

Early Kidney and liver Dysfunction Associated with Exposure to Crude Oil in Al-Ahdab field

Nasser Nafaa Alqurashy¹, Zahraa Z. Al-Mamoori², Sura M.A. Alkadhimi³

¹Department of Pathological Analyses, College of Science, Wasit University, Kut, IRAQ

²Department of Biology, College of Science, University of Kerbala, IRAQ

³Department of Hotel Studies, College of Tourism Sciences, University of Karbala, IRAQ

*Corresponding Author: Nasser Nafaa Alqurashy

DOI: <https://doi.org/10.31185/wjps.264>

Received 13 October 2023; Accepted 2 November 2023; Available online 30 November 2023

ABSTRACT: Although the detrimental effects of crude oil on many types of plants and animals have been broadly studied, not enough research has been done on how exposure to oil affects human health. In the present work, we tried to inspect their effect on the liver and kidney functions of the Al-Ahdab field workers as well as the local population living close to the oil drilling sites (Fadak city). A randomly selected 150 subjects they don't suffer from chronic diseases (50 subjects from workers of Al-Ahdab field They have been working in the field for more than ten years and 50 subjects from Fadak city they don't work in the Al-Ahdab field and 50 subjects from Al- kut city far from the Al-Ahdab field also they don't work in the Al-Ahdab field as control) aged higher than 40 years were taken under consideration. The level of urea, creatinine, bilirubin, aspartate transaminase and alanine transaminase enzymes in serum were evaluated by spectrophotometer. Where it was found that the levels of urea, creatinine, liver enzymes, and bilirubin were higher among the Al-Ahdab field workers than the control. We also found that the residents of Fadak City, which is very close to the Al-Ahdab field oil extraction, were also high compared to the control but less than the Al-Ahdab field workers. The study reveals that long-term exposure to the pollutants produce from oil extraction may lead to harm effect to liver and kidney functions of workers in oil extraction field and populations living in polluted sites.

Keywords: Crude oil, human health, Al-Ahdab field, liver function, kidney function



1. INTRODUCTION

The economy in Iraq is heavily depending on revenue from oil [1]. Crude oil, apart from being an ever-increasing demand of global industry [2]. Oil also documented as one of the main origin of pollutants perceived in terrestrial, atmospheric and ocean ecosystem [3].

Exploration, extraction, refinement, and transportation are among the operational processes involved in oil exploitation. Crude oil and other naturally occurring hydrocarbons must be transported to the surface through the process of extraction; different methods such as mechanical pumping and hydraulic pumping could be done for this activity. These processes all severely damage the environment, causing contamination of the air, water, and soil [4].

Crude oil or its components can enter the human body through a variety of routes, such as skin sites are more liable to various types of diseases [5].

Up to date oil extraction often occurs close to human populations, Approximately 100 countries and 70,000 oil fields globally are estimated to contain more than 1600 billion barrels of known crude oil tanks [6].

Over 600 million people worldwide may be impacted by the health and environment of currently active oil fields [7]. Studying the impact of contamination linked to oil on health has mostly focused on workers and residents of the impacted coastal areas following oil spills [8].

Studies conducted by health workers in Melut and Koch reveal a positive association between these health problems and elevated pollution levels from the oil industry [9]. According to study in 2014, high percent of the women in the oil-producing regions gave birth to children who had birth defects, Additional study conducted in the US state of Colorado found a link between the prevalence of congenital heart defects and nearby to oil and gas fields [10]. There have been observations of hepatotoxicity and nephrotoxicity in response to petrol exposure in humans and animals [11].

In the body kidney and Liver play a vital role, the liver for instance carry out many functions involve ; detoxification, metabolism, digestion, and stores [12]. The kidney remove metabolic waste products from the body, maintain blood volume, and control the amount of minerals in the blood [13].

The aspartate and alanine transaminases (ALT and AST), alkaline phosphatase (ALP), gamma-glutamyl transferase (GGT), prothrombin time (PT), serum bilirubin, the international normalized ratio (INR), total protein, and albumin are mostly used as indicators of liver function, also these tests can assist in identifying the area of the liver that may be impaired [14].

Tests for creatinine concentration, blood urea nitrogen, and urine are also advised in order to assess kidney function [15].

