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#### **ORIGINAL ARTICLE**

Experimental Biology and Medicine



# Effects Inhalation of Kerosene and Naphtha Fumes on Some Blood Indices in Rats

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## **Abstract**

**Background:** Multiple studies, including both humans and animals, have demonstrated that gasoline, kerosene, and diesel fuel exhaust emissions include chemical components harmful to the bone marrow, lymph nodes, and spleen. This study aimed to evaluate the impact of kerosene and naphtha vapors on several blood parameters in rats.

Methods and Results: The study was conducted on 10-12-week-old male Wistar albino rats (Rattus norvegicus) (n=20) weighing 150-200g. The experimental rats were categorized into two groups, each including five animals. The rats were exposed to kerosene and naphtha vapors for 15, 30, and 45 days, with six hours of daily exposure. Two control groups of animals, each including five animals, were exposed to room air. One group of rats was allowed to inhale the vapors emitted by the evaporating kerosene. Another group underwent an identical process for the naphtha vapors. Both groups of animals were exposed to daily vapors for six hours, from 9:00 a.m. to 3:00 p.m., six days a week, for three different durations: 15, 30, and 45 days. Blood samples were tested for hematological indices using a Cell Dyn Ruby Hematology Analyzer (Abbott, USA). On days 15, 30, and 45 of the experiment, rats exposed to kerosene and naphtha vapors had an increase in the total number of leukocytes, an increase in the percentage of lymphocytes, and a decrease in the percentage of neutrophils, compared to the control group (P<0.05 in all cases). At 15, 30, and 45 days of the experiment, the total number of RBCs increased significantly (P<0.05 in all cases). In addition, under the influence of kerosene and naphtha vapors, a higher level of PCV and MCV was noted at 30 and 45 days of the experiment, compared to the control group. At the same time, at the indicated stages of the experiment, there was a significant decrease in MCH and MCHC, compared to the control group (P<0.05 in all cases).

*Conclusion*: exposure to naphtha and kerosene vapor significantly affects a variety of WBC and RBC parameters, exhibiting toxic effects.(International Journal of Biomedicine. 2024;14(2):300-304.)

**Keywords**: white blood cell • red blood cell • kerosene • naphtha • toxic effects

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#### **Abbreviations**

Hb, hemoglobin; MCH, mean corpuscular hemoglobin; MCHC, MCH concentration; MCV, mean corpuscles volume; PCV, packed cell volume; RBC, red blood cell; WBC, white blood cell.

# Introduction

Refineries and petrochemical firms produce a wide range of toxic pollutants discharged into the environment. Petroleum fumes are widely present in our surroundings, and the main places for inhalation or contact are petrochemical businesses (refineries, oil fields, and filling stations) and residential areas. (1) The Al-Najaf refinery (Iraq) is a crucial source of petroleum products (naphtha and kerosene, gas oil, and heavy black petroleum) for Najaf city and its neighboring

regions. It serves the local demand for these products and supplies electrical stations, factories, and other purposes.

Crude petroleum consists of a mixture of different metals and hydrocarbons. Crude oil undergoes a refining process to produce distinct fractions such as petroleum, diesel, kerosene, heavy gas, and lubricating oils. Petrol, diesel, and kerosene are commonly produced from fractional distillation of crude oil. (2) Petrol poses a significant hazard due to its composition of diverse harmful substances, such as volatile aliphatic and aromatic hydrocarbons referred to as BTEX (benzene, toluene,

ethylbenzene, and xylene).<sup>(3)</sup> Furthermore, the widespread presence of processed petroleum chemicals has adversely affected many human biological functions.<sup>(4)</sup>

Multiple studies, including both humans and animals, have demonstrated that gasoline, kerosene, and diesel fuel exhaust emissions include chemical components such as cadmium, benzene, and volatile nitrates harmful to the bone marrow, lymph nodes, and spleen. (5) The presence of pollutants derived from petroleum products has been reported to cause changes in the levels of liver enzymes and in the generation of hormones in the pituitary gland. (6)

Industry emits hazardous air pollutants such as organic compounds (benzene, toluene, formaldehyde, acetaldehyde, phenol, ethylbenzene, xylene), inorganic compounds (hydrogen chloride [HCI], hydrogen cyanide [HCN]), reduced sulfur compounds (carbon disulfide [CS2]), and metals (arsenic, beryllium, cadmium, chromium, cobalt). These pollutants are primarily associated with the development of leukemia. (7,8) This study aimed to evaluate the impact of kerosene and naphtha vapors on several blood parameters in rats.

