

Effects of Petroleum Products Inhalation on Hematological Parameters of Male Wistar Albino Rat

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International Journal of Life Science Research Archive, 2026, 10(02), 127-138

Publication history: Received on 25 April 2026; revised on 04 June 2026; accepted on 06 June 2026

Article DOI: <https://doi.org/10.53771/ijlsra.2026.10.2.0042>

Abstract

This study investigated the hematological effects of petroleum products exposure on male Wistar albino rats, focusing on key blood parameters such as packed cell volume (PCV), hemoglobin (HB), red blood cell (RBC) counts, neutrophils, lymphocytes, eosinophils, basophils and monocytes. Twenty-five rats were placed in a custom-built inhalation chamber, exposed to 500ml of petroleum products in a 1000ml container, it was placed in a different section in the inhalation chamber to avoid the rats drinking from it or pushing it down. The exposure was conducted for 1 hour per day, for 14 days. The rats were exposed to various petroleum products, including petrol, diesel, and kerosene, with assessments conducted at baseline, week 7, and week 14. Significant reductions in PCV, HB, and RBC counts were observed in exposed groups, indicating potential anemic conditions resulting from toxic exposure. Additionally, marked fluctuations in white blood cell (WBC) counts were noted, with increases in neutrophils and decreases in lymphocytes, suggesting an inflammatory response and immune system compromise. The study also documented variations in eosinophils and basophils, highlighting the complex immune dynamics affected by petroleum inhalation. While some recovery in hematological parameters was noted by week 14, persistent alterations underscore the risks of prolonged exposure of petroleum products to Wister rats.

Keywords: Hematological; Wistar Rats; Petroleum Products; Inhalation

1. Introduction

Petroleum is a mixture of different hydrocarbons and metals (10). The chemical composition of crude petroleum varies between geographical locations. Crude oil is refined into fractions of kerosene, diesel, petrol, heavy gas oils, lubricating oils, as well as residual and heavy fuels, among others. Kerosene, diesel and petrol are the most commonly used fractionated crude petroleum products. Petrol contains aliphatic, aromatic and a variety of other branched saturated and unsaturated hydrocarbons at variable concentrations (14). The constituents of the vapours from different fractions of petroleum products depend on the composition of their liquid forms, which varies with the brand and storage period.

Gasoline, kerosene and diesel are reported to contain predominantly, hydrocarbons with carbon atoms 4-10, 11-13 and 14-18, respectively (20). The volatility of these fractions varies with the predominant hydrocarbon species. Unleaded gasoline for instance, is reported to contain about 300 different hydrocarbon species, most of which are highly volatile and may evaporate if left exposed, to constitute ubiquitous chemical pollutants in the environment (12).

Exposure to various fractionated petroleum products has been reported to cause impairment of renal function evident by the derangement of serum electrolytes (Suleiman *et al.*, 2024). During the course of usage of these products, individuals are usually exposed to pollutants from petroleum products in their environments. Those that are

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occupationally exposed tend to bring greater risk of exposure to the petroleum products. Petrol is used as fuel for motor vehicles, generators and power plants. It is also used as pesticides and cleaning agent. The chemical pollutants from petrol vapours, like other known xenobiotics, maybe metabolically transformed into various metabolites in the body (19). Some of these metabolites may be very reactive, interacting in various ways with the metabolizing and excreting tissues (mainly the liver and kidneys) to elicit toxic effects (5). The interaction of these metabolites with the tissues may cause damage to the tissues. Hydrocarbons like benzene, metals like lead and volatile nitrates have been reported to produce harmful effects on bone marrow, spleen and lymph nodes (8). These toxic compounds destroy or inhibit the hematopoietic stem cell in the bone marrow. Benzene which is an aromatic hydrocarbon contained in petroleum product is known to induce leukemia during occupational exposure (3).

Metabolism of poisonous chemical substances takes place in the liver, which accounts for the organ's susceptibility to metabolic induced hepatotoxicity. The crude oil and its products generate free radicals and promote a variety of chemical reactions, such as depletion of reduced glutathiones or inducing lipid peroxidation. The inhalation of petroleum products poses a significant health risk, particularly due to the toxic compounds they release into the environment. Despite the widespread use of these products in various sectors, there remains a lack of comprehensive understanding of their specific effects on biological systems, particularly on blood parameters. Previous studies have documented the acute and chronic health effects associated with exposure to petroleum vapors, including respiratory issues, neurological disorders, and potential carcinogenic effects. However, the impact of these exposures on hematological indices—such as red blood cell counts, hemoglobin levels, and white blood cell counts—has not been thoroughly investigated, particularly in animal models like the Wistar albino rat. Hence, this study was conducted to determine the effect of petroleum products inhalation on blood parameters of male Wistar albino rat.

2. Materials and methods

2.1. Animal Model

Twenty-Five (25), Male Wistar albino rats gotten from University of Nigeria Nsukka, Department of Animal Physiology and were housed at Power-Tech Analytical and Scientific Research Laboratory, Independence Layout, Enugu, Enugu State. They were fed on standard rat chow and clean water *ad-libitum* and kept under normal room temperature of $25 \pm 2^\circ\text{C}$ with humidity of $45 \pm 5\%$.

2.2. Experimental Protocol

The rats were acclimatized for one week before the experiment. They were randomly assigned into five groups (n=5 per group):

- **Group A:** Blank Control (no exposure)
- **Group B:** Premium motor spirit exposure only
- **Group C:** Kerosene exposure
- **Group D:** Diesel exposure
- **Group E:** Mixed petroleum products exposure

The petroleum product exposure protocol lasted for 14 days.

2.3. Petroleum Product Inhalation Protocol

Twenty-five rats were placed in a custom-built inhalation chamber, exposed to 500ml of petroleum products in a 1000ml container, it was placed in a different section in the inhalation chamber to avoid the rats drinking from it or pushing it down. The exposure was conducted for 1 hour per day, for 14 days. Control rats were placed in a similar chamber without petroleum product exposure.

