Short Communication

Blood Lead Level and Biochemical Changes Among Gasoline Stations Workers Exposed to Benzene in Makkah City, Saudi Arabia

Mutasim M. Khalafalla^{1*}, Ahmad Babalghith², Khalil Mohamed³, Mohand Jafar³, Obaidullah H. Allehiany⁴, Hosham Karar⁵, Mohammed Bakri³, Al-Fadil Salih³

> Received: 27 July 2022 Accepted: 24 August 2022

Abstract

Benzene exposure in humans has significant negative health consequences and may be linked to an increased risk of blood biochemical changes. Therefore, the goal of this study was to determine the level of lead in blood of fuel station workers at various petroleum stations in holly Makkah city and look into its effects on specific blood biochemical parameters. Blood samples were collected from 63 individuals, including 44 samples from gasoline station workers (exposed group) and 19 samples from women who were not subjected to any kind of benzene exposure (nonexposed group). Socio-demographic data were collected using a structured questionnaire and 10 ml of venous blood was collected for the determination of lead and biochemical parameters. Data obtained were analyzed using Statistical Package for the Social Sciences (SPSS). The median (range) of blood lead level among exposed group 40.5 (4.0-90.0) $5\mu g/dL$ was significantly higher (P = 0.00) than non-exposed group 3.2 (2.7-15.0) $\mu g/dL$). Exposed workers had decreased level in albumin, glucose, total protein, and hemoglobin when compared with normal level in human blood (p<0.0001). The results also confirmed that, there was significant relation between exposure to benzene and increase in blood cholesterol (p<0.01). The correlation between benzene exposure and the blood lead level was confirmed by comparing with the non-exposed group (p<0.01).

^{*}e-mail: mmkali@uqu.edu.sa

The outcomes from the exposure call upon increased concern for policy action to avoid the adverse effects of benzene on gasoline stations workers' health.

Keywords: benzene exposure, biochemical changes, gasoline stations, lead toxicitie

Introduction

The city of holy Makkah in the Kingdom of Saudi Arabia aside from its tremendous population and physical expansion, attracts a large influx of pilgrims during the Hajj and Umra seasons, as well as many other visitors throughout the year. Therefore, the demand for transportation is increasing as a result of a sharp rise in the number of cars and petrol consumption which consequently reflected in a dramatic surge in the numbers of gasoline filling stations and workers at these stations throughout the city.

Gasoline stations in Makkah are busy and open for longer working hours and are full-service stations where fuel filling is usually performed manually by workers who are exposed to petrol hazardous substances such as benzene from gasoline fumes and motor vehicle emissions during filling time.

Benzene is a natural component of crude oil and also found in different concentrations in refined products such as gasoline. It is a colorless liquid and highly volatile at room temperature [1]. Benzene is considered as one of the most dangerous gasoline constituents due to its carcinogenic potentiality [2-4]. The major routes of exposure pathways of benzene are inhalation, ingestion, and skin contact. Human exposure occurs primarily through inhalation particularly in occupational settings [5].

Makkah city features is a hot desert climate, the temperature is high all year around and it can reach up to 45°C (113°F) in the summer; hence benzene evaporation will be at higher rate in filling stations and lead to significant concentrations in the air.

In both humans and experimental animals, benzene exposure has been linked to high blood lead level (BLL), hematopoietic disorders such as lack of bone marrow which results in a drop in the rate of circulating blood

corpuscles, anemia, thrombocytopenia, leucopenia, aplastic anemia, and severe myelogenous leukemia [6, 7]. Furthermore, the adverse effect of benzene on workers' health has been shown in investigation carried by Choi et al., 2019 [8], who reported that workers exposed to oil industry chemicals could emerge lungs, skin, and other organs diseases to varying degrees rely on the length of exposure.

Biomonitoring the health of workers at gas stations who are chronically and directly exposed to benzene is an important preventative measure for occupational diseases. Therefore, the goal of this study was to look into the effects. of benzene on BLL and selected blood biochemical parameters among workers at different petroleum stations in Makkah city, Kingdom of Saudi Arabia.

