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EVALUATION OF SOME BIOCHEMICAL CHANGES IN WISTAR RATS ADMINISTERED DIFFERENT DOSES OF BENZENE

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ABSTRACT

Benzene is a volatile organic compound recognized for its carcinogenic effects in humans. This study investigated the effects of benzene exposure on lipid profile and selected biochemical parameters namely total protein, albumin, and total bilirubin in Wistar rats. A total of 40 adult male Wistar rats were randomly assigned into 10 groups, each comprising four rats. Group 1 received distilled water and served as the control, while groups 2 through 9 were administered increasing doses of analytical-grade benzene at 0.2, 0.4, 0.6, 0.8, 1.0, 1.2, 1.4, 1.6, and 1.8 ml/kg body weight, respectively, for a duration of 28 days. Unfortunately, animals in groups 6 to 10 did not survive the entire 28-day study period. Rats in groups 1 to 5 were euthanized on day 28 under chloroform anesthesia, and blood samples were collected via cardiac puncture for biochemical analysis. Benzene exposure resulted in a significant ($p < 0.05$) increase in serum levels of total cholesterol, triglycerides, low-density lipoprotein (LDL), and total bilirubin, while levels of high-density lipoprotein (HDL), total protein, and albumin were significantly decreased. These findings suggest that benzene adversely affects lipid metabolism and liver function, likely through mechanisms involving oxidative stress, inflammation, and hepatocellular injury.

KEYWORDS: Benzene, Total Bilirubin, High-Density Lipoprotein, Low-Density Lipoprotein, Total Cholesterol, Triglycerides, Albumin, Wistar rats, Lipid Profile.

INTRODUCTION

Benzene is a naturally occurring aromatic hydrocarbon present in crude oil and is a major contributor to the toxicity of gasoline, alongside other volatile organic compounds (VOCs) such as toluene, ethylbenzene, and xylene (Huff *et al*., 2021). It serves as a critical precursor in the synthesis of numerous industrial chemicals such as styrene, phenol, cyclohexane, and aniline (Bahadar *et al*., 2014). The compound's chemical stability, derived from its conjugated π-electron system, not only underpins its value as an industrial solvent but also contributes to the distinctive odor of petroleum products (Galbraith *et al*., 2010). Due to its well-established link with hematological cancers, particularly acute myeloid leukemia, benzene has been classified as a Group 1 carcinogen by the International Agency for Research on Cancer (IARC) (Snyder 2012; Cordiano *et al*., 2022).

As a pervasive environmental pollutant, benzene contaminates air, water, and soil, primarily through emissions from industrial processes, vehicle exhaust, and tobacco smoke (Horvat *et al*., 2025; Anigilaje et al., 2024). Human exposure occurs predominantly via inhalation, although dermal contact and ingestion of

contaminated food or water also contribute to overall exposure levels (Weisel, 2010). Occupational settings such as petrochemical plants, rubber manufacturing industries, and specialty chemical facilities present heightened exposure risks, particularly among workers who handle benzene directly (Bahadar *et al*., 2014; Saeedi *et al*., 2024). Chronic exposure to benzene has been linked to a range of hematotoxic outcomes, including aplastic anemia, bone marrow suppression, and increased risk of leukemia, all of which are well-documented in epidemiological studies (Snyder, 2012; Yusoff *et al*., 2023).

Following absorption into the body, benzene undergoes biotransformation primarily in the liver through the cytochrome P450 enzyme system, leading to the formation of reactive intermediates such as benzene oxide and phenol. These metabolites can bind to cellular macromolecules, disrupting normal cellular processes and causing tissue damage (Snyder *et al*., 2012). The principal metabolites include phenol (hydroxybenzene), catechol (1,2-dihydroxybenzene), and hydroquinone (1,4-dihydroxybenzene), which are implicated in benzene's toxic effects (IARC, 2018).

Beyond its well-known hematological toxicity, benzene has been shown to affect several organ systems, including the liver, kidneys, and cardiovascular system (Faulhammer *et al*., 2015). Hepatic dysfunction caused by benzene toxicity can be detected through alterations in specific biochemical markers (Tamber *et al*., 2023). Parameters of the lipid profile such as total cholesterol, triglycerides, high-density lipoprotein (HDL), and low-density lipoprotein (LDL) are valuable indicators of hepatic lipid metabolism and systemic lipid homeostasis (Stadler & Marsche, 2020). Deviations in these markers may suggest hepatic injury or metabolic disruptions following toxicant exposure. Likewise, serum levels of total protein and albumin reflect the liver's synthetic capacity, with reduced concentrations pointing to impaired hepatocyte function. Additionally, total bilirubin levels, which reflect hepatic excretory performance and hemoglobin catabolism, often rise in cases of liver or biliary tract dysfunction.