#### **Materials and Methods**

<u>Animals</u>

The experiments were performed in accordance with the norms for the humane treatment of animals, which are regulated by the International Guidelines of the Association for the Assessment and Accreditation of Laboratory Animal Care, following the protocol approved by the Institutional Animal Care and Use Committee of the University of Kufa. The study was conducted on 10-12-week-old male Wistar albino rats (Rattus norvegicus) (n=20) weighing 150–200g. The experimental rats were categorized into two groups, each including five animals. The rats were exposed to kerosene and naphtha vapors for 15, 30, and 45 days, with six hours of daily exposure. Two control groups of animals, each including five animals, were exposed to room air. The animals were kept at the Faculty of Science, University of Kufa's animal facility under controlled environmental conditions, with a temperature range of 25-28°C and a 12hour light-dark cycle. Animals were provided with normal water and feed.

#### Exposure to kerosene & naphtha vapors

The study used inhalation as the technique of exposure. The experimental groups were housed in animal cages and placed inside exposure chambers with dimensions of 150, 90, and 210 cm. Two extensively perforated 1000ml cans carrying 500 ml of fluid kerosene were put in the exposure chamber. One group of rats was allowed to inhale the vapors emitted by the evaporating kerosene. Another group underwent an identical process for the naphtha vapors. Both groups of animals were exposed to daily vapors for six hours, from 9:00 a.m. to 3:00 p.m., six days a week, for three different durations: 15, 30, and 45 days. The study used kerosene and naphtha produced by the Al-Najaf refinery. This investigation utilized a previously documented modified nose-inhalation exposure approach.<sup>(9)</sup>

#### Blood sample collection and analysis

After exposure, each animal received 0.5 ml ketamine and 0.1ml xylazine for moderate anesthesia. (10) After anesthesia, the rats were put in a dissecting dish and sutured at the wrists and ankles with small pins. Blood was drawn correctly using a 3ml and 5ml disposable syringe following a heart puncture. It was mixed correctly in an EDTA tube before being used by an automated analyzer to estimate blood levels. Blood samples taken from the EDTA tube were tested for hematological indices using a Cell Dyn Ruby Hematology Analyzer (Abbott, USA).(11)

Statistical analysis was performed using the statistical software package SPSS version 21.0 (SPSS Inc, Armonk, NY: IBM Corp). For the descriptive analysis, results are presented as mean (M)  $\pm$  standard deviation (SD). The Mann-Whitney U Test was used to compare the differences between the two independent groups (for nonparametric data). The Wilcoxon criterion was used to compare the differences between the paired samples. A value of P<0.05 was considered significant.

#### Results

On days 15, 30, and 45 of the experiment, rats exposed to kerosene and naphtha vapors had an increase in the total number of leukocytes, an increase in the percentage of lymphocytes, and a decrease in the percentage of neutrophils, compared to the control group (P<0.05 in all cases) (Tables 1 and 2).

Some red blood cell (RBC) parameters under the influence of kerosene and naphtha vapors changed at 15, 30 and 45 days of the experiment: The total number of RBCs increased significantly (P<0.05 in all cases). In addition, under the influence of kerosene and naphtha vapors, a higher level of PCV and MCV was noted at 30 and 45 days of the experiment, compared to the control group (P<0.05 in all cases). At the same time, at the indicated stages of the experiment, there was a significant decrease in MCH and MCHC, compared to the control group (Tables 3 and 4).

Table 1.

Effect of kerosene fumes on the total number of WBCs and the differential count in rats.