Exposure to oil and oil products either directly or indirectly causes intense health trouble in humans, Direct skin contact and inhaling contaminated air (volatile fractions released as gases) are examples of direct exposures , while walking in contaminated areas Eating contaminated food and taking a bath in contaminated water can result in indirect oil exposure , and the effects vary greatly depending on the type of spill site (land, river, or ocean) [16]. The amount, way, and duration of exposure in addition to the age and implied medical condition of the exposed individual, all influence how serious a chemical compound poisoning is. [17]. The main objective of this work was to estimate the impact of crude oil exposure on liver (ALT, AST) and total bilirubin, as well as (urea and creatinine) renal function tests in Al hdab field workers as well as among the Fadak city population living close to the oil drilling site.

2. BIOLOGICAL SAMPLES AND ASSAYS

2.1 Plasma biochemical and enzymatic assays:

Blood samples collected into vacutainer Gel tubes were centrifuged at 3000 xg for 10 min. The supernatant was carefully aspirated into centrifuge tubes and then aliquot plasma were transferred into epindorff tubes and stored at -80°C till subsequent biochemical analyses. Creatinine, urea, total bilirubin, alanine aminotransferase and aspartate aminotransferase, were assayed in plasma.

2.2 Biochemical and enzymatic assays

2.2.1 Determination of urea and creatinine

For urea measurement 1ml of the working reagent was mixed with 10 µl of the sample or standard solution. The mixture was vortexed and incubated at 37°C for 3 minutes. Alkaline reagent (200 µl) was added then mixed and incubated at 37°C for 5 minutes. The absorbance of both the sample and standard were measured against reagent blank at 578 nm. while for Creatinine 100 µl of the specimen or standard solution was pipetted into a tube containing 1 ml of the reagent. The mixture was vortexed and the initial absorbance (A1) was measured after 30 seconds. Two minutes later, absorbance (A2) of the standard or specimen was recorded.

2.2.2 Determination of total bilirubin, aspartate aminotransferase and alanine aminotransferase

For bilirubin Reagents containing sulfanilic acid, sodium nitrite and caffeine were mixed with ul sample and incubated at 20-25°C for 10 minutes, then colouring reagent was added then mixed and incubated at (20-25°C). At 546 nm, the sample's absorbance was measured when compared to a blank. Diazotized sulfanilic acid Sulfanilic acid + NaNO₂ HCl Bilirubin + Diazotized sulifanilic acid pH 1.4.

For AST and ALT µl of the sample was added to 500 ml of the reagent and were vortexed and the initial 50 absorbance was read after 90 seconds. The timer was started simultaneously and the absorbance was recorded after 30, 60 and 90 seconds.

The mean absorbance change per. minute was used for the calculation of the enzyme activity

2.3 Statistical Analysis

Data was analyzed using IBM SPSS software package version 20.0. The Shapiro-Wilk test was used to verify the normality of distribution. Quantitative data were illustrated using range (minimum and maximum), mean, standard deviation, median and interquartile range (IQR). Results were considered significance at the 5% level.

3. RESULTS

3.1 Kidney functions biomarkers

Table (1) shows the results of urea and creatinine in the serum of people. The study showed that urea and creatinine increased significantly the Al-Ahdab field workers than the control group.

On the other hand, the residents of Fadak City, which is very close to the Al-Ahdab field oil extraction, were also high compared to the control, but less than the Al-Ahdab field workers (Figure 1, and 2).

Table 1. Comparison between the three studied groups according to renal function

	Al-Ahdab field (n = 50)	Fadak city (n = 50)	Al- kut city (control) (n = 50)	P
Urea				
Min. – Max.	40.0 – 66.0	30.0 – 48.0	22.70 – 44.60	<0.001*
Mean ± SD.	48.43 ± 6.04	40.26 ^a ± 4.40	33.17 ^{ab} ± 6.39	
Sig. bet. grps.	p ₁ <0.001*, p ₂ <0.001*, p ₃ <0.001*			
Creatinine				
Min. – Max.	1.22 – 1.82	1.11 – 1.29	0.72 – 1.33	<0.001*
Mean ± SD.	1.47 ± 0.16	1.20 ^a ± 0.05	0.99 ^{ab} ± 0.14	
Sig. bet. grps.	p ₁ <0.001*, p ₂ <0.001*, p ₃ <0.001*			

