

HAEMATOLOGIC AND BIOCHEMICAL IMPLICATIONS OF INHALATION OF FUMES OF PETROLEUM PRODUCTS

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ABSTRACT

Aims: To investigate the Haematologic and biochemical implications of exposure to fumes of petroleum products in human subjects.

Methods: A total of 100 subjects (40 males and 60 females) aged between 18-30 years participated in the study. Each gender was further categorized into two groups of 10 controls and 30 tests in males and 20 controls and 40 tests in females. Test Group 1 subjects were exposed to fumes of petroleum products for two years and below while test Group 2 subjects were exposed for more than two years. Samples of blood were collected daily and subjected to haematological and biochemical analysis.

Results: There was a significant decrease in red blood cell counts, white blood cell counts, pack cell volume and haemoglobin concentration in the test Groups 1 and 2 compared to control and a significant increase in alkaline phosphatase, alanine transaminase and aspartate aminotransferases. The odds/odds ratios of subjects becoming anaemic progressively rose from less than 1 in the control to greater than 1 in test groups.

Conclusion: Frequent exposure to fumes of petroleum products causes reduction in haematological indices and deleterious effect to the liver cells which worsens with prolonged exposure.

Key Words: Petroleum Fumes, Haematological, Biochemical Parameters.

INTRODUCTION

Fractional distillation of crude oil yields different fractions of petroleum products of which petrol, kerosene and diesel are constituent parts. These fractions of crude petroleum contain aliphatic, aromatic and a variety of saturated and unsaturated hydrocarbons having straight and branched chains (Anderson et al., 1995). Fumes of petroleum products (Petrol, Kerosene and Diesel) are ubiquitous in our environment and the

common sources of contact or exposure are petrochemical industries (refineries, oils fields, filling stations) and homes. The applications of petroleum products as fuels for vehicles, chemical feedstock for industries, cooking and lighting fuels in homes and outside homes, as well as for therapeutic reasons (Hockabey et al., 1995) have resulted in direct exposure of these products to a good percent of the populace. However, the most affected are those who are occupationally exposed

(those who work directly with the petroleum industries) to the fumes (Rothman et al., 1996). It has been demonstrated that lower concentrations of saturated hydrocarbons than unsaturated aromatic hydrocarbons are detected in human and animal blood after inhalation of petroleum vapour through chronic exposure, (Zahlsen et al., 1993). Occupational exposure to petroleum fumes have been shown to produce toxic effects on various organs and systems including the respiratory, immune and nervous systems. Organs such as the heart, skin and kidneys are as well affected, resulting in various diseases ranging from mutagenic, immunotoxic, carcinogenic, genotoxic, and neurotoxic manifestations (Becker CE 1985; Klassen CD 1990; d'Azevedo et al., 1996; Rabble GK and Wong O 1996; Ross D 1996; Rothman et al., 1996). Hydrocarbons like benzene, metals like lead and volatile nitrates from petroleum products have all been shown to inhibit the haematopoietic component in the red marrow and spleen and to cause other harmful effects on the lymph nodes (Marieb EN 1995). In another study benzene has been shown to induce leukaemia during occupational exposure (Austin et al., 1988). Hydrocarbons and other constituents of petroleum products are extensively metabolized in the liver (Sims P 1980). Ueng et al., (1998) reported that exposure of rats to motorcycle exhaust and organic extracts of the exhaust particulate caused a dose- and time-dependent increase in cytochrome P-450-dependent monooxygenases and glutathione-S-transferase in the liver, kidney and lung microsomes. Since kerosene and petrol contain some of these constituents, chronic or frequent exposure to their fumes may affect the normal liver functions. The appearance of toxicity of xenobiotics is usually determined biochemically by monitoring some plasma enzymes and lipids. A rise in AST, ALT, ALP, TG and cholesterol are common indices for measuring levels of damage of the liver cells (Abdel-Baset et al., 1997). The main objective of this study was to investigate the effect of inhalation of fumes of petroleum products on some haematological and biochemical parameters in humans occupationally exposed to them. these include Packed cell Volume (PCV), Red blood cell (RBC), White blood cells (WBC), Haemoglobin (Hb), alkaline phosphatase (ALP),

alanine transaminase (ALT) and aspartate aminotransferases (AST).