Variable	Control (1)	Time			
		15 (2)	30 (3)	45 (4)	Statistics
WBC count (×10³/mm³)	6.52±0.13	18.06±1.70	13.33±2.13	13.95±2.42	P <sub>1-2*</sub> P <sub>1-3*</sub> P <sub>1-4*</sub> P <sub>3-2*</sub> P <sub>4-2*</sub> P <sub>4-3^</sub>
Neutrophils (%)	51.40±2.55	19.12±3.32	13.34±2.09	10.42±3.13	P <sub>1-2*</sub> P <sub>1-3*</sub> P <sub>1-4*</sub> P <sub>3-2*</sub> P <sub>4-2*</sub> P <sub>4-3*</sub>
Lympho- cytes (%)	41.26±0.25	67.38±2.18	79.74±3.86	84.24±3.18	P <sub>1-2*</sub> P <sub>1-3*</sub> P <sub>1-4*</sub> P <sub>3-2*</sub> P <sub>4-2*</sub> P <sub>4-3*</sub>
Mid cells (%)	7.28±2.50	13.50±4.05	6.84±2.27	5.06±2.71	P <sub>1-2*</sub> P <sub>1-3^</sub> P <sub>1-4*</sub> P <sub>3-2*</sub> P <sub>4-2*</sub> P <sub>4-3^</sub>

<sup>\*-&</sup>lt;0.05; ^->0.05

Table 2.

Effect of naphtha fumes on the total number of WBCs and the differential count in rats.

Variable	Control (1)	Time			
		15 (2)	30 (3)	45 (4)	Statistics
WBC count (×10³/mm³)	6.25±0.38	16.07±2.38	13.34±2.54	14.43±0.82	$\begin{array}{c} P_{1\text{-}2^*} \; P_{1\text{-}3^*} \\ P_{1\text{-}4^*} \; P_{3\text{-}2^{\wedge}} \\ P_{4\text{-}2^{\wedge}} \; P_{4\text{-}3^{\wedge}} \end{array}$
Neutrophils (%)	51.40±2.55	13.24±3.35	13.24±3.34	13.80±4.61	$\begin{bmatrix} P & P \\ P_{1-2*}^{1-2*} & P_{3-2^{\wedge}}^{1-3*} \\ P_{4-2^{\wedge}}^{1-4*} & P_{4-3^{\wedge}}^{1-2} \end{bmatrix}$
Lympho- cytes (%)	41.26±2.37	74.28±3.65	82.16±3.72	80.30±3.07	P <sub>1-2*</sub> P <sub>1-3*</sub> P <sub>1-4*</sub> P <sub>4-2^</sub>
Mid cells (%)	7.34±2.76	12.48±2.90	7.22±2.66	6.10±2.49	P <sub>1-2*</sub> P <sub>1-3^</sub> P <sub>1-4*</sub> P <sub>3-2*</sub> P <sub>4-2*</sub> P <sub>4-3^</sub>

<sup>\*-&</sup>lt;0.05; ^->0.05

Table 3.

Effect of kerosene fumes on the total number of RBCs and RBC indices in rats.

Variable	Control (1)	Time			
		15 (2)	30 (3)	45 (4)	Statistics
RBCs (×10 <sup>6</sup> /mm <sup>3</sup> )	6.21±0.42	7.18±0.49	7.26±0.48	7.98±0.78	P <sub>1-2*</sub> P <sub>1-3*</sub> P <sub>1-3*</sub> P <sub>4-2^</sub> P <sub>4-3^</sub>
Hb (g/dl)	12.54±0.02	12.78±0.33	13.28±0.26	13.32±0.50	P <sub>1-2^</sub> P <sub>1-3^</sub> P <sub>1-3^</sub> P <sub>4-2^</sub> P <sub>4-2^</sub>
PCV (%)	38.42±1.16	40.16±1.60	46.44±2.80		
MCV (fL)	51.50±1.49	55.76±1.80	64.42±2.22	63.90±2.14	P <sub>1-2^</sub> P <sub>1-3*</sub> P <sub>1-3*</sub> P <sub>3-2*</sub> P <sub>4-2*</sub> P <sub>4-3^</sub>
MCH (pg)	19.44±2.71	19.14±2.18	18.02±2.86		
MCHC (g/dl)	31.88±0.82	30.94±0.51	28.10±1.16		

<sup>\*-&</sup>lt;0.05; ^->0.05

# **Discussion**

The results of our research are mainly consistent with the data of various authors. According to a study by Rabee, (12) exposure to air pollution at the oil refinery led to a considerable elevation in white blood cell (WBC) and lymphocyte counts.