Although extensive research has addressed benzene's hematotoxic and carcinogenic effects (Saeedi *et al*., 2024), there remains a gap in understanding its impact on liver function, particularly concerning lipid metabolism and protein synthesis. A clearer picture of how benzene affects these biochemical markers is crucial to comprehending its broader toxicological profile and informing strategies for prevention and treatment.

Accordingly, this study investigated the effect of benzene exposure on lipid profile, total protein, albumin, and total bilirubin levels in Wistar rats. By assessing these key indicators, the study aims to elucidate the hepatic and metabolic consequences of benzene toxicity and contribute valuable data to the ongoing evaluation of its public health implications.

MATERIALS AND METHODS

Experimental Animals

Forty (40) Healthy male albino rats of Wistar strain weighing between 150-200g were purchased from the animal house of Department of Pharmacology, Faculty of Basic Clinical Sciences, College of Health Sciences, University of Port Harcourt, Rivers State. They were kept in standard rat cages in the animal house of Faculty of Basic Medical Sciences, College of Health Sciences, Niger Delta University, Wilberforce Island, Bayelsa State. The animals were allowed to acclimatize for 14 days under standard laboratory conditions with free access to commercial grower's mash (Delta Feeds), water ad libitum, 12 h/ 12 h light/darkness cycle and fresh air prior to the inception of this study.

Experimental Design

The animals were randomly grouped into ten (10) groups of four rats each in a standard plastic rat cage and treated as follows using an oral gavage tube:
Group I: Normal control: distilled water for 28 days.
Group II: Benzene (0.04ml/kg body weight) for 28 days.
Group III: Benzene (0.06ml/kg body weight) for 28 days.
Group IV: Benzene (0.08ml/kg body weight) for 28 days.
Group V: Benzene (0.2ml/kg body weight) for 28 days.
Group VI: Benzene (0.4ml/kg body weight) for 28 days.
Group VII: Benzene (0.6ml/kg body weight) for 28 days.
Group VIII: Benzene (0.8ml/kg body weight) for 28 days.
Group IX: Benzene (1.0ml/kg body weight) for 28 days.
Group X: Benzene (1.2ml/kg body weight) for 28 days.
At the end of 28th day, all animals in all the groups were anaesthetized with chloroform and sacrificed.

Collection of Samples

Blood was collected via cardiac puncture into plain bottles and allowed to stand for 30 minutes for coagulation to take place. Afterwards, the blood samples were centrifuged at 2000 RPM for ten minutes and the supernatant (serum) was collected for biochemical analysis.

DETERMINATION OF BIOCHEMICAL PARAMETERS

Serum total protein, albumin, total bilirubin, total cholesterol, triglycerides, high-density lipoprotein (HDL), and low-density lipoprotein (LDL) were measured using spectrophotometry applying the guide contained in the kit manual of Randox Laboratories (Crumlin, Co, Antrim, United Kingdom).

STATISTICAL ANALYSIS

All the results were expressed as Mean ± Standard deviation. The statistical significance was evaluated using the One-way Analysis of Variance (ANOVA) (SPSS 10.0). Statistical significance was set at $p<0.05$.

ETHICAL APPROVAL

Approval for this study was obtained from the Research and Ethics Committee of the College of Health Sciences, Niger Delta University, Wilberforce Island, Bayelsa State, Nigeria.

RESULTS

The results from this study are expressed in the bar charts below.