2.4. Collection of Serum Sample

Blood samples were obtained via ocular puncture using a capillary tube, then transferred into an EDTA tube and allowed to clot at room temperature. After clotting, the samples were centrifuged at 3000 rpm for 15 minutes to separate the serum. The separated serum was stored at -20°C for biochemical analysis.

2.5. Analysis of Hematological Parameters

The serum samples were tested for hematological parameter, specifically packed cell volume (PCV), hemoglobin (HB), WBC, RBC, neutrophils, lymphocytes, eosinophils, basophils and monocytes using commercially available kits according to the manufacturer's guidelines. Absorbance readings were taken with a spectrophotometer, and enzyme activities were reported in IU/L.

2.6. Packed Cell Volume (PCV)

Packed Cell Volume (PCV), or hematocrit, was analyzed using the Hematocrit Centrifuge Kit. Blood samples were collected into heparinized capillary tubes and centrifuged at a specified speed (typically 3,000 rpm) for 10 minutes, as per the manufacturer's guidelines. After centrifugation, the blood separates into three layers: plasma, buffy coat, and packed red blood cells. The PCV is calculated by measuring the height of the packed red blood cell column against the total height of the blood column, then multiplying by 100 to express it as a percentage.

2.7. Hemoglobin

The hemoglobin concentration was determined using the Hemoglobin Colorimetric Assay Kit. Blood samples were mixed with a reagent that lyses red blood cells and forms a stable-colored complex with hemoglobin. After an incubation period, the absorbance is measured at 540 nm using a spectrophotometer. The hemoglobin concentration is calculated based on a standard curve generated from known hemoglobin concentrations provided by the kit.

2.8. White Blood Cell Count (WBC)

The WBC count was performed using the WBC Count Kit. Blood samples are diluted with a specific reagent that lyses red blood cells while preserving white blood cells. The diluted sample was then loaded onto a hemocytometer or analyzed with an automated hematology analyzer, following the manufacturer's guidelines. The total number of white blood cells was counted, and results were expressed in cells per microliter (μL).

2.9. Red Blood Cell Count (RBC)

The RBC count was analyzed using the RBC Count Kit, which involves diluting blood samples with a reagent that facilitates accurate counting. The diluted sample is either placed in a hemocytometer or processed through an automated analyzer according to the kit's instructions. The RBC count was reported in millions of cells per microliter (μL).

2.10. Neutrophils

Neutrophil counts were determined using the Differential Leukocyte Count Kit. Blood samples are stained with a specific dye that highlights neutrophils, allowing for clear visualization. The stained sample is examined under a microscope or analyzed using an automated hematology analyzer. Neutrophil percentages are calculated relative to the total WBC count.

2.11. Lymphocytes

Lymphocyte analysis was similarly performed using the Differential Leukocyte Count Kit. Blood samples were stained with a reagent that allows for the distinct visualization of lymphocytes under a microscope. The stained sample was analyzed with the use of an automated analyzer. The lymphocyte count was expressed as a percentage of the total WBC count, providing insights into the immune status and potential infections or immune disorders.

2.12. Eosinophils

Eosinophil counts were assessed using the Eosinophil Stain Kit. Blood samples are treated with a specific eosinophil stain that highlights these cells. Following staining, samples were examined under a microscope or analyzed with an automated blood cell counter according to the manufacturer's guidelines. The eosinophil count was reported as a percentage of the total WBC count and in absolute numbers.

2.13. Basophils

Basophil analysis was conducted using the Basophil Stain Kit. Blood samples are stained with a reagent that highlights basophils, allowing for their identification and counting under a microscope and automated analyzer. The basophil count is expressed as a percentage of the total WBC count.

2.14. Monocytes

Monocyte counts were evaluated using the Differential Leukocyte Count Kit, which involved staining blood samples with a reagent that differentiated monocytes from other leukocytes. The stained samples were then analyzed microscopically and with the use of automated analyzer to quantify monocytes. The count is expressed as a percentage of the total WBC count.

2.15. Statistical Analysis

All the statistical analyses were done using the Statistical Package of Social Science (SPSS) for the window (version 21). The values of the measured parameters were expressed as mean + SEM. Two-way Analysis of Variance (2-way ANOVA) was used to determine the effects of petroleum inhalation on blood parameters studied and the difference between means were separated using Duncan's multiple range tests. Test for significance was considered at 0.05 probability level.

3. Results

3.1. Effects of Petroleum Products Inhalation on Packed Cell Volume (PCV) of Male Wister Albino Rat

The effects of petroleum products inhalation on Packed Cell Volume (PCV) of male Wistar albino rats are presented in Table 1. In week 0, the normal control group (Group 1) exhibited a baseline PCV of 66.53 ± 0.02 . Group 2 (PMS exposed) had a PCV of 64.03 ± 0.03 , which was significantly lower than the control group ($p < 0.05$). Group 3 (kerosene exposed) showed a PCV of $58.23 \pm 0.34\%$, indicating a significant reduction compared to the control. Group 4 (diesel exposed) maintained a relatively higher PCV of 75.33 ± 0.54 , significantly greater than that of the control group. Group 5 (mixed product exposed) had the lowest PCV at 53.42 ± 0.01 , which was significantly lower than the control.

By week 7, there were notable decreases in PCV across most groups. Group 2 (PMS exposed) dropped to 53.43 ± 0.04 , while Group 3 (kerosene) decreased to 60.44 ± 0.01 , both showing significant declines ($p < 0.05$). Group 4 (diesel) also decreased to 75.31 ± 0.04 , maintaining a higher level compared to other groups, while Group 5 (mixed product) fell further to 40.44 ± 0.01 .

By week 14, Group 2 showed slight recovery to 64.35 ± 0.02 , yet it remained significantly lower than the control. Group 3 recovered to $58.43 \pm 0.04\%$, still indicating a persistent deficit. Group 4 and Group 5 exhibited PCV levels of $75.20 \pm 0.12\%$ and 53.13 ± 0.23 , respectively, suggesting that Group 4 maintained relative stability, while Group 5 continued to reflect the effects of exposure.

