

## Evaluation Some Heavy Metals and Antioxidants in the Blood of Workers in the Benzene Filling Station in the City of Hilla

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### ABSTRACT

The results of presents showed evaluation of lead , cadmium elements and liver function such as Alkaline phosphatase, Alanine transaminase and Aspartate transaminase and blood parameters such as White blood cells , red blood cell , hemoglobin and package cell volume in workers station of filling benzene in Al-Hilla city, whereas lead and cadmium concentration in blood significantly increased at  $P \geq 0.05$  when compared with control , All liver function markers and blood parameters were significantly increased in workers as compared with control.

**Keywords:** Heavy Metals, Antioxidants, Blood of Workers, Iraq

### INTRODUCTION

Benzene is a major monocyclic aromatic hydrocarbons largely used as solvent in a variety of industrial and commercial processes. Benzene is used as a feeder chemical in the manufacture of lubricants, detergents, rubber, dyes and pesticides. It is often found in automobiles and solvent benzene. A large section of population is occupationally exposed to benzene through work environment (Abbas *et al.*, 2020). Chronic exposure to benzene results in progressive decline of hematopoietic function and may lead to the onset of various disorders, including aplastic anemia, melody's plastic syndrome and leukemia (Caruso *et al.*, 2013). Liver injury had long been associated with occupational exposure to a wide variety of chemicals (Beelen *et al.*, 2014).

Its susceptibility to chemical injury is a result of its unique position within the circulatory system, and also because it is the primary organ for the biotransformation of chemicals within the body, as liver is the main organ involved in the metabolism of toxins and medicinal agents Such metabolism is always associated with the disturbance of hepatocyte biochemistry and generation of reactive oxygen species (ROS) which are assumed to induce oxidative damage on liver, kidney and hematopoietic system (AL-Janabi *et al.*, 2025; Bae *et al.*, 2010).

The concentrations of ROS have to be controlled by several defense mechanisms, which involve also a number of antioxidant and detoxifying enzymes, their induction reflects a specific response to pollutants, A balance between free radical reactions and antioxidant activities is very important for normal liver functioning (Qassim *et al.*, 2023). This balance is altered in pathological processes, A lot of liver damages ranging from subclinical hepatitis to necrosis inflammatory hepatitis, cirrhosis, and carcinoma have been proved to associate with the redox imbalance and oxidative stress, The most common chemicals known to cause liver injury are the organic solvents as benzene, acetone, ether, and so on (Saini *et al.*, 2013)

There is some evidence that organic solvents especially benzene may express their toxicity by the way of ROS that was found to induce cell damage [4] Moreover, higher incidence of cancer is suspected in subjects exposed to organic solvents as benzene characterized by reactive metabolic intermediates, Workers in the fuel stations are routinely exposed to benzene vapor that has been reported to increase the risks for acute and chronic health problems in motor fuel workers (Mohammed *et al.*, 2019).

Activation of benzene and its reactive metabolites leads to continuous production of reactive oxygen species (ROS), which leads to lipid peroxidation and damages DNA, RNA, leading to genetic modification and alterations in the functions of important enzymes and proteins chronic benzene exposure leads to decrease in antioxidant enzymes activity and hematologic disorders Benzene affects many enzyme activities in the liver, tissues, and peripheral blood and this can lead to a decrease in the activity of antioxidants enzymes and may result in oxidative stress which can be defined as the unbalance between the generation of reactive oxygen species (ROS) and the rate of their consumption by antioxidants, Oxidative stress occurs when the critical balance between oxidants and antioxidants is disrupted due to the depletion of antioxidants or excessive accumulation of the reactive oxygen species (ROS), or both, leading to a damage of the cells ,No single component of serum antioxidant complex could fully reflect the protective efficiency of blood, probably because of interactions that occur in vivo among different antioxidant compounds, Total antioxidant capacity (TAC) considers the cumulative effect of all antioxidants present in blood and body fluids. (Sadhana *et al.*, 2025). This study was aimed to evaluate the hazardous effects of occupational exposure to benzene vapor in fuel-station workers on the liver and hemopoietic system through Measurement some heavy metals such as Lead Pb and Cadmium Cd in whole blood in fuel station workers , Measurement of liver enzymes such as Alkaline phosphatase ALP, Alanine transaminase ALT and Aspartate transaminase AST and Measurement of WBC, RBC Hb and PCV. (Prasad *et al.*, 2013).