Table 4.

Effect of naphtha fumes on the total number of RBCs and RBC indices in rats.

Variable	Control (1)	Time			
		15 (2)	30 (3)	45 (4)	Statistics
RBCs (×10 <sup>6</sup> /mm <sup>3</sup> )	6.21±0.42	7.47±0.46	7.64±0.10	7.44±0.36	P <sub>1-2*</sub> P <sub>1-3*</sub> P <sub>1-3*</sub> P <sub>4-2^</sub> P <sub>4-2^</sub>
Hb (g/dl)	12.54±0.28	12.64±0.37	12.26±0.29	12.70±0.51	$\begin{array}{c} P_{1\text{-}2^{\wedge}} \ P_{1\text{-}3^{\wedge}} \\ P_{1\text{-}4^{\wedge}} \ P_{3\text{-}2^{\wedge}} \\ P_{4\text{-}2^{\wedge}} \ P_{4\text{-}3^{\wedge}} \end{array}$
PCV (%)	38.42±0.14	39.84±1.77	44.18±2.86	49.56±1.83	P <sub>1-2^</sub> P <sub>1-3*</sub>
MCV (fL)	51.51±4.49	55.80±3.83	66.44±3.78	65.04±3.16	$\begin{array}{c} P_{1\text{-}2^{\wedge}} \ P_{1\text{-}3^{*}} \\ P_{1\text{-}4^{*}} \ P_{3\text{-}2^{*}} \\ P_{4\text{-}2^{*}} \ P_{4\text{-}3^{\wedge}} \end{array}$
MCH (pg)	19.44±0.71	17.02±0.65	16.50±0.46	16.80±0.48	P <sub>1-2*</sub> P <sub>1-3*</sub> P <sub>1-4*</sub> P <sub>3-2^</sub> P <sub>4-2^</sub> P <sub>4-3^</sub>
MCHC (g/dl)	31.88±0.82	30.86±0.56	27.52±0.82	25.88±0.53	$\begin{array}{c} P_{1\text{-}2^{\wedge}} \ P_{1\text{-}3^{*}} \\ P_{1\text{-}4^{*}} \ P_{3\text{-}2^{*}} \\ P_{4\text{-}2^{*}} \ P_{4\text{-}3^{*}} \end{array}$

<sup>\*-&</sup>lt;0.05; ^->0.05

As known, WBCs are responsible for safeguarding the body against infectious illnesses and external substances. Inhaling polluted air might stimulate the release of WBCs into the circulatory system, leading to inflammation. Furthermore, the elevated WBC counts may be attributed to activating a defensive mechanism in response to exposure to xenobiotics. A study by Uboh et al. (13) documented the hepatotoxic effects observed in albino Wistar rats following exposure to kerosene and gasoline vapors. Additionally, the hazardous constituents, particularly those present in petroleum vapors, can alter the chemistry of the blood.

A study by Johnson et al.<sup>(14)</sup> also revealed a statistically significant increase (P<0.05) in WBC count and lymphocyte number in the group exposed to household kerosene, compared to the control group. Leukocytosis was seen in the exposure group with a dosage of 1ml/kg body weight of household kerosene. This increase in WBCs may be attributed to bone marrow response and inflammatory illness in the animals exposed to kerosene.<sup>(15)</sup>

A study by Getu et al.<sup>(16)</sup> revealed that the mean RBC count, hemoglobin level, and the absolute lymphocyte count, of petrol-filling workers in Gondar town (Northwest Ethiopia), showed a significant increase, compared with the control group. Moreover, the duration of exposure to petrol showed a significant positive correlation with RBC count and mean cell hemoglobin concentration; however, a significant negative correlation was observed with mean cell volume.