Materials and Methods

Study Population

Characteristics of the study population, obtained through questionnaire interview. This study enrolled 63 people, including 19 samples from women as non-exposed group and 44 gasoline station workers as exposed group. All of the exposed workers had been working in their current positions for at least one year. Each participant was interviewed to learn about their general health, lifestyle, smoking habits, and history of exposure.

Collection of Blood Samples

Blood samples were collected from 44 male individuals as exposed group by visiting the petrol pumps located in Makkah city at different sites as well

TC 1 1 1	D: / '1 /'	C/1 D1 11	11 1 .	1: 4 4:	1 1	non-exposed groups.
Table 1.	Distribution	or the brook ic	iu icveis iii	Easonine stations	workers and	HOH-CADOSCU ETOUDS.

Blood lead level (µg/L)	Exposed group	Non - exposed group
≤ 5 (The reference blood lead level for adults)	1 (2.27%)	18 (94.7%)
5-	1 (2.27%)	0 (0.0%)
10-	3 (4.54%)	1 (5.3%)
20 -	2 (4.54%)	0 (0.0%)
30-	7 (15.90%)	0 (0.0%)
40-	10 (22.73%)	0 (0.0%)
> 50	21 (47.73%)	0 (0.0%)
Total	44	19

as 19 female individuals as non-exposed group who were not subjected to any kind of benzene exposure. Approximately 10 ml of blood was collected from each individual with sterilized or disposable syringes, equipped with stainless steel tips, which were used once only. 5 mL sterile syringes were used for each venipuncture, and the blood was placed in a dry and clean plain container. After coagulation, the tube was centrifuged at 4000 rpm for 15 min. The serum was transferred into plain container equipped with tight-fitting caps by disposable tips, then stored at -20°C. 5 mL sterile syringes were used, and the blood was placed in an ethylenediaminetetraacetic acid (EDTA) container for hemoglobin estimation using an automated complete blood count (CBC).

The kits for determination of lead, glucose, total protein, cholesterol, albumin, and creatinine were obtained from Medical Scope Company, Saudi Arabia. The measurements were made using a flameless atomic absorption spectrometer (AAS).

Data Analysis

Statistical Package for the Social Sciences (SPSS) version 23 was used to entering and analyzing the data obtained from questionnaire. One sample t-test was used to detect the effect of benzene exposure on glucose, total protein, cholesterol, albumin, and creatinine by comparing with the normal level. The analysis of variance (ANOVA) test was used to explore the lead toxicity in benzene exposed group and non-exposed group. Individual test was used to detect the correlation between BLLs and years of working in gasoline stations.

Results and Discussion

A total of 63 individuals, including 44 samples from gasoline station workers as exposed group and 19 samples from women who were not subjected to any kind of benzene exposure as non-exposed group

were recruited to investigate the effects of benzene on BLL and selected blood biochemical parameters among workers at different petroleum stations in Holy Makkah city. the results obtained for the distribution, median and range values of lead levels in blood of non-exposed subjects and gas stations employees are shown in Tables 1 and 2. The findings showed that fuel station workers have significantly higher BLL compared to the nonexposed group. According to the Centers for Disease Control and Prevention in US (CDC) elevated BLL for adult are defined as BLL ≥5 µg/dL [9]. The obtained result showed that 94.7% of exposed workers compared to only 2.27%% of non-exposed group have elevated BLL. This result gives a clue to the distinct relationship between exposure to gasoline and the blood level of lead intoxication. The median (range) blood lead value of the exposed fuel stations workers was 40.5 (4.0-90.0) µg/dl. While the median (range) blood lead value in non-exposed healthy women was 3.2 (2.7-15.0) µg/dl. The difference between the two groups was extremely significant (p = 0.00). These findings are in consistent with that previously reported by Eltayeb et al., 2014 [6].

On the other hand, correlation between benzene inhalation exposure and the BLL was studied. Positive correlation was revealed by comparing the findings in the exposed group to the non-exposed group and was considered significant (p<0.01) as shown in Table 3. This finding supports the hypothesis that exposure to gasoline and benzene increased BLL in fuel station workers.