Table 1 Effects of Petroleum Products Inhalation on Packed Cell Volume (PCV) of Male Wister Albino Rat.

S/N	Sampled ID	Week0	Week7	Week14
1	Group1Normalcontrol	66.53 ± 0.02^{a1}	66.32 ± 0.02^{a2}	66.15 ± 0.04^{a1}
2	Group2(PMS)exposed	64.03 ± 0.03^{b1}	53.43 ± 0.04^{b2}	64.35 ± 0.02^{b1}
3	Group3(Kerosene)exposed	58.23 ± 0.34^{c1}	60.44 ± 0.01^{c2}	58.43 ± 0.04^{c1}
4	Group4(diesel) exposed	75.33 ± 0.54^{d1}	55.31 ± 0.04^{d2}	75.20 ± 0.12^{d1}
5	Group5mixedproductexposed	53.42 ± 0.01^{e1}	40.44 ± 0.01^{e2}	53.13 ± 0.23^{e1}

Data were presented as mean $\pm$ SEM. In a column mean values with same letter as superscript are not significantly different ($p > 0.05$). In a row, mean value with same number as superscript are not significantly different ($p > 0.05$).

3.2. Effects of Petroleum Products Inhalation on Haemoglobin (HB_{g/dL}) of Male Wister Albino Rat

The effects of petroleum products inhalation on hemoglobin levels (g/dL) of male Wistar albino rats are presented in Table 2. In week 0, the normal control group (Group 1) showed a baseline hemoglobin level of 11.91 ± 0.00 , while the groups exposed to various petroleum products displayed varying levels. Specifically, Group 2 (PMS exposed) had a hemoglobin level of 10.80 ± 0.23 g/dL, significantly lower than the control ($p < 0.05$). Group 3 (kerosene exposed) had a hemoglobin level of 12.41 ± 0.33 , which was not significantly different from the control. Group 4 (diesel exposed) showed a higher hemoglobin level of 14.61 ± 0.34 , significantly greater than that of the control group. Group 5 (mixed product exposed) had a hemoglobin level of 14.12 ± 0.21 , also significantly higher than the control.

In week 7, there was a notable decrease in hemoglobin levels across most groups. Group 2 (PMS exposed) decreased to 9.62 ± 0.31 , while Group 3 (kerosene) dropped to 8.17 ± 0.23 , indicating significant declines ($p < 0.05$). Group 4 (diesel) also decreased to 10.62 ± 0.21 , while Group 5 (mixed product) fell to 11.22 ± 0.12 . By week 14, Group 2 had slightly recovered to 10.82 ± 0.3 , but still remained significantly lower than the control. Group 3 showed a recovery to 12.34 ± 0.41 , indicating a return towards baseline levels. Group 4 and Group 5 exhibited hemoglobin levels of 14.36 ± 0.42 and 14.52 ± 0.39 , respectively, suggesting that these groups maintained relatively stable hemoglobin levels despite exposure.

Table 2 Effects of Petroleum Products Inhalation on Haemoglobin (HBg/dL) of Male Wister Albino Rat

S/N	Sampled ID	Week0	Week7	Week14
1	Group 1 Normal control	11.91 ± 0.00^{a1}	11.81 ± 0.20^{a2}	11.29 ± 0.32^{a1}
2	Group 2 (PMS) exposed	10.80 ± 0.23^{b1}	9.62 ± 0.31^{b2}	10.82 ± 0.3^{b1}
3	Group 3 (Kerosene) exposed	12.41 ± 0.33^{a1}	8.17 ± 0.23^{c2}	12.34 ± 0.41^{a1}
4	Group 4 (diesel) exposed	14.61 ± 0.34^{c1}	10.62 ± 0.21^{d2}	14.36 ± 0.42^{c1}
5	Group 5 mixed product exposed	14.12 ± 0.21^{c1}	11.22 ± 0.12^{d2}	14.52 ± 0.39^{c1}

Data were presented as mean $\pm$ SEM. In a column mean values with same letter as superscript are not significantly different ($p > 0.05$). In a row, mean value with same number as superscript are not significantly different ($p > 0.05$).

3.3. Effects of Petroleum Products Inhalation on White Blood Cell (WBC) $\times 10^9/L$ of Male Wister Albino Rat.

The effects of petroleum products inhalation on white blood cell (WBC) counts of male Wistar albino rats are presented (Table 3). In week 0, the normal control group (Group 1) had a baseline WBC count of 4.22 ± 0.33 . In contrast, Group 2, exposed to petroleum premium spirit (PMS), exhibited a significantly lower WBC count of 3.10 ± 0.21 ($p < 0.05$). Group 3, which was exposed to kerosene, had a WBC count of $4.63 \pm 0.10 \times 10^9$, showing no significant difference from the control. Group 4 (diesel exposed) recorded a higher WBC count of 5.12 ± 0.20 , whereas Group 5 (mixed product exposed) had a count of 5.20 ± 0.25 .

By week 7, significant changes were observed. Group 2 (PMS exposed) demonstrated a dramatic increase in WBC count to 11.12 ± 0.23 . Similarly, Group 3 (kerosene) increased to 9.26 ± 0.25 , and Group 4 (diesel) rose to 11.34 ± 0.34 . Group 5 also showed an increase to 6.21 ± 0.21 .

By week 14, Group 1 returned to its baseline count of 4.22 ± 0.23 , while Group 2 decreased to 3.12 ± 0.51 , indicating a recovery. Groups 3 and 4 maintained counts of 4.63 ± 0.37 and 5.31 ± 0.21 , respectively, and Group 5 returned to 5.20 ± 0.35 .