* p₁:comparing between Al-Ahdab field and Fadak city, p₂: comparing between Al-Ahdab field and Al-kut city (control), p₃: comparing between Fadak city and Al-kut city (control). Values are expressed as of Mean ± Standard deviation.

a: Significant with Al-Ahdab field

b: Significant with Fadak city

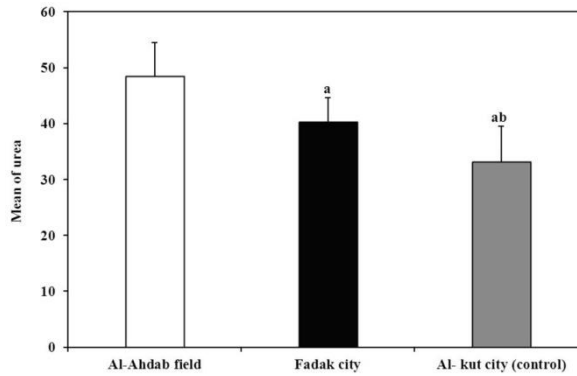


Figure 1. Statistical comparison between the three studied groups according to urea

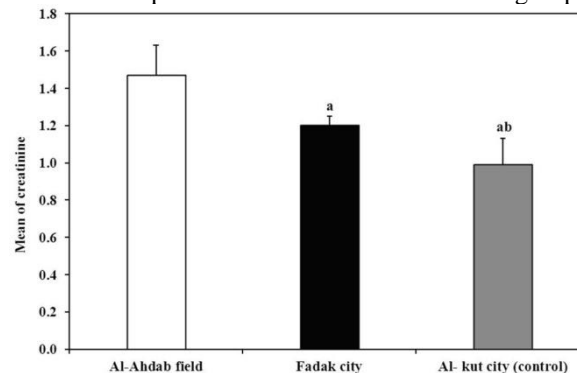


Figure 2. Statistical comparison between the three studied groups according to creatinine

3.2 Liver diagnostic biochemical markers

Table (2) shows the statistical comparison between different study groups according to liver enzymes. The results showed a significant increase in the activities of AST, ALT and bilirubin in serum of human as compared to the control group.

On the other hand, the residents of Fadak City, which is very close to the Al-Ahdab field oil extraction, were also high compared to the control, but less than the Al-Ahdab field workers (Figure 3,4, and 5).

Table 2. Comparison between the three studied groups according to liver enzymes

	Al-Ahdab field (n = 50)	Fadak city (n = 50)	Al- kut city (control) (n = 50)	p
AST				
Min. – Max.	123.0 – 230.0	122.0 – 169.0	91.0 – 152.0	<0.001*
Mean ± SD.	156.1 ± 24.76	142.7 ^a ± 13.54	118.6 ^{ab} ± 14.51	
Sig. bet. grps.	p ₁ <0.001*, p ₂ <0.001*, p ₃ <0.001*			
ALT				
Min. – Max.	42.0 – 77.0	32.0 – 49.0	24.0 – 45.0	<0.001*
Mean ± SD.	59.32 ± 8.47	41.30 ^a ± 4.04	34.30 ^{ab} ± 5.76	
Sig. bet. grps.	p ₁ <0.001*, p ₂ <0.001*, p ₃ <0.001*			
Bilirubin				
Min. – Max.	0.29 – 0.49	0.29 – 0.44	0.21 – 0.37	<0.001*
Mean ± SD.	0.43 ± 0.05	0.36 ^a ± 0.04	0.30 ^{ab} ± 0.04	
Sig. bet. grps.	p ₁ <0.001*, p ₂ <0.001*, p ₃ <0.001*			

* p₁: comparing between Al-Ahdab field and Fadak city, p₂: comparing between Al-Ahdab field and Al-kut city (control), p₃: comparing between Fadak city and Al-kut city (control). Values are expressed as of Mean ± Standard deviation.