MATERIALS AND METHODS

Subjects: This study was conducted on adult human subjects of age range between 18 to 30 years who gave informed consent to the study. Questionnaires were distributed and filled; candidates who met the criteria for participation in this study were admitted into the study. Several fuel stations located within the Port Harcourt metropolis were used as sites for the study for test subjects, while students who were not exposed to petroleum products served as controls. A total of one hundred subjects took part in this study of which forty were males and sixty females. These were divided into two test groups and a control group for both sexes. All control groups were classified as Group A and people who had worked for 2 years or less (test group 1) were classified as Group B, while test 2 group (people who had worked for more than 2 years) were classified as Group C. Venous blood (5 ml) was taken from a peripheral vein on the arm of each subject and immediately transferred into sterile potassium EDTA anticoagulant bottles. The blood samples collected were analyzed on a daily basis. The Hb, PCV, RBC and WBC were done with auto-haematological analyzer with the use of diluents, cell lyse and E-Z cleanser as reagents. Furthermore, ALP, ALT and AST were done with the Auto-chemistry analyzer using the kit method. **Statistical analysis:** This was carried out by employing the student's t-test to compare mean values of test groups with control. A value of $P \leq 0.05$ was considered to be statistically significant. Furthermore, the odds and odds ratios for the various RBC indices were calculated to determine the likelihood of subjects becoming anaemic in both control and test groups. Values of odds and odds ratios which were below one were considered unlikely to become anaemic while values above one were more likely to be anaemic, after the method of Bandolier (2006). In this method, the odds of an event are calculated as the number of events divided by the number of non-events. An odds-ratio result is calculated by dividing the odds in the treated, exposed or test group by the odds in the control group.

RESULTS

The results obtained are presented as comparison of mean \pm SEM values of Hb, PCV, RBC, WBC, ALP, ALT and AST of control group with test group 1 and test group 2 for males and females. They are presented as figures 1 through 7. The Hb, PCV, RBC and WBC for the control males are 14.67 ± 0.15 (g/dl), 45.55 ± 0.17 (%), 4.52 ± 0.18 ($10^6/\text{mm}^3$), and 5.49 ± 0.18 ($10^3/\text{mm}^3$) respectively. The values of Hb, PCV, RBC and WBC of test group 1 males are 12.53 ± 0.20 (g/dl), 40.82 ± 0.18 (%), 3.83 ± 0.19 ($10^6/\text{mm}^3$), and 4.60 ± 0.07 ($10^3/\text{mm}^3$) respectively. The result of test group 2 for Hb, PCV, RBC and WBC were 10.99 ± 0.01 (g/dl), 37.46 ± 0.23 (%), 3.23 ± 0.08 ($10^6/\text{mm}^3$), and 4.08 ± 0.03 ($10^3/\text{mm}^3$) respectively. These results show significant difference ($P \leq 0.001$, 0.001, 0.004 and 0.01) between control and test groups 1 and 2 for Hb, PCV, RBC and WBC respectively. Similarly, the Hb, PCV, RBC and WBC for the control females are 13.55 ± 0.25 (g/dl), 40.34 ± 0.33 (%), 3.59 ± 0.23 ($10^6/\text{mm}^3$), and 3.33 ± 0.17 ($10^3/\text{mm}^3$) respectively. The values of Hb, PCV, RBC and WBC of test group 1 females are 11.45 ± 0.12 (g/dl), 40.34 ± 0.33 (%), 3.59 ± 0.23 ($10^6/\text{mm}^3$), and 4.00 ± 0.01 ($10^3/\text{mm}^3$) respectively. The result of test group 2 for Hb, PCV, RBC and WBC were 9.99 ± 0.01 (g/dl), 35.97 ± 0.03 (%), 2.97 ± 0.04 ($10^6/\text{mm}^3$), and 3.33 ± 0.17 ($10^3/\text{mm}^3$) respectively. In a similar manner the results show

significant difference ($P \leq 0.001$, 0.001, 0.004 and 0.01) between control and test groups 1 and 2 for Hb, PCV, RBC and WBC respectively. The ALP, ALT and AST for control males are 25.88 ± 0.59 (u/l), 8.36 ± 0.32 (u/l), and 8.36 ± 0.07 (u/l), respectively. The values of ALP, ALT and AST of test group 1 males were respectively 36.00 ± 0.58 (u/l), 10.77 ± 0.24 (u/l), and 11.66 ± 0.17 (u/l), while ALP, ALT and AST values for test group 2 males were 70.00 ± 2.65 (u/l), 29.93 ± 0.58 (u/l), and 38.07 ± 1.6 (u/l) respectively. These results show significant difference ($P \leq 0.001$) between control and test groups 1 and 2 for ALP, ALT and AST. Similarly, ALP, ALT and AST for control females were 24.33 ± 0.88 (u/l), 7.11 ± 0.06 (u/l), and 8.10 ± 0.10 (u/l), respectively. The values of ALP, ALT and AST of test group 1 males were respectively 31.67 ± 1.76 (u/l), 9.40 ± 0.60 (u/l), and 11.22 ± 0.100 (u/l), while ALP, ALT and AST values for test group 2 males were 63.33 ± 2.33 (u/l), 27.69 ± 0.70 (u/l), and 36.73 ± 1.5 (u/l) respectively. In a similar manner, the results show significant difference ($P \leq 0.001$) between control and test groups 1 and 2 for ALP, ALT and AST. The results of Odds and Odds ratios presented in tables 1 and 2 respectively, shows that the Odds values for control in RBC, Hb and PCV were all less than 1, while those for test groups for RBC, Hb and PCV were all more than 1.