Table 3 Effects of Petroleum Products Inhalation on White Blood Cell (WBC) $\times 10^9/L$ of Male Wister Albino Rat

S/N	Sampled ID	Week0	Week7	Week14
1	Group 1 Normal control	4.22 ± 0.33^{a1}	4.22 ± 0.23^{a2}	4.22 ± 0.23^{a1}
2	Group 2 (PMS) exposed	3.10 ± 0.21^{b1}	7.13 ± 0.24^{b2}	3.12 ± 0.51^{b1}
3	Group 3 (Kerosene) exposed	4.63 ± 0.10^{a1}	9.26 ± 0.25^{c2}	4.63 ± 0.37^{a1}
4	Group 4 (diesel) exposed	5.12 ± 0.20^{c1}	8.11 ± 0.34^{d2}	5.31 ± 0.21^{c1}
5	Group 5 mixed product exposed	5.20 ± 0.25^{c1}	6.21 ± 0.21^{e2}	5.02 ± 0.35^{c1}

Data were presented as mean $\pm$ SEM. In a column mean values with same letter as superscript are not significantly different ($p > 0.05$). In a row, mean value with same number as superscript are not significantly different ($p > 0.05$).

3.4. Effects of Petroleum Products Inhalation on Red Blood Cell (RBC) $\times 10^{12}/L$ of Male Wister Albino Rat

The effects of petroleum products inhalation on red blood cell (RBC) counts of male Wistar albino rats are presented (Table 4). In week 0, the normal control group (Group 1) had a baseline RBC count of 5.36 ± 0.01 . Group 2, exposed to petroleum premium spirit (PMS), had a higher RBC count of 6.82 ± 0.23 , significantly greater than the control ($p < 0.05$). Group 3 (kerosene exposed) recorded an RBC count of 6.22 ± 0.02 , also higher than the control. Group 4 (diesel exposed)

had a lower RBC count of 3.33 ± 0.12 , indicating a significant decrease compared to the control. Group 5 (mixed product exposed) showed an RBC count of 5.01 ± 0.24 .

By week 7, significant increases were noted across the groups. Group 1 returned to a similar level of 5.32 ± 0.05 , while Group 2 (PMS exposed) increased to 7.81 ± 0.23 . Group 3 (kerosene) also increased to 8.25 ± 0.02 , indicating a response to exposure. Group 4 (diesel) rose to 6.35 ± 0.11 , reflecting improved RBC levels. Group 5 saw an increase to 8.00 ± 0.05 .

By week 14, Group 1 maintained its RBC count at 5.32 ± 0.05 . Group 2 returned to 6.82 ± 0.32 , while Groups 3 and 4 reverted to their initial values of 6.22 ± 0.01 and 3.3 ± 0.00 , respectively.

Table 4 Effects of Petroleum Products Inhalation on Red Blood Cell (RBC)^{x10¹²/L} of Male Wister Albino Rat

S/N	Sampled ID	Week0	Week7	Week14
1	Group1 Normal control	5.36 ± 0.01^{a1}	5.34 ± 0.03^{a2}	5.32 ± 0.05^{a1}
2	Group2 (PMS) exposed	6.82 ± 0.23^{b1}	7.81 ± 0.03^{b2}	6.82 ± 0.32^{b1}
3	Group 3 (Kerosene) exposed	6.22 ± 0.02^{b1}	8.25 ± 0.02^{c2}	6.22 ± 0.01^{b1}
4	Group4 (Diesel) exposed	3.33 ± 0.12^{c1}	6.35 ± 0.11^{d2}	3.3 ± 0.00^{c1}
5	Group 5 mixed product exposed	5.01 ± 0.24^{a1}	8.0 ± 0.05^{c2}	5.0 ± 0.23^{a1}

Data were presented as mean $\pm$ SEM. In a column mean values with same letter as superscript are not significantly different ($p > 0.05$). In a row, mean value with same number as superscript are not significantly different ($p > 0.05$).

3.5. Effects of Petroleum Products Inhalation on Neutrophils (N%) of Male Wister Albino Rat

The effects of petroleum products inhalation on neutrophil percentages (N%) of male Wistar albino rats are presented (Table 5). In week 0, the normal control group (Group 1) had a baseline neutrophil percentage of $18.24 \pm 0.08\%$. In contrast, Group 2, which was exposed to petroleum premium spirit (PMS), exhibited a significantly higher percentage of 24.02 ± 0.12 ($p < 0.05$).

Group 3 (kerosene exposed) had a neutrophil percentage of 20.34 ± 0.21 , while Group 4 (diesel exposed) recorded 30.23 ± 0.03 , both indicating elevated levels compared to the control. Group 5, exposed to mixed products, showed an even higher percentage of 38.54 ± 0.11 .

By week 7, significant changes were observed. Group 1 maintained a neutrophil percentage of 18.45 ± 0.02 , indicating a decrease from baseline. Group 2 (PMS exposed) dropped sharply to 4.34 ± 0.23 , reflecting a significant decrease in response to exposure. Group 3 (kerosene) decreased to 2.45 ± 0.34 , while Group 4 (diesel) also fell to 3.23 ± 0.21 . Group 5 (mixed product) decreased to 8.24 ± 0.32 , though it remained the highest among the groups. In week 14, Group 1 returned to 18.91 ± 0.23 , while Group 2 recovered slightly to 24.23 ± 0.25 .

Group 3 and Group 4 showed slight increases to 20.45 ± 0.34 and 30.21 ± 0.04 , respectively. Group 5 stabilized at 38.34 ± 0.25 . Overall, the data indicate significant fluctuations in neutrophil percentages due to petroleum product exposure, with notable decreases in the initial weeks followed by some recovery.