A study by Sajid Jabbar and Ali (17) aimed to investigate the effect of benzene exposure on some blood parameters of

workers at several fuel stations in Basra city. The authors found significant hematological changes in the exposed workers and concluded that anemia was a common disorder among them. In addition, there was a significant decline in WBC and different types of WBC, including lymphocytes, monocytes, and neutrophils, due to continuous exposure to vapors of petrol products.

The results of a study by Imo et al.<sup>(18)</sup> showed that exposure of albino rats to inhalation of petroleum products could cause slight alteration in hematological parameters but can cause significant alteration in levels of liver function parameters and distortion in normal histoarchitecture of the liver tissue.

A study by Okoh et al. $^{(19)}$  showed a significant increase (P<0.05) in the PCV and the total number of leukocytes of Wistar rats exposed to diesel fumes, as compared to the control. Exposure to diesel fumes also caused elevated levels of liver and kidney biomarkers.

High kerosene and naphtha fume exposure may obstruct airways, causing alveolar hypoxia. In response to hypoxia, renal erythropoietin secretion increases, stimulating RBC production and maintenance. In this study, we also found a significant decrement in MCH and MCHC in rats exposed to kerosene and naphtha vapors for 30 and 45 days compared with the control. This may be caused by affected heme biosynthesis under kerosene and naphtha vapors that, with increased production of erythrocytes, lead to the development of hypochromic erythrocytosis. A study by Ufelle et al. (20) showed that exposure to volatile petroleum hydrocarbons raised the absolute RBC indices and liver enzymes and could stimulate a combined increase in the release of erythropoietin and interleukin-3, leading to ineffective hematopoiesis.

**In conclusion**, exposure to naphtha and kerosene vapor significantly affects a variety of WBC and RBC parameters, exhibiting toxic effects.

# **Disclosure and Competing Interests**

The authors declare that they have no competing interests. The views presented in this paper are the views of the authors and not an official position of the institution.

#### References

- 1. McDonald BC, de Gouw JA, Gilman JB, Jathar SH, Akherati A, Cappa CD, Jimenez JL, Lee-Taylor J, Hayes PL, McKeen SA, Cui YY, Kim SW, Gentner DR, Isaacman-VanWertz G, Goldstein AH, Harley RA, Frost GJ, Roberts JM, Ryerson TB, Trainer M. Volatile chemical products emerging as largest petrochemical source of urban organic emissions. Science. 2018 Feb 16;359(6377):760-764. doi: 10.1126/science.aaq0524. PMID: 29449485.
- 2. Berepubo NA, Johnson NC, Sese B.T. Growth potentials and organ weights of weaner rabbits exposed to crude oil contaminated feed. Int J Anim. Sci.1994; 9:73-76.
- 3. Kodidala SR, Ahanger AM, Gandhi A. Comparison of pulmonary functions in petrol pump workers and