a: Significant with Al-Ahdab field
b: Significant with Fadak city

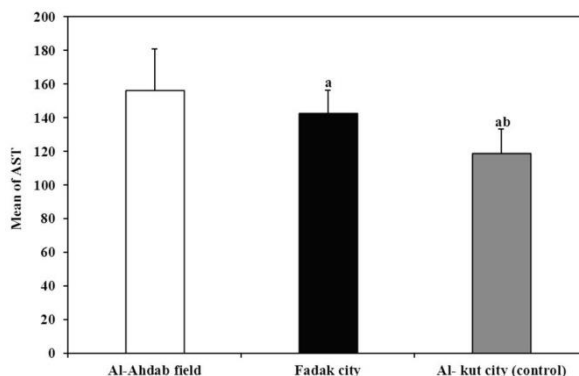


Figure 3. Statistical comparison between the three studied groups according to AST

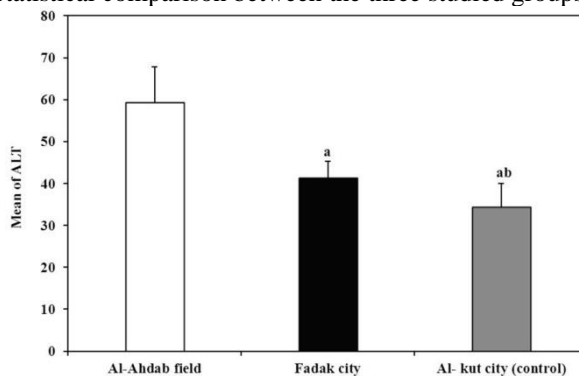


Figure 4. Statistical comparison between the three studied groups according to ALT

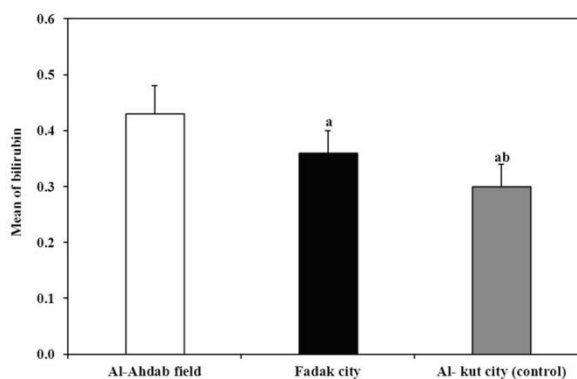


Figure 5. Statistical comparison between the three studied groups according to bilirubin

4. DISCUSSION

Large populations are possibly exposed to oil extraction associated contamination due to habitation in regions where there is oil extraction, particularly in LMICs between oil spills cleanup workers and oil industry workers [8, 18]. However, research on populations exposed in residential settings is remarkably insufficient. Research involving methodologically sound studies in populations exposed to exposure is needed on a global scale [19]. According to studies, communities close to operations like oil extraction activities are likely to have higher disease prevalence. As a result, exposure pathways like these could become crucial for public health [6, 20].

One of the main sites where xenobiotic toxicity is induced is thought to be the kidney [21]. The high blood flow rate is linked to kidney-specific toxicity because it causes the kidneys to receive higher than normal concentrations of xenobiotics. Particularly, the proximal tubule epithelium is more susceptible to nephrotoxicity because it expresses a variety of transporters that allow metabolites and toxic compounds to be actively intake and accumulated inside the cell. [22, 23].

The waste product creatinine is produced by the muscles, whereas urea is derived from the breakdown of proteins. [24]. Serum urea and creatinine are useful measurements to evaluate the state of renal function, with creatinine levels usually serving as a more precise indicator of kidney function than urea [25]. Elevated serum urea levels could indicate impaired renal discharge.

Due to the Al-Ahdab oil field's close proximity to residential areas, environmental pollution has resulted in elevated levels of urea and creatinine. The presence of heavy elements and environmental pollution from burning crude oil was observed in the samples of workers whose levels of urea and creatinine were measured. These workers reside in the areas adjacent to the Al-Ahdab field. [26].

The population's liver physiology may be significantly impacted by prolonged exposure to gaseous and particulate pollutants in surrounding areas of polluted sites. According to our research The Ahdab oil field, which is close to residential areas, has caused environmental pollution, which has raised ALT and AST levels., which is located near residential areas. This was evident in samples and individuals who were measured for ALT, and was evident in samples and individuals who were measured for ALT, and AST levels and who lived in the adjacent areas surrounding the Ahdab field. This pollution is caused by the heavy metals and smoke released from burning crude oil [17, 27].