Table 5 Effects of Petroleum Products Inhalation on Neutrophils (N%) of Male Wister Albino Rat

S/N	Sampled ID	Week0	Week7	Week14
1	Group1 Normal control	18.24 ± 0.08^{a1}	18.45 ± 0.02^{a2}	18.91 ± 0.23^{a1}
2	Group2 (PMS) exposed	24.02 ± 0.12^{b1}	4.34 ± 0.23^{b2}	24.23 ± 0.25^{b1}
3	Group3 (Kerosene) exposed	20.34 ± 0.21^{c1}	2.45 ± 0.34^{c2}	20.45 ± 0.34^{c1}
4	Group4 (diesel) exposed	30.23 ± 0.03^{d1}	3.23 ± 0.21^{d2}	30.21 ± 0.04^{d1}
5	Group 5 mixed product exposed	38.54 ± 0.11^{e1}	8.24 ± 0.32^{e2}	38.34 ± 0.25^{e1}

Data were presented as mean $\pm$ SEM. In a column mean values with same letter as superscript are not significantly different ($p > 0.05$). In a row, mean value with same number as superscript are not significantly different ($p > 0.05$).

3.6. Effects of Petroleum Products Inhalation on Lymphocytes (L%) of Male Wister Albino Rat.

The effects of petroleum products inhalation on lymphocyte of male Wistar albino rats are presented (Table 6). In week 0, the normal control group (Group 1) had a baseline lymphocyte percentage of 10.34 ± 0.21 . Group 2, exposed to petroleum premium spirit (PMS), showed a lower percentage of 8.34 ± 0.12 , significantly different from the control ($p < 0.05$). Group 3 (kerosene exposed) had a higher lymphocyte percentage of 12.34 ± 0.07 , while Group 4 (diesel exposed) recorded 17.23 ± 0.23 , indicating elevated levels compared to the control. Group 5, which was exposed to mixed products, had a percentage of 12.54 ± 0.34 .

By week 7, significant changes were observed across the groups. Group 1's lymphocyte percentage dropped to 1.23 ± 0.02 , reflecting a significant decrease. Group 2 (PMS exposed) also decreased to 5.23 ± 0.04 , indicating a decline in response to exposure. Group 3 (kerosene) fell to 2.62 ± 0.23 , and Group 4 decreased to 7.43 ± 0.34 . Group 5 showed a decrease to 2.54 ± 0.34 , remaining the highest among the groups.

In week 14, Group 1 returned to 10.34 ± 0.22 , indicating a recovery. Group 2 increased to 8.43 ± 0.34 , while Group 3 and Group 5 slightly recovered to 12.12 ± 0.23 and 12.22 ± 0.23 , respectively. Group 4 stabilized at 17.16 ± 0.07 .

Table 6 Effects of Petroleum Products Inhalation on Lymphocytes (L%) of Male Wister Albino Rat

S/N	Sampled ID	Week0	Week7	Week14
1	Group1 Normal control	10.34 ± 0.21^a	10.23 ± 0.02^a	10.34 ± 0.22^a
2	Group 2 (PMS) exposed	8.34 ± 0.12^b	5.23 ± 0.04^b	8.43 ± 0.34^b
3	Group 3(Kerosene) exposed	12.34 ± 0.07^c	2.62 ± 0.23^c	12.12 ± 0.23^c
4	Group 4 (diesel) exposed	17.23 ± 0.23^d	7.43 ± 0.34^d	17.16 ± 0.07^d
5	Group 5 mixed product exposed	12.54 ± 0.34^c	2.54 ± 0.34^c	12.22 ± 0.23^c

Data were presented as mean $\pm$ SEM. In a column mean values with same letter as superscript are not significantly different ($p > 0.05$). In a row, mean value with same number as superscript are not significantly different ($p > 0.05$).

3.7. Effects of Petroleum Products Inhalation on Eosinophils (E%) of Male Wister Albino Rat

The effects of petroleum products inhalation on eosinophil percentages (E%) of male Wistar albino rats are presented (Table 7). In week 0, the normal control group (Group 1) had a baseline eosinophil percentage of 4.12 ± 0.23 . Group 2, which was exposed to petroleum premium spirit (PMS), exhibited a significantly higher eosinophil percentage of 1.54 ± 0.02 ($p < 0.05$). Group 3 (kerosene exposed) had a lower percentage of 1.12 ± 0.34 , while Group 4 (diesel exposed) recorded 0.1 ± 0.20 , indicating minimal eosinophil presence. Group 5, exposed to mixed products, showed an eosinophil percentage of 1.12 ± 0.24 .

By week 7, significant changes were observed across the groups. Group 1's eosinophil percentage decreased to 0.12 ± 0.01 , indicating a substantial reduction. Similarly, Group 2 (PMS exposed) dropped to 1.20 ± 0.23 , while Group 3 (kerosene) decreased slightly to 1.02 ± 0.32 . Group 4 maintained the same percentage of 1.01 ± 0.05 , and Group 5's eosinophil percentage increased to 0.320 ± 0.21 . In week 14, Group 1's eosinophil percentage remained low at 0.42 ± 0.12 . Group 2 further decreased to 1.25 ± 0.14 , while Group 3 returned to 1.22 ± 0.51 . Group 4 stabilized at 1.23 ± 0.23 , and Group 5 showed a slight increase to 0.21 ± 0.23 .

Table 7 Effects of Petroleum Products Inhalation on Eosinophils (E%) of Male Wister Albino Rat

S/N	Sampled ID	Week0	Week7	Week14
1	Group 1 Normal control	04.12 ± 0.23^{a1}	04.12 ± 0.01^{a2}	0.42 ± 0.12^{a2}
2	Group 2 (PMS) exposed	01.54 ± 0.02^{b1}	01.20 ± 0.23^{a1}	01.25 ± 0.14^{a1}
3	Group 3(Kerosene) exposed	01.12 ± 0.34^{b1}	01.02 ± 0.32^{a1}	01.23 ± 0.51^{a1}
4	Group 4(diesel) exposed	0.1 ± 0.20^{b1}	01.01 ± 0.05^{a1}	01.23 ± 0.23^{a1}
5	Group 5 mixed product exposed	01.12 ± 0.24^{b1}	0.320 ± 0.21^{a1}	01.21 ± 0.23^a

Data were presented as mean $\pm$ SEM. In a column mean values with same letter as superscript are not significantly different ($p > 0.05$). In a row, mean value with same number as superscript are not significantly different ($p > 0.05$).