## 2 MATERIALS AND METHODS

### 2-1 Blood collection

Blood Samples Collection and Assay: About 10 ml of venous blood was collected from each of them by sterile disposable syringes. Venous blood samples collected in plain tubes without using an anticoagulant and was left to clot for 30 min. at 25°C, the blood was centrifuged at 4,000 rpm for 15 min. at 4°C. The top yellow serum layer was pipette off without distributing the white buffy layer. The separated serum was used for estimation of liver enzymes; the other part was collected in clean tubes with EDTA as anticoagulant substance for estimation of blood parameters and lead and cadmium elements (Munajad *et al.*, 2018).

### 2-2 Measurement of Heavy metals in blood

Taken 2 ml of Whole blood were collected from Workers in EDTA tube , At the laboratory, samples were acid-digested by adding (1.5) ml of nitric acid to maintain in ionic form then put the sample at heater to evaporate the sample to pre-dry then complete the sample to 10 ml by deionized water then measure by Atomic Spectrometer type of AA-7000 Shimadzu (Japan)in Laboratory of advance environmental –Sciences in Al-Qasim green university for lead and cadmium elements (Qassim *et al.*, 2019).

### 2-3 Enzymes of liver function

The activity of alkaline phosphatase ALP, Alanine transaminase ALT and Aspartate transaminase AST had been measured for serum of a blood by reflotron plus roche (Qassim *et al.*, 2021).

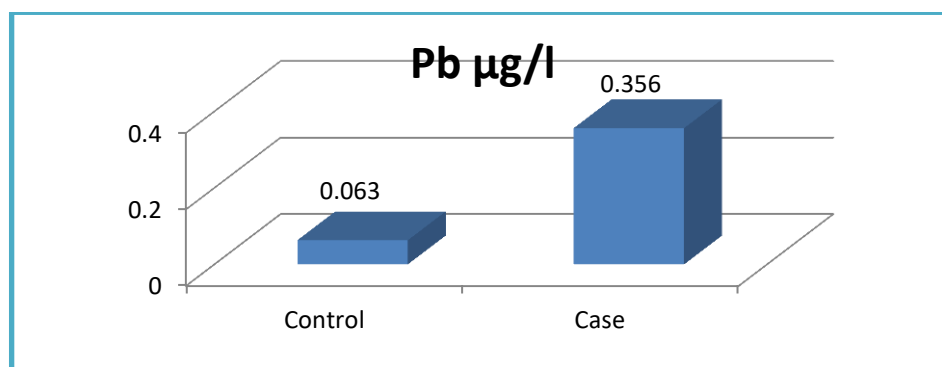
### 2-4 Hematological parameters

All blood parameters were determined by complete blood count (CBC) system. All sample of treatment were done in triplicate (Qassim *et al.*, 2024).