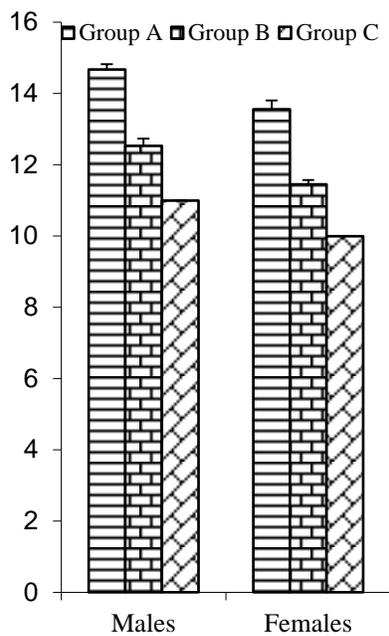
Table 1: Odds/odd ratios for anaemia in male subjects

RBC	n	Anaemic			Non anaemic	Odds (Anaemic/ Non anaemic)				Odds ration
Control	1	0	1		9	0	-	1	1	-
Test	1	5	1	1	4	2	-	7	5	18-18
Test	1	5	1	3	2	6	-		5	59-09
Hb										
Control	1	0	2		8	0	-	2	5	-
Test	1	5	1	2	3	4				1-6
Test	1	5	1	4	1	1			4	5-6
PCV										
Control	1	0	1		9	0	-	1	1	-
Test	1	5	1	3	2	6	-		5	59-09
Test	1	5	1	4	1	1			4	127-27

Table 2: Odds/odd ratios for anaemia in female subjects

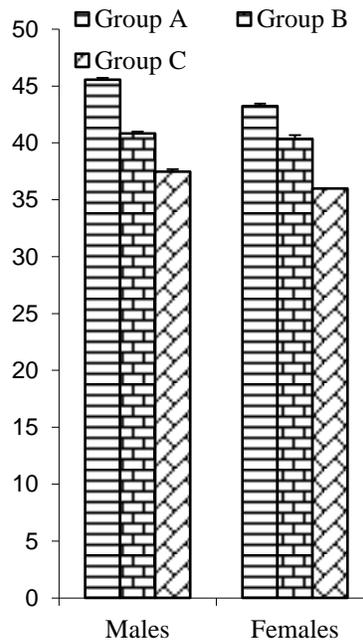
RBC	n	Anaemic			Non anaemic		Odds (Anaemic/ Non anaemic)			Odds ration
Control	2	0	2		1	8	0	1	1	-
Test	12	0	1	6	4		4			36-36
Test	22	0	1	8	2		9			81-82
Hb										
Control	2	0	1		1	9	0	0	5	-
Test	12	0	1	5	5		3			6 0
Test	22	0	1	7	3		5	6	7	113-4
PCV										
Control	2	0	2		1	8	0	1	1	-
Test	12	0	1	6	4		4			36-36
Test	22	0	1	8	2		9			81-82

Figure 1: Effect of petroleum products on Hb (g/dl)



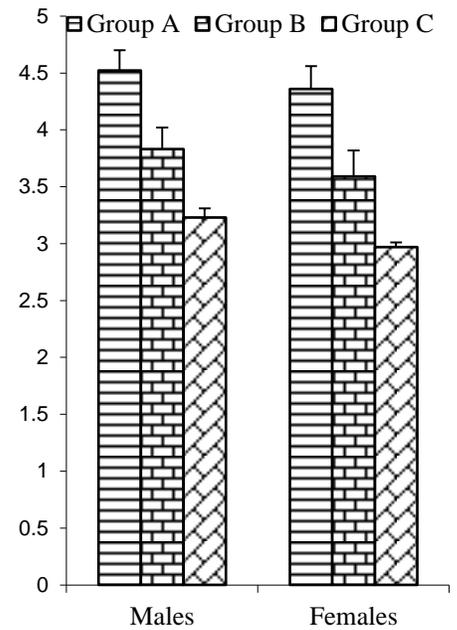
Group A= Non- petrol station attendants, Group B = Petrol station attendants who worked for less than two years and Group C = workers who have worked for two years and above.