ALP and aminotransferases (ALT, AST) are the most sensitive biomarkers that are directly embroiled in the bulk of hepatic harm and toxicity thus, Individuals in this study had significantly elevated levels of ALT and AST, which is consistent with Nwanjo and Ojiako [28] and Saadat and Ansari-Lari [29]. This increase may be explained by the cytoplasmic release of these enzymes into the bloodstream, which would indicate inflammation and necrosis. [30, 31]. The disruption of hepatocytes' transport function in liver injury leads to plasma membrane leakage and an increase in the level of liver enzymes in serum [32].

Michailova et al. (1998) [33], and Oladele (2003) [34] revealed a considerable rise in the activity of AST and ALT in workers in the oil production.

Ansari-Lari et al. (2004) [35] revealed that in a study involving 56 Shiraz pump station workers a considerable rise plasma levels of urea, creatinine, serum activity , ALT and AST, as well as considerable decreased in albumin and protein values in the exposed group as compared to a control group .

There is enough scientific confirmation to support the hypothesis that either acute or chronic exposure to crude oil (or its derivatives) causes a number of health effects in humans, including those who work in the oil industry and live in close proximity to oil fields, and significantly increased the liver and renal parameters with duration of exposure of gasoline station attendants. This indicates that there is a greater probability of changes in the way many essential organs function, and that these changes are linked to a higher risk of liver and renal cancer [17].

The findings of the present research show that exposure to crude oil can cause notable changes in the renal and hepatic functions of study participants, especially when compared to study participants in the control group, Alahdab field workers had significantly higher levels of ALT, AST, urea, creatinine, and bilirubin.

The significant rise in ALT and AST coincides with findings from studies carried out in Brazil, Egypt, Nigeria, India, and other countries [36-38].

We evaluated the effects of exposure to petroleum products and their derivatives on liver and renal parameters in field workers from Al-Ahdab and in the local population living close to oil drilling sites in Fadak City to properly understand the health risks connected to residential exposure to oil-related contamination, effective control policies to prevent such contamination, and public health recommendations and policies to prevent exposure in already contaminated places, All of these studies ought to be supported.