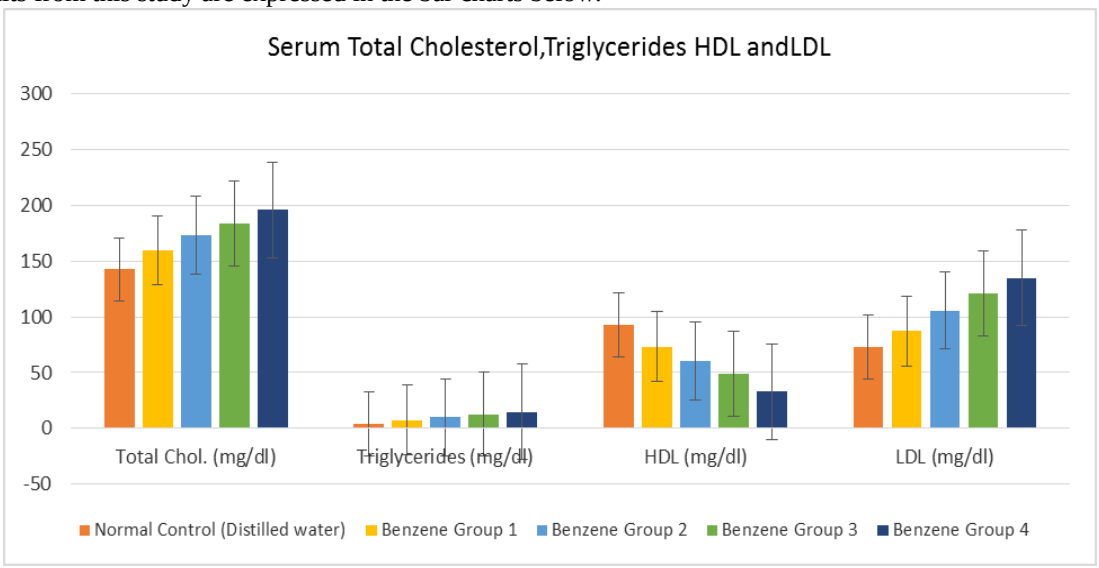

Figure 1: The effects of benzene on serum concentrations of total cholesterol, triglycerides, high-density lipoprotein (HDL), and low-density lipoprotein (LDL) in benzene-treated Wistar rats.

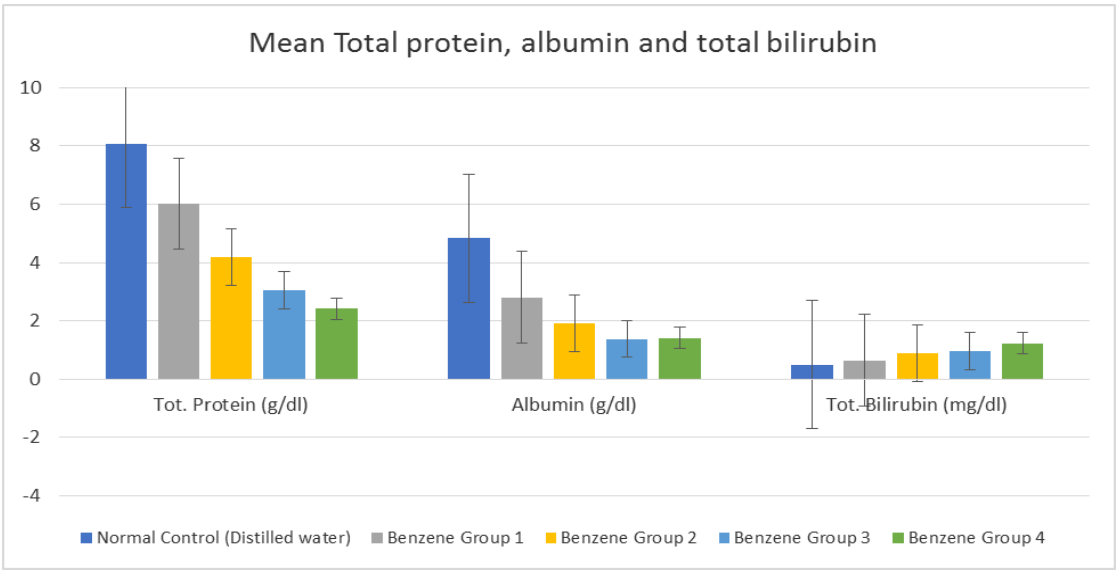

Figure 2: The effects of benzene on total protein, albumin and total bilirubin in benzene treated Wistar rats.

DISCUSSION

Volatile organic compounds (VOCs) are a class of chemicals commonly released through industrial processes, vehicle emissions, and household products, making them ubiquitous environmental contaminants present in air, water, and soil (EPA, 2022). Benzene is a VOC that the International Agency for Research on Cancer (IARC) has classified as a Group 1 carcinogen, a conclusive evidence of its cancer-causing potential in humans (IARC, 2012). This is mainly because benzene exposure has been clearly linked to a type of blood cancer called myeloid leukemia (AML), with numerous studies demonstrating increased risks among occupationally exposed populations (Spatari *et al*., 2021; Wang *et al*., 2024).

Recent studies have demonstrated a consistent relationship between benzene exposure and various hematological malignancies, showing consistent dose-response patterns across different exposure scenarios. The 2018 IARC evaluation confirmed benzene's Group 1 status based not only on human epidemiological data but also on compelling evidence from animal studies showing tumor development in multiple organ systems (Chiavarini *et al*., 2021). The carcinogenic effects of benzene occur through metabolic activation by liver

enzymes, particularly CYP2E1, which converts benzene into reactive metabolites such as hydroquinone and benzoquinone (Carbonari *et al*., 2016). These compounds generate oxidative stress by producing reactive oxygen species (ROS), leading to DNA damage and chromosomal abnormalities that can initiate cancer development (D'Souza *et al*., 2024).