- residents of oil refinery. Indian Journal of Medical Specialities.2020;11(4):197-200.
- 4. Ajeel MA, Ajeel AA, Nejres AM, Salih RA. Assessment of Heavy Metals and Related Impacts on Antioxidants and Physiological Parameters in Oil Refinery Workers in Iraq. J Health Pollut. 2021 Aug 17;11(31):210907. doi: 10.5696/2156-9614-11.31.210907. PMID: 34434599; PMCID: PMC8383791.
- 5. Menanteau-Ledouble S, El-Matbouli M. Chemical hazards associated with fish as a food. In: Chemical hazards in foods of animal origin. Wageningen Academic Publishers 2019:296-303.
- 6. Al-Hulfi RA, Al Salem BAM, Al-Naiema IM. The Effect of Air Pollutants on liver Enzymes and Pituitary Gland Hormones of Smokers and non-smokers of Oil Refinery and Gas Station Workers in Basra/Iraq. Indian Journal of Forensic Medicine & Toxicology. 2022;16(3):297-301.
- 7. Dantes E, Fildan AP, Toma CL, Voicu GH, Oancea C. Respiratory impact in workers exposed to air pollutants from petroleum refinery. J Environ Prot Ecol.2016; 17(2), 523-531 8. Vogelmeier CF, Criner GJ, Martinez FJ, Anzueto A, Barnes PJ, Bourbeau J, Celli BR, Chen R, Decramer M, Fabbri LM, Frith P, Halpin DM, López Varela MV, Nishimura M, Roche N, Rodriguez-Roisin R, Sin DD, Singh D, Stockley R, Vestbo J, Wedzicha JA, Agustí A. Global Strategy for the Diagnosis, Management, and Prevention of Chronic Obstructive Lung Disease 2017 Report. GOLD Executive Summary. Am J Respir Crit Care Med. 2017 Mar 1;195(5):557-582. doi: 10.1164/rccm.201701-0218PP. PMID: 28128970.
- 9. Uboh FE, Akpanabiatu MI, Ekaidem IS, Ebong PE, Umoh IB. Effect of inhalation exposure to gasoline fumes on sex hormones profile in Wistar albino rats. Acta Endocrinol. 2007;(Buc) 3(1): 23-30.
- 10. Gecit I, Kavak S, Meral I, Pirinçci N, Güneş M, Demir H, Cengiz N, Ceylan K. Effects of shock waves on oxidative stress, antioxidant enzyme and element levels in kidney of rats. Biol Trace Elem Res. 2011 Dec;144(1-3):1069-76. doi: 10.1007/s12011-011-9124-8. Epub 2011 Jun 24. PMID: 21701835.
- 11. Mathieleers I, Lsabel G, Fadila S, Rick G. Abbott apromise for life. Com, Case Book. 2012; 1-91
- 12. Rabee, A. M. Evaluation of Biomarkers in Workers Exposed to Air Pollutants in Oil Refineries. Indian Journal of Forensic Medicine & Toxicology. 2022;16(1).
- 13. Uboh FE. Evaluation of toxicological implications of inhalation exposure to kerosene fumes and petrol fumes in rats. Acta Biologica Szegediensis. 2005;49(3-4):19-22:
- 14. Johnson M, Damazio OA. Hepatotoxicity of House Hold Kerosene (HHK) on Liver Enzyme Markers and Its Effect on Hematological and Oxidative Stress Parameters on Wistar Albino Rats. Science Journal of Medicine and Clinical Trials. 2014: Article ID sjmct-232
- 15. Marieb EN. Human Anatomy and Physiology. 3rd ed. Benjamin and Cummnings Pub Co, California.1995. 585-611.

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- 16. Getu S, Shiferaw E, Melku M. Assessment of hematological parameters of petrol filling workers at petrol stations in Gondar town, Northwest Ethiopia: a comparative cross-sectional study. Environ Health Prev Med. 2020 Aug 29;25(1):44. doi: 10.1186/s12199-020-00886-1. PMID: 32861244; PMCID: PMC7456503.
- 17. Sajid Jabbar A, Ali ET. Impact of Petroleum Exposure on Some Hematological Indices, Interleukin-6, and Inflammatory Markers of Workers at Petroleum Stations in Basra City. J Environ Public Health. 2020 Aug 4;2020:7693891. doi: 10.1155/2020/7693891. PMID: 32831856;
- 18. Imo C, Uhegbu FO, Ifeanacho NG. Effect of exposure to inhalation of selected petroleum products on liver function of male albino rats: A comparative study. IOSR Journal of

- Environmental Science, Toxicology and Food Technology. 2015;19:2319-23993
- 19. Okoh HI, Odoh IM, Ahmed J, Adewemimo T, Nwana AO, Akano OA. Biochemical and Histopathological Effects of Exposure to Diesel Fumes on Vital Organs of Wistar Rats (*Rattus Norvegicus*). FUOYE Journal of Engineering and Technology.2023; 8(2),136-141.
- 20. Ufelle S, Onyekwelu K, Chinweoke A, Ibegbu D, Okoli U, Ikekpeazu J. Assessment of hepatic functions, hematopoietic cytokines and haematological parameters in people occupationally exposed to volatile petroleum hydrocarbons. Arch Environ Occup Health. 2021;76(8):567-571. doi: 10.1080/19338244.2020.1832037. Epub 2020 Oct 12. PMID: 33043855.