significant decrease ($P < 0.05$) was observed in the RBC of both test groups when compared to the control. A significant decrease ($P < 0.05$) was observed in the WBC of test group two while the decrease observed in test group one was not significant. A non-significant decrease was observed in the Hb of both test groups. There was no significant change in the PCV of both test groups. A non-significant increase and a significant increase were observed in the MCV and MCH of test group one and test group two respectively. There was no significant increase in the MCHC of both test groups.

4. Discussion

The results obtained from this research work showed significant changes in the haematological parameters of the albino rats used. These changes were higher in test group two which were exposed for a longer period. These findings obtained in this experiment are in line with the observations of Ita and Sirdah[6,8]. The decrease in RBC could also be attributed to the cytotoxic effects of the various gasoline constituents. These may cause oxidative stress thereby damaging the membrane of the RBC[9]. Toxic components of gasoline such as benzene and lead are capable of altering the functions of DNA of a cell. There have been several established cases of toxic effects of other petroleum fractions such as petrol, kerosene and engine oil on human blood and other organs of the body. This is because these petroleum fractions also contain heavy metals which are toxic to human system. The increase in the RBC indices vis-à-vis PCV, Hb, MCV, MCH, and MCHC observed in this experiment may be due to dysfunction in the secretion of erythropoietin caused by the exposure as a result of the heavy metal content. This could be due in part to the hypoxia caused by the exposure to gasoline vapour and also to the decrease in the RBC. However, the higher decrease observed in test Group 2 which were exposed to gasoline vapour for a longer duration supported the claim of Okoro *et al.*[5], that exposure to petroleum fumes decreases red cells in a manner that is time-dependent. This suggests that chronic exposure to gasoline may have more adverse effects on the haematological parameters. The decrease in RBC observed in this experiment is not in line with the increased RBC in people exposed to gasoline vapour reported by Sirdah *et al.*[8]. Furthermore, the increase in the PCV, Hb and other RBC indices observed in this experiment tallies with the finding of Sirdah *et al.*[8], but does not tally with the reports of Okoro *et al.*[5]. This difference may be due to environmental factors. Also, it was observed in this experiment that the male rats were more susceptible to the changes in haematological indices induced by the gasoline vapour when compared to the females. These changes may be as a result of hormonal imbalance.

This study has shown that exposure of male and female rats to gasoline vapour may be hematotoxic. This toxicity may be, however, due to the toxic components contained in the gasoline vapour. The hematotoxic effects were also time-dependent. The concomitant relative pancytopaenia and the fluctuations observed in the RBC dependent indices of the rats could also occur in humans who are exposed to gasoline vapours on daily basis. However, before that the above results are extrapolated to human beings, epidemiological studies on "the effects of acute gasoline exposures" on human beings need to be carried out.

Conflict of interest statement

The authors report no conflict of interest.

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