The findings of this study demonstrate that benzene exposure induces significant biochemical alterations in Wistar rats, particularly affecting lipid metabolism and liver function, as evidenced by changes in serum lipid profile, total protein, albumin, and total bilirubin concentrations.

As illustrated in Figure 1, benzene administration led to a dose-dependent significant ($p<0.05$) increase in serum total cholesterol, triglycerides, and low-density lipoprotein (LDL) levels, accompanied by a marked reduction in high-density lipoprotein (HDL) concentrations. These alterations in lipid profile parameters suggest that benzene disrupts normal lipid metabolism. Elevated total cholesterol and triglycerides are indicators of impaired lipid clearance or increased lipid synthesis, both of which may arise from benzene-induced hepatocellular damage. The increase in LDL and reduction in HDL further point to heightened cardiovascular risk, as these changes are characteristic of dyslipidemia, a known precursor to atherosclerosis (Vekic *et al*., 2023). The observed dyslipidemia may be attributed to oxidative stress and inflammatory responses triggered by reactive metabolites of benzene, such as benzene oxide and hydroquinone, which can impair hepatic lipid regulatory enzymes and lipid transport mechanisms (Snyder, 2012).

Results (Figure 2) show that benzene exposure significantly reduced serum levels of total protein and albumin. These two parameters are critical indicators of hepatic synthetic function, and their reduction suggests impaired protein synthesis, likely due to hepatocellular injury. Albumin, being the most abundant plasma protein, produced exclusively by the liver, plays essential roles in maintaining oncotic pressure and transporting endogenous and exogenous compounds. A decline in its concentration often reflects hepatic dysfunction or chronic inflammation (Belinskaia *et al*., 2024). The reduction in total protein may also indicate a generalized impairment in the liver's capacity to produce other plasma proteins, possibly due to structural and functional damage to hepatocytes by benzene metabolites.

Total bilirubin levels were significantly elevated in benzene-treated rats, as also presented in Figure 2. Bilirubin is a breakdown product of hemoglobin and is normally conjugated and excreted by the liver. Elevated serum bilirubin levels may result from increased hemolysis, impaired hepatic uptake, conjugation defects, or biliary obstruction. The increase in total bilirubin likely reflects hepatic excretory dysfunction due to benzene-induced hepatotoxicity. The elevation may also be exacerbated by oxidative damage to red blood cells or impaired clearance of bilirubin conjugates, both of which are associated with systemic toxicity from benzene exposure (Hansen *et al*., 2020).

The death of animals in the higher dose groups (1.2–1.8 ml/kg) further supports the systemic toxicity of benzene. This acute mortality suggests a threshold beyond which benzene's hepatotoxic and systemic effects become rapidly lethal, possibly due to multi-organ failure, severe oxidative stress, or metabolic acidosis.

The alterations in lipid profile, total protein, albumin, and total bilirubin reflect substantial hepatic and systemic derangements following benzene exposure. These findings underscore the hepatotoxic potential of benzene and suggest that biochemical markers of liver function and lipid metabolism can serve as early indicators of benzene toxicity. While benzene's hematotoxic and carcinogenic effects have been well documented (Cordiano *et al*., 2022; Saeedi *et al*., 2024), this study expands the understanding of its impact on liver function and metabolism, which may have broader implications for occupational and environmental health monitoring.

CONCLUSION

The study reveals that benzene exposure adversely affects lipid homeostasis and hepatic function in Wistar rats. The dose-dependent elevations in serum total cholesterol, triglycerides, LDL, and total bilirubin, along with reductions in HDL, total protein, and albumin, highlight the systemic and liver-specific toxicity of benzene. These findings emphasize the need for stringent regulatory controls and protective measures in benzene-related industries, as well as further investigations into therapeutic interventions that may mitigate its toxic effects.