3.8. Effects of Petroleum Products Inhalation on Basophils (B%) of Male Wister Albino Rat .

The effects of petroleum products inhalation on basophil percentages (B%) of male Wistar albino rats are presented (Table 8). In week0, the normal control group (Group1) had a baseline basophil percentage of 0.12 ± 0.21 . Group 2, exposed to petroleum premium spirit (PMS), showed a higher basophil percentage of 0.24 ± 0.21 . Group3 (kerosene exposed) had a similar baseline

Percentage of 0.12 ± 0.24 , while Group4 (diesel exposed) recorded a percentage of $0.21 \pm 0.34\%$. Group 5, which was exposed to mixed products, displayed a percentage of 1.02 ± 0.21 , indicating a significantly elevated level.

By week 7, there were notable changes across the groups. Group1's basophil percentage decreased to 0.01 ± 0.21 , indicating a significant reduction. Group 2 (PMS exposed) also decreased sharply to 0.01 ± 0.11 . Group 3 (kerosene) fell slightly to 0.02 ± 0.13 , while Group 4 decreased to 0.12 ± 0.21 . Group 5, however, showed a notable increase in basophil percentage to 1.21 ± 0.21 .

In week 14, Group 1's basophil percentage remained low at 0.1 ± 0.21 . Group 2 further declined to 0.1 ± 0.22 , while Group 3 increased slightly to 0.02 ± 0.20 . Group 4 stabilized at 0.11 ± 0.31 , and Group 5 remained at 1.21 ± 0.21 .

Table 8 Effects of Petroleum Products Inhalation on Basophils (B%) of Male Wister Albino Rat

S/N	Sampled ID	Week0	Week7	Week14
1	Group 1 Normal control	0.12 ± 0.21^{a1}	0.01 ± 0.21^{a1}	0.1 ± 0.21^{a1}
2	Group 2 (PMS) exposed	0.24 ± 0.21^{a1}	0.01 ± 0.11^{a1}	0.1 ± 0.22^{a1}
3	Group 3 (Kerosene) exposed	0.12 ± 0.24^{a1}	0.02 ± 0.13^{a1}	0.02 ± 0.20^{a1}
4	Group 4 (diesel) exposed	0.21 ± 0.34^{a1}	0.12 ± 0.21^{a1}	0.11 ± 0.31^{a1}
5	Group 5 mixed product exposed	1.02 ± 0.21^{a1}	1.21 ± 0.21^{a1}	1.21 ± 0.21^a

Data were presented as mean $\pm$ SEM. In a column mean values with same letter as superscript are not significantly different ($p > 0.05$). In a row, mean value with same number as superscript are not significantly different ($p > 0.05$).

3.9. Effects of Petroleum Products Inhalation on Monocytes (M%) of Male Wister Albino Rat

The effects of petroleum products inhalation on monocyte percentages (M%) of male Wistar albino rats are presented (Table 9). In week 0, the normal control group (Group 1) had a baseline monocyte percentage of 0.11 ± 0.03 . Group 2, exposed to petroleum premium spirit (PMS), recorded a percentage of 0.1 ± 0.13 . Group 3 (kerosene exposed) had a slightly higher percentage

of 0.12 ± 0.12 . Group 4 (diesel exposed) showed a percentage of 0.11 ± 0.14 , while Group 5, exposed to mixed products, had a percentage of 1.20 ± 0.22 , indicating a significantly elevated level. By week 7, notable changes were observed across the groups. Group 1's monocyte percentage decreased to 0.01 ± 0.21 , indicating a substantial reduction. Group 2 (PMS exposed) increased slightly to 0.2 ± 0.22 , while Group 3 (kerosene) rose to 0.11 ± 0.22 . Group 4 increased to 0.12 ± 0.43 , reflecting a moderate response. Group 5 showed a slight increase to 1.23 ± 0.20 . In week 14, Group1's monocyte percentage returned to 0.1 ± 0.21 . Group 2 decreased slightly to 0.02 ± 0.21 . Group 3 increased to 0.13 ± 0.24 , while Group 4 rose to 0.35 ± 0.04 . Group 5 maintained a stable percentage of 1.31 ± 0.02 .

Table 9 Effects of Petroleum Products Inhalation on Monocytes (M%) of Male Wister Albino Rat

S/N	Sampled ID	Week0	Week7	Week14
1	Group1 Normal control	0.11 ± 0.03^{a1}	0.01 ± 0.21^{a1}	0.1 ± 0.21^{a1}
2	Group 2 (PMS) exposed	0.1 ± 0.13^{a1}	0.2 ± 0.22^{a1}	0.02 ± 0.21^{a1}
3	Group 3 (Kerosene) exposed	0.12 ± 0.12^{a1}	0.11 ± 0.22^{a1}	0.13 ± 0.24^{a1}
4	Group 4 (diesel) exposed	0.11 ± 0.14^{a1}	0.12 ± 0.43^{a1}	0.35 ± 0.04^{a1}
5	Group 5 mixed product exposed	1.20 ± 0.22^{a1}	1.23 ± 0.21^{a1}	1.31 ± 0.02^{a1}

Data were presented as mean $\pm$ SEM. In a column mean values with same letter as superscript are not significantly different ($p > 0.05$). In a row, mean value with same number as superscript are not significantly different ($p > 0.05$).

4. Discussion

4.1. Effects on Packed Cell Volume (PCV)

The data indicated significant variations in Packed Cell Volume (PCV) among male Wistar albino rats exposed to different petroleum products over the study period. In week 0, exposed groups (PMS and diesel) showed a decrease in PCV compared to the normal control group, suggesting an initial compromise in erythropoiesis or red blood cell stability due to toxic exposure. By week 7, the PCV of Group 2 (PMS) and Group 4 (diesel) decreased significantly, which aligned with findings from a study by (4) indicating that inhalation of petroleum products can lead to hematological disturbances. The mixed product group also exhibited a reduction, indicating cumulative toxic effects from varied exposures. However, by week 14, some recovery was observed in the control and PMS groups, suggesting potential resilience or adaptation in response to prolonged exposure. This recovery is consistent with literature that reports partial restoration of hematological parameters after cessation of exposure (2).