Figure 2: Effect of petroleum products on PCV



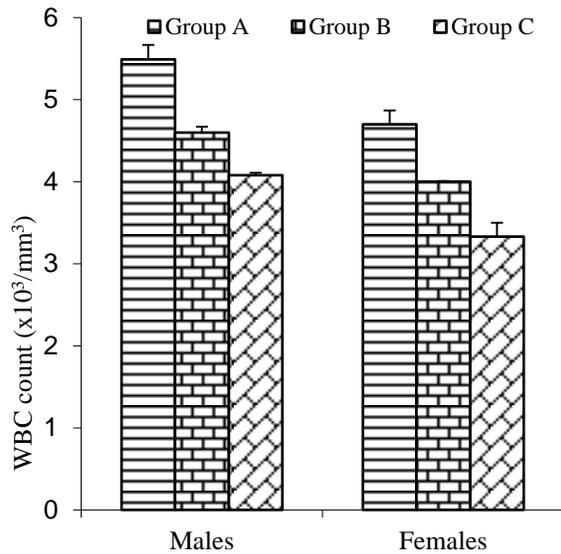
Group A = Non- petrol station attendants, Group B = Petrol attendants who have worked for less than two years. Group C= workers who have worked for two years and above.

Figure 3: Effect of petroleum products on RBC (x10⁶/mm³)



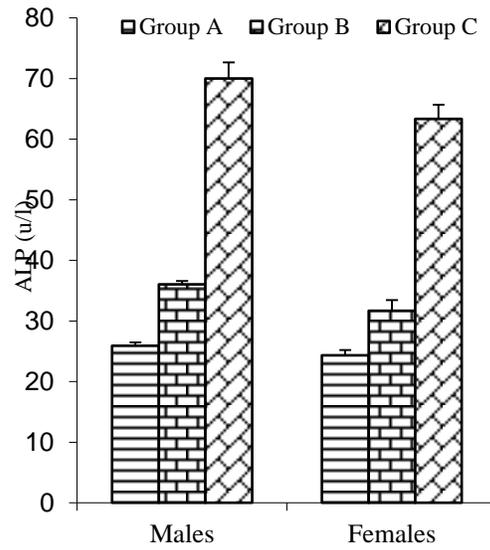
Group A = Non- petrol station attendants, Group B = Petrol station attendants who have work for less than two years and Group C = workers who have worked for two years and above.

Fig 4: Effect of petroleum products on WBC



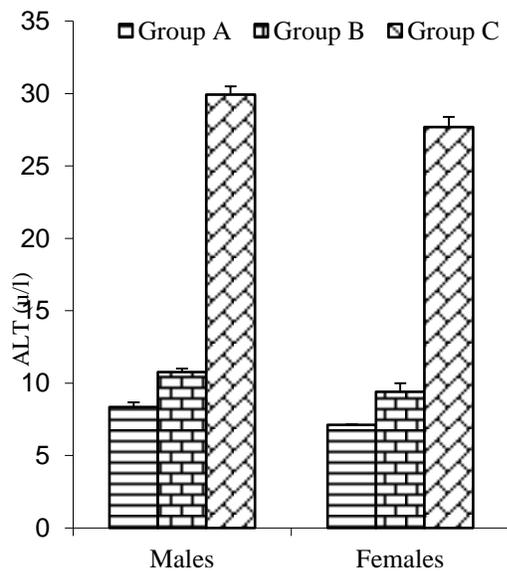
Group A = Non- petrol attendants, Group B=Petrol attendants who have work for less than two years and Group C=workers who worked for two years and above.

Fig 5: Effect of petroleum products on ALP



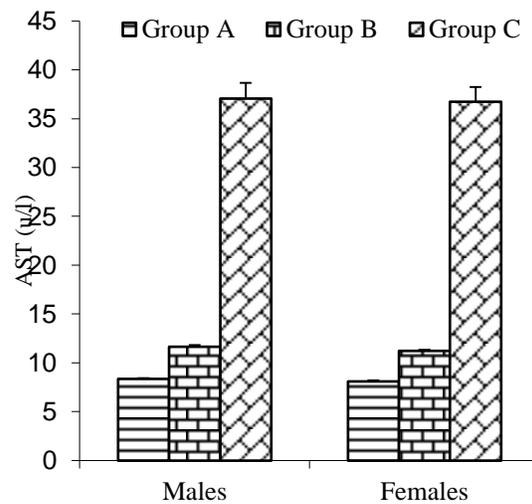
Group A= Non- petrol attendants, Group B= Petrol attendants who have work for less than two years. Group C= workers who worked for two years and above.