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REFERENCES

1. Anigilaje, E. A., Nasir, Z. A., & Walton, C. (2024). Exposure to benzene, toluene, ethylbenzene, and xylene (BTEX) at Nigeria's petrol stations: a review of current status, challenges and future directions. *Frontiers in Public Health, 12:* 1295758.
2. Bahadar, H., Mostafalou, S., & Abdollahi, M. (2014). Current understandings and perspectives on non-cancer health effects of benzene: A global concern. Toxicology and Applied Pharmacology, 276(2): 83-94.
3. Belinskaia, D. A., Jenkins, R. O., & Goncharov, N. V. (2024). Albumin Is an Integrative Protein of

Blood Plasma and Beyond. *International Journal of Molecular Sciences*, *25*(23): 12627.
4. Carbonari, D., Chiarella, P., Mansi, A., Pigini, D., Iavicoli, S., & Tranfo, G. (2016). Biomarkers of susceptibility following benzene exposure: influence of genetic polymorphisms on benzene metabolism and health effects. *Biomarkers in medicine*, *10*(2): 145-163.
5. Chiavarini, M., Rosignoli, P., Sorbara, B., Giacchetta, I., & Fabiani, R. (2024). Benzene Exposure and Lung Cancer Risk: A Systematic Review and Meta-Analysis of Human Studies. *International Journal of Environmental Research and Public Health*, *21*(2): 205.
6. Cordiano, R., Papa, V., Cicero, N., Spatari, G., Allegra, A., & Gangemi, S. (2022). Effects of benzene: hematological and hypersensitivity manifestations in resident living in oil refinery areas. *Toxics*, *10*(11): 678.
7. D'Souza, L. C., Paithankar, J. G., Stopper, H., Pandey, A., & Sharma, A. (2024). Environmental chemical-induced reactive oxygen species generation and immunotoxicity: a comprehensive review. *Antioxidants & Redox Signaling*, *40*(10-12): 691-714.
8. Faulhammer, F., van Ravenzwaay, B., Schnatter, A. R., Rooseboom, M., Kamp, H., Flick, B., ... & Kocabas, N. A. (2024). The short-term toxicity and metabolome of Benzene. *Toxicology Letters*, *400:* 58-70.
9. Horvat, T., Pehnec, G., & Jakovljević, I. (2025). Volatile Organic Compounds in Indoor Air: Sampling, Determination, Sources, Health Risk, and Regulatory Insights. *Toxics*, *13*(5): 344-344.
10. Huff, J., Infante, P. F., & Bird, M. G. (2021). Benzene-induced cancers: Abridged history and occupational health impact. International Journal of Environmental Research and Public Health, 18(21): 10885.
11. IARC, I. (2018). Working Group on the Evaluation of Carcinogenic Risks to Humans. DDT, Lindane, and 2, 4-D.
12. International Agency for Research on Cancer. (2012). A review of human carcinogens. F. Chemical Agents and Related Occupations: IARC Monographs on the Evaluation of Carcinogenic Risks to Humans.
13. Saeedi, M., Malekmohammadi, B., & Tajalli, S. (2024). Interaction of benzene, toluene, ethylbenzene, and xylene with human's body: Insights into characteristics, sources and health risks. *Journal of Hazardous Materials Advances*, *16:* 100459.
14. Snyder, R. (2012). Leukemia and benzene. International Journal of Environmental Research and Public Health, 9(8): 2875-2893.
15. Spatari, G., Allegra, A., Carrieri, M., Pioggia, G., & Gangemi, S. (2021). Epigenetic effects of benzene in hematologic neoplasms: the altered gene expression. *Cancers*, *13*(10): 2392.
16. Stadler, J. T., & Marsche, G. (2020). Obesity-related changes in high-density lipoprotein metabolism and function. *International journal of molecular sciences*, *21*(23): 8985.
17. Tamber, S. S., Bansal, P., Sharma, S., Singh, R. B., & Sharma, R. (2023). Biomarkers of liver diseases. *Molecular Biology Reports*, *50*(9): 7815-7823.
18. United States Environmental Protection Agency (USEPA). (2021). *Safer Choice*. https://www.epa.gov/saferchoice
19. Vekic, J., Stromsnes, K., Mazzalai, S., Zeljkovic, A., Rizzo, M., & Gambini, J. (2023). Oxidative stress, atherogenic dyslipidemia, and cardiovascular risk. *Biomedicines*, *11*(11): 2897.
20. Wang, T., Cao, Y., Xia, Z., Christiani, D. C., & Au, W. W. (2024). Review on novel toxicological effects and personalized health hazard in workers exposed to low doses of benzene. *Archives of Toxicology*, *98*(2): 365-374.
21. Weisel, C. P. (2010). Benzene exposure: An overview of monitoring methods and their findings. Chemico-Biological Interactions, 184(1-2): 58-66.
22. Yusoff, N. A., Abd Hamid, Z., Budin, S. B., & Taib, I. S. (2023). Linking benzene, in utero carcinogenicity and fetal hematopoietic stem cell niches: a mechanistic review. *International Journal of Molecular Sciences*, *24*(7): 6335.