Adverse biochemical effects of benzene exposure and high BLL are well known today [10, 11]. Therefore, here in this study we analyzed different biochemical parameters in blood of exposed workers and their values were compared to normal human references to find out are there any changes in workers' biochemical profile.

The results obtained showed that exposed workers had decreased levels of albumin, glucose, total protein, and hemoglobin when compared with normal level in human blood (p<0.0001) and increased the level of cholesterol as shown in Table 4.

Table 2. Blood lead levels in μ g/dl among fuel stations workers and non-e	-exposed group
--	----------------

Group	Number	Median (range)	Std Deviation	P- Value
Exposed group	44	40.5(4.0-90.0)	20.7	0.00
Non - exposed	19	3.2 (2.7-15.0)	2.7	0.00

Table 3. The Relationship between benzene exposure and blood lead level

Group	Number	Mean	Std Deviation	F	P- Value
Exposed	44	4.4591	1.7813	6.142	0.016
Non-exposed	19	3.4105	0.6854		
Total	63		_		_

1476 Khalafalla M.M., et al.

Mean of Normal	Mean	SD	Mean Difference	SD error mean	P-Value
Albumin g/dl (4.5)	3.6045	0.64338	0.89545	0.0969	0.0001
Glucose g/dl (140)	110.7	45.177	29.29	6.8108	0.0001
Cholesterol mg/dl (200)	219.022	47.39	19.022	7.1444	0.011
Creatinine mg/dl (1.05)	1.097	0.3944	0.0477	0.0594	0.427
Total-Protein g/dl (7.15)	5.747	1.3592	0.2049	1.4022	0.0001
Hemoglobin (HB) g/dl	12.6364	2.2056	2.8636	0.3325	0.0001

Table 4. The effect of benzene exposure to biochemical parameters in the blood.

Generally, the relation was significant between exposure to benzene aerosol inhalation and effects on levels of these biomedical parameters. Previous studies carried by Teklu et al., 2021 [12] have declared that the harmful effects of gasoline are mainly caused by benzene metabolites. Following gasoline inhalation, benzene, which is the primary components of gasoline, enters the lungs, then travels through the bloodstream to the liver, where three major phenolic metabolites of benzene are produced and accumulated in relatively high levels, and then to bone marrow, where actual toxic elements are produced [13-15]. Moreover, our results showed that exposed workers had increased level of blood cholesterol and significant relationship between benzene exposure and increased blood cholesterol levels. (p<0.01) (Table 4). This complies with the result previously reported by Abplanalp et al., 2017 [16] who found that total cholesterol was increased in benzeneexposed mice compared with mice breathing filtered air. Moreover, Khan et al., 2013 [17] reported significant increase in total cholesterol of participants compared to subjects who were not exposed to organic solvents. All these investigations declared that exposure to different chemicals including benzene at working place has an adverse effect on the cholesterol level. Since cholesterol is one of the important predictors of cardiovascular diseases, these workers are at risk of developing cardiovascular diseases [12].

(15.5)

For blood creatinine level, our result showed that exposed workers had no evident effect on blood creatinine level (p>0.05), according to the investigation of the methodology used in this study (Table 4). However, other studies found significant increase in the creatinine levels of the gasoline station workers compared with the non-exposed subjects [18, 19]. This disparity could be attributed to the chemical concentrations, duration of exposure, and experimental subjects used.

The effects of benzene on other blood parameters such as hemoglobin which is the carrier of oxygen to the body tissues, blood glucose the main nutrient of human cells, total protein and albumin which is the main protein serum responsible for many vital processes in the human body indicated the decrease in hemoglobin level in the exposed subjects (p>0.0001) as compared to normal value. In line with this finding Teklu et al. 2021, Robert et al., 2010, Nair et al., 2015 and Neghab et al., 2014 [12, 20-22], reported drop in hemoglobin levels among gas station employees who were exposed to gasoline versus those who were not. They attributed this drop in the measured hemoglobin level to the shortened life cycle of red blood cells (*RBCs*) and a reduction in heme production by the final product of metabolism of benzene free radicals and other gasoline aliphatic hydrocarbon elements.