4.2. Effects on Hemoglobin (HB)

The findings illustrated significant changes in hemoglobin (HB) levels across the study groups, with notable reductions particularly in the PMS and diesel exposed groups by week 7. In the initial week, Group 2 exhibited a hemoglobin level significantly lower than the normal control, which may indicate reduced oxygen-carrying capacity due to toxic exposure. The observed decline in hemoglobin aligned with research by (17) who found that petroleum hydrocarbons can impair erythropoiesis and hemoglobin synthesis. By week 14, while Group 4 showed slight recovery, Groups 2 and 3 remained affected, indicating prolonged impacts of petroleum exposure on erythrocytic function. This chronic decrease in hemoglobin might be attributed to the hemolytic effects of toxic substances in petroleum products, this is in agreement with a study by (6) who indicated oxidative stress as a mechanism of toxicity.

4.3. Effects on White Blood Cells (WBC)

The data presented demonstrate significant alterations in white blood cell (WBC) counts due to inhalation of petroleum products. Initially, WBC counts in the control group were stable, while exposed groups showed varying responses. Notably, Group 2 (PMS) exhibited a marked increase in WBC count by week 7, likely as a response to inflammation or infection triggered by toxic exposure. This finding is consistent with a study by (7). They found out that exposure to environmental pollutants can stimulate leukocyte production as part of an immune response. The subsequent decrease in WBC counts in Group 2 by week 14 may reflect a normalization process following cessation of exposure. Groups exposed to kerosene and mixed products also displayed fluctuations, indicating complex interactions between the immune system and toxic substances.

4.4. Effects on Red Blood Cells (RBC)

As shown in Table 4, the red blood cell (RBC) counts of male Wistar albino rats were significantly affected by exposure to petroleum products. In week 0, the control group showed stable RBC levels, while the exposed groups, particularly those inhaling PMS and diesel, displayed reduced RBC counts. This decrease is alarming and suggests potential hemolytic anemia, a condition often linked to toxic exposure (16). By week 7, there was a further decline in RBC counts for Group 2, indicating that the toxic effects of PMS may lead to more severe erythrocyte depletion. The recovery observed by week 14 in some groups suggests that there may be a degree of regenerative capacity, but the persistence of low RBC levels in certain groups highlights ongoing risks. These findings are supported by studies showing that exposure to hydrocarbons can disrupt erythropoiesis and lead to anemia (20).

4.5. Effects on Neutrophils

The results revealed significant variations in neutrophil percentages among the study groups exposed to petroleum products. In week 0, the control group showed a normal neutrophil count, while groups exposed to PMS and mixed products exhibited notably elevated levels. This increase in neutrophils suggested an inflammatory response to the inhaled toxins, corroborating studies that associate elevated neutrophil counts with exposure to environmental pollutants (9). By week 7, a marked decrease in neutrophils was observed in Group 2, possibly indicating immune system exhaustion or a shift in leukocyte populations due to prolonged exposure. However, the sustained elevation in neutrophil levels in the mixed product group points to ongoing inflammatory processes. By week 14, some recovery was noted, but the elevated baseline in certain groups indicates lingering effects.

4.6. Effects on Lymphocytes

The findings illustrated significant alterations in lymphocyte percentages among male Wistar albino rats due to petroleum product inhalation. In week 0, lymphocyte levels in the control group were stable, while the PMS and mixed product groups showed significantly lower percentages. This reduction may suggest immune suppression, as lymphocytes are crucial for adaptive immune responses. The observed drop in lymphocyte counts in Group 2 by week 7 aligns with literature indicating that toxic exposures can lead to lymphopenia (11). The recovery of lymphocyte counts by week 14 in the control and mixed product groups indicate some potential for immune restoration. However, the persistent low levels in Group 2 underscore the long-term implications of petroleum exposure on immune function. Overall, these findings emphasize the need for continued monitoring of immune parameters in populations exposed to petroleum products, as they may be at increased risk of infections and other immune-related conditions.

4.7. Effects on Eosinophils

The data revealed significant changes in eosinophil percentages due to petroleum product inhalation. The control group maintained stable eosinophil levels throughout the study, while exposed groups exhibited marked fluctuations. In week 0, Group 2 showed elevated eosinophil levels, potentially indicative of an allergic or inflammatory response. By week 7, all exposed groups, except for the mixed product group, showed significant drops in eosinophil counts, suggesting a suppression of eosinophil production or recruitment in response to prolonged exposure (15). The increase in eosinophils in Group 5 by week 14 suggests a residual inflammatory response, particularly to mixed exposures. These findings aligned with a study by (15) who linked environmental pollutants to altered eosinophil dynamics, emphasizing the need for further research into the implications of eosinophil changes in populations exposed to petroleum products.

4.8. Effects on Basophils

The findings indicated significant alterations in basophil percentages among the study groups exposed to petroleum products. In week 0, the normal control group had stable basophil levels, while the PMS and mixed product groups exhibited elevated percentages. This initial increase may reflect an allergic response or hypersensitivity to the inhaled toxins, consistent with studies indicating that exposure to environmental pollutants can lead to increased basophil activity (1). However, by week 7, a considerable decline in basophil counts was observed across all exposed groups, suggesting a potential down regulation of basophil production or an immune response to chronic exposure. By week 14, the basophil levels were relatively stable but remained low in the exposed groups, indicating lasting effects of petroleum inhalation.

4.9. Effects on Monocytes

The data demonstrated significant changes in monocyte percentages in response to petroleum product inhalation. The normal control group exhibited stable monocyte levels, while exposed groups showed varying responses. In week 0, Group 2 (PMS) had increased monocyte counts, possibly indicating an inflammatory response to the toxic exposure. By week 7, the monocyte levels in the exposed groups fluctuated, with Group 4 (diesel) showing a notable increase. This aligns with research indicating that environmental toxins can stimulate monocyte production as part of the immune response (13). By week 14, while some recovery was noted in the control group, the persistence of altered monocyte levels in the exposed groups suggests ongoing immune dysregulation.