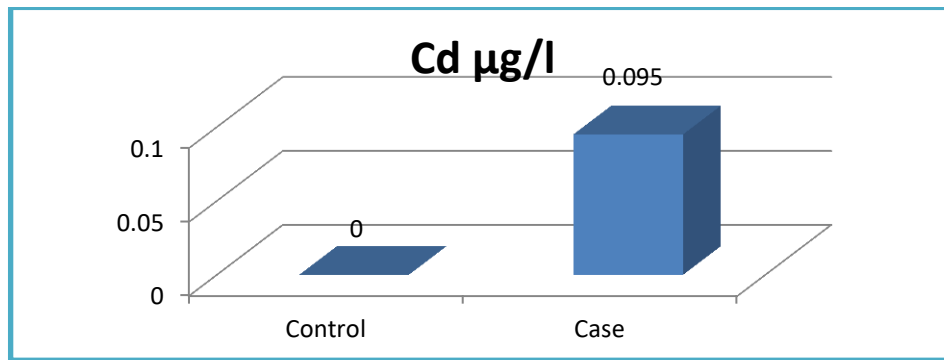
## 3 RESULTS

### 3-1-1 Heavy Metals in Serum of workers in filling benzene station

The results of presents study showed significant differences between control and case with workers in station of filling benzene at ( $P < 0.05$ ) in heavy metals elements, the lead pb concentration in serum control (mean  $\pm$ SD) was  $0.063 \pm 0.01 \mu\text{g/l}$  While its concentration in serum case with workers in station of filling benzene were significantly increased and reached to  $0.356 \pm 0.021 \mu\text{g/l}$  Figure (1) Whereas the concentration of cadmium cd was nil ( $0 \mu\text{g/l}$ ) in control While in case with workers in station of filling benzene, its concentration were  $0.095 \pm 0.001 \mu\text{g/l}$  Figure (2).



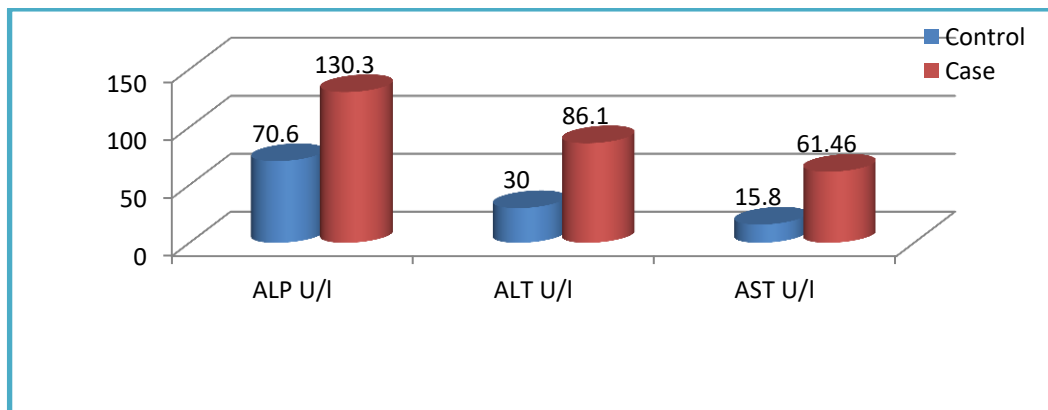
**Figure 1: Concentration of lead in serum of workers in station of filling benzene**



**Figure 2: Concentration of cadmium in serum of workers in station of filling benzene**

### 3-1-2 Liver function Parameters

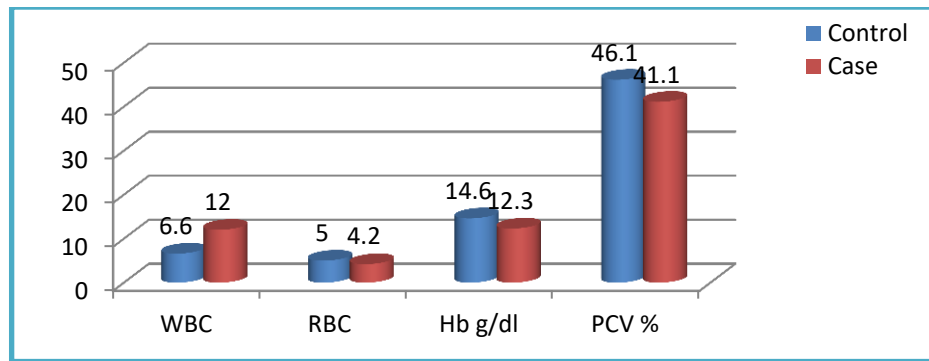
The result of present study showed significant differences between control and cases in serum of workers of filling benzene station in all liver function parameters, the activity of alkaline phosphatase ALP in cases (workers of filling benzene station) was significantly increased and reached to (130.3±4.56) U/l While alkaline phosphatase activity in control (70.6± 10.8) U/l, Whereas Alanine transaminase activity ALT in control was reached to (30 ± 2.65) U/l, While in case of workers of filling benzene station, its activity was significantly elevated to (86.1± 17.91) U/l and the activity of aspartate transaminase AST in case was significantly increased to reach (61.46±18.4) U/l compared with control groups (15.8±3.57) U/l figure (3).