Furthermore, our results showed a decrease in blood glucose level (29.29 mean difference from the normal level (p>0.0001). This could be a very critical issue when workers with conditions of metabolic disorders such as diabetes mellitus are associated. This finding is in agreement with previous results which declared that the exposure to benzene affected blood glucose and caused reduction in liver and muscle glycogen stores [23].

For the total serum protein, the data showed that it was negatively affected. A mean drop of approximately 20% from the normal was noticed, as well as a same mean reduction in serum albumin value (p>0.0001). These finding are in agreement with the previous finding reported by Mohamed et al., 2016 [24] and Saeed et al., 2015 [25], where significantly decreased albumin levels in benzene exposed people compared to the non-exposed ones was shown.

Conclusion

This investigation showed that lead blood level and some biochemical parameters of gasoline station's workers could negatively be affected by the influence of the chronic exposure to benzene, leading to adverse effects on their general health condition, especially when combined with their expected low-socio-economic status and low health consciousness. Such findings call for alert to more vigilance and prompt action, both in regard to health authorities and policy and regulation of labor, in pursuit of protection of this vulnerable group. Moreover, these workers should have regular medical checkups, which should include an evaluation of their blood profile as well as blood benzene and lead levels measurements. Furthermore, administrative controls and proper policies by relevant authorities such as using protective equipment's like coverall and facemask to reduce exposure, educating and training gasoline station's workers on the exposure health hazards associated with gasoline components, the typical routes of exposure, improving work practices, and exposure avoidance techniques (e.g., standing upwind of the vehicle tank openings) are recommended.

Conflicts of Interest

The authors declare they have no conflicts of interest.

Acknowledgment

The authors would like to thank Institute of Scientific Research and Revival of Islamic Heritage at Umm Al-Qura University (Project # 43409065) for the financial support.

References

- D'ANDREA M.A., REDDY G. Health risks associated with benzene exposure in children: A Systematic Review. Glob. Pediatr. Health. 5, 1, 2018.
- AHMADI Z., MORADABADI A., ABDOLLAHDOKHT D., MEHRABANI M., NEMATOLLAHI M.H.J. Association of environmental exposure with hematological and oxidative stress alteration in gasoline station attendants. Environmental science and pollution research international 1, 2019.
- 3. ABOU-ELWAFA H.S., ALBADRY A.A., EL-GILANY A. H., BAZEED F.B. Some biochemical and hematological parameters among petrol station attendants: a comparative study. BioMed research international. 1, 2015.
- ABUBAKAR M. B., ABDULLAH W. Z., SULAIMAN S. A., ANG B. S. The effects of exposure to petrol vapors on growth, hematological parameters and oxidative markers in SpragueDawley male rats. Malaysian Journal of Medical Sciences. 22, (1), 23, 2015.
- POLI D., MOZZONI P., PINELLI S., CAVALLO D., PAPALEO B., CAPOROSSI L. Sex difference and benzene exposure: Does It Matter? Int. J. Environ. Res. Public Health. 19, 2339, 2022.
- ELTAYEB T., NAGEEB A., ALI K.T. Blood lead level among fuel station workers at Khartoum city. American Journal of Research Communication. 2 (6), 74, 2014