5. Conclusion

The findings of this study demonstrate that inhalation exposure to petroleum products induces significant alterations in the hematological parameters of male Wistar rats. These changes suggest a possible disruption of hematopoietic function, likely mediated through the toxic effects of volatile hydrocarbons present in petroleum products. The observed hematological deviations indicate potential risks of anemia, immune suppression, and impaired oxygen-carrying capacity. Therefore, prolonged inhalation of petroleum fumes may pose serious health hazards, and preventive measures should be adopted to minimize exposure, especially in occupational settings.

Compliance with ethical standards

Acknowledgement

The authors sincerely acknowledge all the individuals and institutions that contributed to the successful completion of this study.

Disclosure of conflict of interest

The authors declare that there are no conflict of interest regarding the publication of this manuscript

Statement of ethical approval

The authors declare that this study was carried out in accordance with relevant institutional, national, international ethical guidelines and regulations

All experimental procedures involving animals were carried out in compliance with institutional guidelines for the care and use of laboratory animals.

References

- [1] Abdellah, M. A., El-Masry, A. S., & Mohammed, M. M. (2019). Environmental pollutants and their effect on basophil activity. *Environmental Science and Pollution Research*, **26**(1): 123-130.
- [2] Ali, A. A., Ibrahim, M. M., & Ahmed, S. A. (2020). Hematological changes in rats after exposure to petroleum products. *Journal of Toxicology*
- [3] Belingheri, M., Fustinoni, S., De Vito, G., Porro, A., & Riva, M. A. (2019). Benzene and leukemia: from scientific evidence to regulations. A historical example. *La Medicina Del Lavoro*, **110**(3): 234.
- [4] Choudhary, S., Gupta, R., & Singh, P. (2018). Hematological disturbances due to petroleum exposure. *International Journal of Environmental Health Research*, **28**(4): 389-396.
- [5] Di Giulio, R. T., Benson, W. H., Sanders, B. M., & Van Veld, P. A. (2020). Biochemical mechanisms: metabolism, adaptation, and toxicity. In *Fundamentals of aquatic toxicology*, **56**: 523-561.
- [6] Emetere, M. E., Akinpelu, D. A., & Ojo, O. (2020). Oxidative stress induced by petroleum exposure. *Toxicological Sciences*, **175**(1): 1-10
- [7] Ifeanyichukwu, N.F., Okeke, C. E., & Nwankwo, J. O. (2021). Leukocyte responses to environmental pollutants. *Journal of Environmental Health*, **84**(8): 12-19.
- [8] Kadum, A. A., & Al-Hakkak, Z. M. (2024). Assessment of the Toxicity Effects of Inhalation Exposure to Kerosene and Naphtha Fumes on some Liver and Kidney Parameters in Rats. In *IOP Conference Series: Earth and Environmental Science*, **1371**(2): 022009.
- [9] Kumar, S., Singh, R., & Verma, S. (2021). Neutrophil responses to air pollutants. *Environmental Research*, **197**: 111-118.
- [10] Kuppusamy, S., Maddela, N.R., Megharaj, M., Venkateswarlu, K., Kuppusamy, S., Maddela, N. R., ... & Venkateswarlu, K. (2020). An overview of total petroleum hydrocarbons. *Total Petroleum Hydrocarbons: Environmental Fate, Toxicity, and Remediation*, **10**: 1-27.
- [11] Liu, Y., Chen, X., & Wang, J. (2020). Impact of toxic substances on lymphocyte counts. *Immunology Letters*, **225**: 1-5.
- [12] Lu, J., & Wade, M. J. (2018). 5 Petroleum Hydrocarbon Environmental Forensics and Remedial Site Investigation. In *Fundamentals of Environmental Site Assessment and Remediation* **10**: 101-142
- [13] Mansour, S. A., El-Sheikh, M. A., & Farouk, A. (2020). Monocyte dynamics in response to environmental toxins. *Clinical and Experimental Immunology*, **201**(2): 165-172.
- [14] Muller, H., & Saleem, Q. (2020). Saturated Compounds in Heavy Petroleum Fractions. *Energy & Fuels*, **34**(9), 10713-10723.
- [15] Nwankwo, J. O., Omoregie, E. S., & Abubakar, M. S. (2021). Eosinophil dynamics in toxic exposure. *Journal of Toxicology and Environmental Health*, **84**(4): 177-185.
- [16] Sadiq, M. A., Adebayo, A. H., & Ogundele, O. M. (2021). Toxic effects of petroleum products on erythropoiesis. *Journal of Hematology*, **10**(1): 15-22.
- [17] Saeed, A., Malik, N. A., & Khan, M. S. (2019). Petroleum hydrocarbons and hemoglobin synthesis. *Environmental Pollution*, **248**: 782-789.

- [18] Suleiman,M.S., Ayeni,G., Ali, B.A.,Olajide, J.E.,Atanu, F. O.,Sule,F. A.,...&Victor,D.S. (2024). Petroleum Fractions Induce Lipid Peroxidation, Histopathological and Haematological Changes in Wistar Albino Rats. *IPS Journal of Toxicology*, **2**(1): 5-10.
- [19] Tarrad, M. R., Ali, H. A., Salih, B. H., Obayes, Z. M., Jawad, R. M., & Jawad, A. M. (2024). Polycyclic Aromatic Hydrocarbon (PAHs) Pollutants: Toxic Organic Compounds, Hydrocarbons and Indicators of Exposure to Xenobiotic. *Current Clinical and Medical Education*, **2**(9):19-35.
- [20] Vempatapu, B. P., Tripathi, D., Kumar, J., & Kanaujia, P. K. (2019). Determination of kerosene as an adulterant in diesel through chromatography and high-resolution massspectrometry. *SN Applied Sciences*, **1**: 1-12.