REFERENCES

- [1] E. A. Hassan, M. Rankin, and W. Lu, "The development of accounting regulation in Iraq and the IFRS adoption decision: an institutional perspective," *The International Journal of Accounting*, vol. 49, pp. 371-390, 2014.
- [2] H. Rajabi, M. H. Mosleh, P. Mandal, A. Lea-Langton, and M. Sedighi, "Emissions of volatile organic compounds from crude oil processing—Global emission inventory and environmental release," *Science of The Total Environment*, vol. 727, p. 138654, 2020.
- [3] C. Li, H. Wang, X. Liao, R. Xiao, K. Liu, J. Bai, *et al.*, "Heavy metal pollution in coastal wetlands: A systematic review of studies globally over the past three decades," *Journal of Hazardous Materials*, vol. 424, p. 127312, 2022.
- [4] A. B. Vakylabad, L. Zand, and Z. Moravvej, "Environmental challenges of extracting unconventional petroleum reserves," in *Crises in Oil, Gas and Petrochemical Industries*, ed: Elsevier, 2023, pp. 355-392.
- [5] S. A. Salami, F. O. Oreagba, H. M. Salahdeen, I. I. Olatunji-Bello, and B. A. Murtala, "Vitamin C supplementation modulates crude oil contaminated water induced gravid uterine impaired contractile mechanism and foetal outcomes in Wistar rats," *Journal of Complementary and Integrative Medicine*, 2023.
- [6] J. E. Johnston, E. Lim, and H. Roh, "Impact of upstream oil extraction and environmental public health: A review of the evidence," *Science of the Total Environment*, vol. 657, pp. 187-199, 2019.
- [7] L. M. Fusco, M. S. Schutter, and A. M. Cisneros-Montemayor, "Oil, Transitions, and the Blue Economy in Canada," *Sustainability*, vol. 14, p. 8132, 2022.
- [8] C. O'Callaghan-Gordo, M. Orta-Martínez, and M. Kogevinas, "Health effects of non-occupational exposure to oil extraction," *Environmental Health*, vol. 15, pp. 1-4, 2016.
- [9] S. Kuch and J. P. Bavumiragira, "Impacts of crude oil exploration and production on environment and its implications on human health: South Sudan Review," *International Journal of Scientific and Research Publications (IJSRP)*, vol. 9, p. 8836, 2019.
- [10] H. Faborode and G. Koledoye, "Perceived effects of oil exploration among among male and female fishers in Ilaje and Ese-Odo local government areas of Ondo State," *Nigerian Journal of Rural Sociology*, vol. 15, pp. 66-80, 2014.
- [11] M. Neghab, K. Hosseinzadeh, and J. Hassanzadeh, "Early liver and kidney dysfunction associated with occupational exposure to sub-threshold limit value levels of benzene, toluene, and xylenes in unleaded petrol," *Safety and health at work*, vol. 6, pp. 312-316, 2015.
- [12] V. Lala, M. Zubair, and D. Minter, "Liver function tests," *StatPearls*, 2023.
- [13] T. J. Hameed, M. M. Ibraheem, and M. A. Mezher, "Estimation of Renin enzyme activity and some biochemical parameters among chronic renal failure patients in Tikrit city," *Journal of Population Therapeutics and Clinical Pharmacology*, vol. 29, pp. 134-139, 2022.
- [14] D. H. Seo, Y. J. Suh, Y. Cho, S. H. Ahn, S. Seo, S. Hong, *et al.*, "Advanced liver fibrosis is associated with chronic kidney disease in patients with type 2 diabetes mellitus and nonalcoholic fatty liver disease," *Diabetes & Metabolism Journal*, vol. 46, pp. 630-639, 2022.
- [15] Divya, S. Mahapatra, and P. Chandra, "Design and Engineering of a Palm-Sized Optical Immunosensing Device for the Detection of a Kidney Dysfunction Biomarker," *Biosensors*, vol. 12, p. 1118, 2022.
- [16] S. Kuppusamy, N. R. Maddela, M. Megharaj, and K. Venkateswarlu, "Total petroleum hydrocarbons," *Environ. Fate Toxic. Remediat*, 2020.
- [17] T. Asefaw, M. Wolde, A. Edao, A. Tsegaye, G. Teklu, F. Tesfay, *et al.*, "Assessment of liver and renal function tests among gasoline exposed gas station workers in Mekelle city, Tigray region, Northern Ethiopia," *PLoS One*, vol. 15, p. e0239716, 2020.
- [18] E. S. Peters, A. L. Rung, M. H. Bronson, M. M. Brashear, L. C. Peres, S. Gaston, *et al.*, "The Women and Their Children's Health (WaTCH) study: methods and design of a prospective cohort study in Louisiana to examine the health effects from the BP oil spill," *BMJ open*, vol. 7, p. e014887, 2017.
- [19] K. Sam, N. Zabbey, I. F. Vincent-Akpu, G. Komi, P. O. Onyagbodur, and B. B. Babatunde, "Socio-economic Baseline for oil-impacted communities in Ogoniland: Towards a Restoration Framework in Niger Delta, Nigeria," 2023.