- ZEINAB A.K., GHADA M.K., GAAFAR M.A., AHMED A.I. Environmental and health effects of benzene exposure among Egyptian taxi drivers. Journal of Environmental and Public Health. 1, 2019.
- CHOI S., KWAK S., PARK D., JEONG J.Y. Potential Risk of Benzene in Petroleum-derived products used from 1974 to 2012 in Korea. Aerosol Air Qual. Res. 19, 548, 2019.
- CENTERS FOR DISEASE CONTROL AND PREVENTION (C. D. C.). Lead Available at: https:// www.cdc.gov/nceh/lead/default.htm. (Accessed July 11, 2018), 2018.
- ZHOU Y., WANG K., WANG B., PU Y., ZHANG J. Occupational benzene exposure and the risk of genetic damage: a systematic review and meta-analysis. BMC Public Health., 20, 1113, 2020.
- DONGRE N. S., ARUN J. P., JEEVAN G. A., DILEEP B. R. Biochemical effects of lead exposure on systolic and diastolic blood pressure, heme biosynthesis and hematological parameters in automobile workers of North Karnataka (India)., Indian J Clin Biochem. 26, (4), 400, 2011.
- 12. TEKLU G., NEGASH M., ASEFAW T., TESFAY F., GEBREMARIAM G., TEKLEHAIMANOT G., WOLDE M., TSEGAYE A. Effect of gasoline exposure on hematological parameters of gas station workers in Mekelle City, Tigray Region, Northern Ethiopia. Journal of Blood Medicine. 12, 839, 2021.
- 13. CARBONARI D., CHIARELLA P., MANSI A., PIGINI D., IAVICOLI S., TRANFO G. Biomarkers of susceptibility following benzene exposure: influence of genetic polymorphisms on benzene metabolism and health effects. Biomarkers in Medicine. 10 (2),145, 2016.
- 14. MITRI S., FONSECA A.S., OTERO U.B., TABALIPA M.M., MOREIRA J.C., SARCINELLI P.D. Metabolic polymorphisms and clinical findings related to benzene poisoning detected in exposed Brazilian gas-station workers. International Journal of Environmental Research and Public Health. 12 (7), 8434, 2015.
- KNUTSEN J.S., KERGER B.D., FINLEY B., PAUSTENBACH D.J.A. Calibrated human PBPK model for benzene inhalation with urinary bladder and bone marrow compartments. Risk Analysis. 33 (7), 1237, 2013.
- 16. ABPLANALP W., DEJARNETT N., RIGGS D.W., CONKLIN D.J., MCCRACKEN J.P., SRIVASTAVA S., XIE Z., RAI S., BHATNAGAR A., O' TOOLE T. E., Benzene exposure is associated with cardiovascular disease risk. PLOS ONE. 12, 1, 2017.
- 17. KHAN A.A., SULTAN R., ZAMANI G.Y. Biochemical and hematological analysis after exposure to hazardous materials during shoe making. Journal of Biology and Life Science. 4 (2), 116, 2013.
- BIN-MEFRIJ M., ALWAKEEL S. The effect of fuel inhalation on the kidney and liver function and blood indices in gasoline station workers. Advances in Natural and Applied Sciences. 11 (1), 45, 2017.
- OLMEDO-BUENROSTRO B.A., ORTEGA-ORTIZ J.G., GUZMAN-ESQUIVEL J., DELGADO-ENCISO O.G., CEJA-ESPIRITU G., PAZ-MICHEL B.A. Workplace gasoline exposure increases the risk for early renal dysfunction: A case-control study in Mexico. Biomed Research. 28 (22), 859, 2017.
- ROBERT SCHNATTER A., KERZIC P.J., ZHOU Y. Peripheral blood effects in benzene-exposed workers. Chem. Biol. Interact. 184, (1-2), 174, 2010.
- NAIR D.S., BEDEKAR M.Y., AGRAWAL M.J. Deleterious effects of petrol fumes on erythrocytes.

- International Journal of Health Sciences and Research. 5, (9), 237, 2015.
- 22. NEGHAB M., HOSSEINZADEH K., HASSANZADEH J. 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. 6, (4), 312, 2015.
- 23. IYANDA A.A., ANETOR J.I. Biomarkers of hepatorenal damage of fuel filling station attendants using or abstaining from use of protective gears. Clinical Chemistry and Laboratory Medicine. 3, 15, 2017.
- 24. MOHAMED A.H., HASAN M.I., IBRAHIM A., SULIMAN O., ELIMAIRI G. Assessment of liver function tests in benzene station workers in Khartoum state-Sudan. International Journal of Current Research. 8, 30683, 2016.
- 25. SAEED H.S.A., ABDELLAH A.M., ABDALLA F.A. B., ABBAS A.R.A., ADAM F.A., ELGAZALI N.A. Biochemical effects of lead toxicity on serum total protein, albumin and globulin levels in occupationally exposed workers in major Sudanese cities. International Journal of Emerging Technology and Advanced Engineering. 7, 132, 2015.