Fig 6: Effect of petroleum products on ALAT



Group A = Non- petrol station attendants, Group B = Petrol station attendants who have work for less than two years and Group C = workers who have worked for two years and above.

Fig 7: Effect of petroleum products on ASAT.



Group A =Non- petrol attendants, Group B =Petrol attendants who worked for less than two years. Group C= workers who worked for two years and above.

DISCUSSION

The haematological parameters Hb, PCV, RBC and WBC provide information on the general state of the blood of the subjects. This study has demonstrated that exposure to fumes of petroleum products cause a significant decrease in Hb, PCV, RBC and WBC values of subjects. The toxic components of fumes of petroleum products have been reported to change the blood chemistry and to induce anaemia by causing bone marrow hypoplasia in experimental animals (Marieb EN 1995). This study suggests a similar effect on humans. It has also been reported that the toxic components in petroleum such as benzene and lead activate the bone marrow and the resultant cytotoxic depressant effects mediated through disturbances in the DNA function leads to inadequate production of red blood cells and other formed elements (Rabble et al., 1996). This is in line with findings in this study. White blood cell whose principal function is in body defense against foreign bodies is achieved through leukocytosis and antibody production (Marieb 1995). In this study, there was a significant decrease in white blood cell count in both males and females exposed to fumes of petroleum products and the decrease was greater in those exposed for more than two years. Studies have reported that benzene produce haematological changes ranging from pancytopenia (deficiency of all cell elements of the blood) to absolute bone marrow aplasia, through its myelotoxic action (d'Azevedo et al., 1996). The decrease in WBC observed in this study could have possibly resulted from pancytopenia and leucopenia. The findings in this study is similar to previous reports of toxicity from constituents of crude oil (Ovuru SS and Ekweozor IKE 2004; Dede EB and Kagbo HD 2002). The result of this study agrees with the findings of Okoro et al., (2006), that the odds of a subject becoming anaemic increased progressively from control values to test group 1 values and were highest in test group 2. The odds were less than one in the control for Hb, PCV and RBC in both sexes, implying that the control subjects were not likely to become anaemic. Contrarily, subjects exposed to petroleum fumes for two years and below (test Group 1) and those exposed to petroleum fumes for more than two years (test group 2) all had odds and odds ratios above 1, implying that these subjects were more likely to

become anaemic. In fact those, with very large odd values are certain to become anaemic. This shows that exposure to fumes of petroleum products decreases red cell indices in a manner that is duration dependent. The level of ALT was significantly higher in test groups 1 and 2 when compared to the controls. It has been reported that the level of serum ALT activity increased because of liver injury in patients developing severe hepatotoxicity (Beckett et al., 1989). McIntyre NR (1992) reported a possible leakage of ALT from damaged cells, due to increased permeability of the hepatocellular membrane, or due to necrosis, indicating organ dysfunction. Similarly, the level of AST was also significantly higher in the test groups 1 and 2 than in the control. Increased activity of AST has been reported in CCl₄-intoxicated experimental animals (Abdel-Baset et al., 1997), which have been attributed to abnormal dynamic properties of cellular membranes following exposure to hydrocarbon fractions. Leighton et al., (1985) and Bondy et al., (1995) have reported that metabolism of aliphatic and aromatic hydrocarbons which are the major constituents of petroleum fumes have resulted in changes in the cell membrane due to reactive free radical species. Alkaline phosphatase activity in test groups 1 and 2 were significantly higher ($p < 0.001$) than those of the control. This implies that damages may have occurred in the liver cells, since the activity of this enzyme in the serum is reported to be increased in liver damage (Abdel-Baset et al., 1997). Alkaline phosphatase is involved in the transport of metabolites across the cell membranes, protein synthesis, synthesis of certain enzymes, secretory activities and glycogen metabolism. The increase in this enzyme activity may not be unconnected with a disturbance in the transport of metabolites or alteration in the synthesis of certain enzymes as in other hepatotoxic conditions (Sharma et al., 1995). In conclusion, the findings from this study reveal that, repeated exposure to fumes of petroleum products causes a depression of total white blood cell count as well as red blood cell count and its dependent haematological indices (PCV, Hb) and also elicit hepatotoxicity, thereby impairing the normal liver function. Petroleum workers therefore should have regular medical check-up to ascertain their health condition.

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