- [20] J. E. Johnston, T. Enebish, S. P. Eckel, S. Navarro, and B. Shamasunder, "Respiratory health, pulmonary function and local engagement in urban communities near oil development," *Environmental research*, vol. 197, p. 111088, 2021.
- [21] S. Hart and L. Kinter, "Assessing renal effects of toxicants in vivo," *Toxicology of the Kidney (JB Tarloff and LH Lash, Eds.)*, pp. 81-147, 2005.
- [22] A. Varshney, M. Rehan, N. Subbarao, G. Rabbani, and R. H. Khan, "Elimination of endogenous toxin, creatinine from blood plasma depends on albumin conformation: site specific uremic toxicity & impaired drug binding," *PLoS One*, vol. 6, p. e17230, 2011.
- [23] G. Vijaywargi, S. Kollipara, T. Ahmed, and S. Chachad, "Predicting transporter mediated drug–drug interactions via static and dynamic physiologically based pharmacokinetic modeling: A comprehensive insight on where we are now and the way forward," *Biopharmaceutics & Drug Disposition*, vol. 44, pp. 195-220, 2023.
- [24] S. N. Prabhu, S. C. Mukhopadhyay, and G. Liu, "Sensors and techniques for creatinine detection: a review," *IEEE Sensors Journal*, vol. 22, pp. 11427-11438, 2022.
- [25] S. Saggu, M. I. Sakeran, N. Zidan, E. Tousson, A. Mohan, and H. Rehman, "Ameliorating effect of chicory (*Chichorium intybus* L.) fruit extract against 4-tert-octylphenol induced liver injury and oxidative stress in male rats," *Food and chemical toxicology*, vol. 72, pp. 138-146, 2014.
- [26] T. Dey, K. Gogoi, B. Unni, M. Bharadwaz, M. Kalita, D. Ozah, *et al.*, "Role of environmental pollutants in liver physiology: special references to peoples living in the oil drilling sites of Assam," *PloS one*, vol. 10, p. e0123370, 2015.
- [27] M. Bin-Mefrij and S. Alwakeel, "The effect of fuel inhalation on the kidney and liver function and blood indices in gasoline station workers," *Advances in Natural and Applied Sciences*, vol. 11, pp. 45-50, 2017.
- [28] A. A. Ahmed, W. Alsalmi, A. A. Elhadi, and M. M. Almarabet, "Adverse effects of benzene exposure on hematological and hepatic biochemical parameters of petrol filling workers in Wadi Al-Hayah, Libya," *European journal of pharmaceutical and medical research*, vol. 9, pp. 40-45, 2022.
- [29] M. P. Singh and R. Himalian, "Monocyclic Aromatic Hydrocarbons (MAHs) Induced Toxicity in Drosophila: How Close How Far?," *Trends in Insect Molecular Biology and Biotechnology*, pp. 53-65, 2018.
- [30] M. L. Contreras-Zentella and R. Hernández-Muñoz, "Is liver enzyme release really associated with cell necrosis induced by oxidant stress?," *Oxidative medicine and cellular longevity*, vol. 2016, 2016.
- [31] N. Kuzu, K. Metin, A. F. Dagli, F. Akdemir, C. Orhan, M. Yalniz, *et al.*, "Protective role of genistein in acute liver damage induced by carbon tetrachloride," *Mediators of inflammation*, vol. 2007, 2007.
- [32] A. Mohammadyari, S. T. Razavipour, M. Mohammadbeigi, M. Negahdary, and M. Ajdary, "Explore in-vivo toxicity assessment of copper oxide nanoparticle in Wistar rats," *J Biol Today's world*, vol. 3, pp. 124-28, 2014.
- [33] P. Michailova, N. Petrova, G. Sella, L. Ramella, and S. Bovero, "Structural–functional rearrangements in chromosome G in *Chironomus riparius* (Diptera, Chironomidae) collected from a heavy metal-polluted area near Turin, Italy," *Environmental pollution*, vol. 103, pp. 127-134, 1998.
- [34] A. Akintonwa and T. Oladele, "Health effect of exposure to hydrocarbons on petrol filling station attendants in Lagos," *Nigerian Quarterly Journal of Hospital Medicine*, vol. 13, pp. 88-92, 2003.
- [35] M. Ansari-Lari, M. Saadat, and N. Hadi, "Influence of GSTT1 null genotype on the offspring sex ratio of gasoline filling station workers," *Journal of Epidemiology & Community Health*, vol. 58, pp. 393-394, 2004.
- [36] K. El-Said and A. El-Noueam, "Biological Monitoring of Fuel Stations Workers Occupationally Exposed to Petroleum Products," *Journal of High Institute of Public Health*, vol. 40, pp. 586-595, 2010.
- [37] A. Eltom and H. Hamd, "Assessment of liver Enzymes level among Sudanese Gasoline Station Workers," *Scholars Journal of Applied Medical Sciences*, vol. 5, pp. 738-743, 2017.
- [38] R. Gali, A. Daja, Y. Mamza, G. Ani, and G. Ani, "Liver enzymes and protein among petrol hawkers and petrol-pump attendants in a Nigerian population," *Advance laboratory medicine international*, vol. 2, pp. 123-129, 2012.