**Figure 3: Activities of Alkaline phosphatase ALP, Alanine transaminase ALT and aspartate transaminase AST in serum of workers in station of filling benzene**

### 3-1-3 Blood parameters

The number of white blood cells in case of workers of filling benzene station were significantly increased and reached to (12±3.54) 10<sup>3</sup>/µL as compared with control groups (6.6 ± 1.37) 10<sup>3</sup>/ µL, Whereas number red blood cells in control was reached to (5±1.76) 10<sup>6</sup>/µL, While in workers of filling benzene station, its number was decreased to (4.2 ±1.21) 10<sup>6</sup>/µL as compared with control which its numbers increased (5± 0.78 ) 10<sup>6</sup>/µL and the level of hemoglobin in cases was to reach (12.3 ± 2.90) g/dl While Hb level in control was (14.6 ± 3.56) g/dl, the packed cell volume PCV % in the control was (46.1 ± 16.43) % and its percentage in workers of filling benzene station were to reach (41.1± 13.4)% Figure (4).



**Figure 4: Blood parameters in whole blood of workers in station of filling benzene**

#### 4 DISCUSSION

Petroleum hydrocarbons such as benzene are converted into free radicals or activated metabolites during their oxidation in cells especially mammalian liver and kidney cells, It is these activated metabolites that react with some cellular components such as membrane lipids to produce peroxidation products which lead to membrane change,[11] They may react with enzymes and cause inactivation through protein oxidation and DNA breakage of the strands (Menden *et al.*, 2019).

A common clinical finding in benzene hematotoxicity is cytopenia, which is a decrease in various cellular elements of the circulating blood manifested as anemia, leukopenia, or thrombocytopenia in humans and in animals, Benzene associated cytopenias vary and may involve a reduction in one (unicellular cytopenias) to all three (pancytopenia) cellular elements of the blood. The results of present study showed the lead and cadmium concentration in whole blood of workers of filling benzene station was significantly increased when compared with control group due to workers of filling benzene was daily exposure to automobile exhaust (Warren *et al.*, 2014).

Enzymes are useful bio-markers used in assessing specific functions and integrity of a cell, especially hepatocytes, an increase levels in these enzymes activities in the plasma are linked to hepatocellular damage caused by either toxins, toxins in drugs or herbs, This study was conducted to determine the activities of liver enzymes in serum of workers, There is a significant increase in the levels of serum liver enzyme such as aspartate aminotransferase, alanine aminotransferase and alkaline phosphatase in petrol workers when compared with control group due to benzene fumes The hepatic enzymes serum levels such as ALP, ALT and AST had been used as markers for hepatic injury (Singh *et al.*, 2015). The alkaline phosphatase (ALP) plays a significant role in phosphate hydrolysis and in membrane transport as well as is a good bio-indicator of stress in biological systems, the importance of measuring alkaline phosphatase is to check the liver dysfunction and the cellular membrane health (Qassim, *et al.*, 2021).

RBC counts, Hb concentration, and PCV% were measured in the study and control groups. Study shows significant decrease in RBC counts and Hb concentration along with a decreasing in PCV%, which is non-significant due to bone marrow depression caused by benzene. White blood cells was significant increase in workers of filling benzene as compared with control groups due to petrol fumes Petrol fumes are therefore environmental pollutants that could have serious consequences on hematological parameters in exposed humans, Benzene also causes a life-threatening disorder called aplastic anemia in humans and animals, This disorder is characterized by reduction of all cellular elements in the peripheral blood and in bone marrow, leading to fibrosis, an irreversible replacement of bone marrow. Benzene has also been associated with acute non-lymphocytic leukemia in humans, and aplastic anemia may be an early indicator of developing acute non-lymphocytic leukemia in some cases (AL-Janabi *et al.*, 2025).

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