Evaluation of Biomarkers in Workers Exposed to Air Pollutants in Oil Refineries

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Abstract

Air pollution is nowadays a complex problem due to industrial prosperity. Among the chemical industry, oil refineries have been identified as major emitters of a wide range of pollutants, the workers of the oil refinery are exposed to a great variety of toxic compounds. Air pollution is the main environmental cause of human disease and death. Therefore, it is necessary to develop early warning signals or biomarkers that convincingly reflect adverse biological responses towards anthropogenic environmental toxins even at minute concentrations. This work aims to study the effects of exposure to the air pollution of oil refinery, on the parameter blood, TNF-a, antioxidant glutathione peroxidase(GPx), and oxidative stress malondialdehyde (MDA), in workers of the oil refinery. Results showed that exposure to air pollution in the oil refinery lead to(a significant increase in levels Hb, WBC, Lymphocyte, TNF-a, and MDA, decrease levels of GPx) $P<0.05$. And non –significant in(RBC, HCT, Neutrophils) $P>0.05$ in workers, compared to healthy control.

Keywords: *air pollution; oil refinery workers; oxidative stress*

Introduction

Air pollution is a phenomenon in which(liquid or solid) particles and gases contaminate the atmosphere and such contamination can impact the population[1]. In other words, an air pollutant is any gas or particulate that may be harmful to life, the environment, and/or property when its concentration is high enough. Among the chemical industry, oil refineries have been identified as major emitters of a wide range of pollutants [2]. The workers of the oil refinery are exposed to a great variety of toxic compounds [3]. The most important gases emitted from oil refineries, which are considered air pollutants are hydrocarbons (HC), CO, NOx, H2S, CH4, CO2, SO2, and particulate matter(PM) [4,5]. Environmental contaminants create harmful conditions for living organisms, including humans. This represents the growing interest in early warning tools for the detection of adverse biological responses to toxins in both humans and wildlife. Molecular and cellular biomarkers of pollution meet this requirement. pollution biomarker is an alteration in a biological response occurring at molecular, cellular, or physiological levels which can be related to exposure to or toxic effects of environmental chemicals [6].

Air pollutants stimulate the immune system to activate leukocytes and macrophages[7]. Cytokines are a group of proteins of low molecular weight secreted by the cells of the immune system. they regulate the inflammatory and immune response. these include,pro-inflammatory(TNF-a)[8]. Oxidative

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stress is a biochemical imbalance in which the production of reactive oxygen species exceeds the natural antioxidant capacity. In the body, this imbalance may occur after exposure to pro-oxidant air pollutants. Reactive oxygen species cause tissue damage and dysfunction in the presence of oxidative stress by targeting and denaturing structural and functional molecules (lipids, proteins, carbohydrates, DNA, etc.)[9]. Chemical components of air pollution exposures that cause oxidative stress and subsequent inflammation may be responsible for associations of cardiovascular morbidity and mortality with pollutant gasses associated with airborne particulate matter combustion. One feasible approach is to measure systemic oxidative stress in the blood for health effects. It is important to measure the genes and/or protein expression of endogenous antioxidant enzymes when measuring oxidative stress, since they may modify the relationship between biomarkers of oxidative stress and air pollutants [10].

Materials and Methods

A total of 70 Iraqi individuals (males) in Basra (Southern Iraq) were included in this study, 40 individuals working in the Shuaiba refinery in Basra exposed to air pollutants during work(Exposed group). their ages ranging between 18-55 years. The other 30 individuals are non-exposed to air pollutants(not work in the refinery), they were considered as (Control group), this group ages ranging between 18-55years.

Blood parameters determined from complete blood count that was performed by hematology analyzer Sysmex. Total serum tumor necrosis factor-alpha (TNF-a) was determined by the use of a total TNF-a ELISA(enzyme-linked immunosorbent Assay) kit(Sunlong, china). The level of GPx in serum was estimated by using commercially available kit ELASA (Elabscience, USA). The level of MDA was determined by the Buege and Aust method [11].

Spss program (version 26) was used to elucidate the difference in parameters. The results were expressed as mean±SD. Comparisons were made between two groups using the T-test for categorical. The p-value of<0.05 was considered to indicate statistical significance.

Results and Discussion

The means of red blood cell (RBC)in the exposed workers was 5.41 ± 0.5and in controls 5.21 ± 0.4, Hematocrit (HCT) in exposed workers was 43.58±3.9% and in controls were 44.08± 3.1, mean corpuscular hemoglobin (MCH) in exposed workers was 28.45 ± 2.5 and in controls was 28.43 ± 1.9, and Neutrophil count in exposed workers was 3.87±1.7 while in control 4.24±1.3. These results showed that the values of previous parameters within the normal range with insignificant differences between exposed workers to air pollution and controls. However, hemoglobin in exposed workers was 15.33 ± 1.4 g/dL and in controls was 14.64 ± 1.1, WBC in exposed workers was 7.64± 2.1(10^3/uL) and in controls was 6.68 ± 1.4, and Lymphocyte(LYM) in exposed workers was 2.87± 0.7and in controls was 2.31± 0.4. All previous results showed a significant increase in exposed workers compares to control, as shown in table 1.

Table 1: Mean and standard deviation of the blood parameters

parameters	Exposed (mean±SD)	Controls (mean±SD)	P-value	
Hb(g/dL)	15.33±1.4	14.64±1.1	0.040	S
RBC(10^6/uL)	5.41±0.5	5.21±0.4	0.108	NS
HCT%	43.58±3.9	44.08±3.1	0.568	NS

***Cont.*. Table 1: Mean and standard deviation of the blood parameters**

MCH(pg)	28.45±2.5	28.43±1.9	0.973	NS
WBC(10^3/uL)	7.64±2.1	6.68±1.4	0.041	S
NEU(10^3/uL)	3.87±1.7	4.24±1.3	0.348	NS
Lym(10^3/uL)	2.87±0.7	2.31±0.4	0.000	S

S:significant,NS:non-significant

Table 2:level TNF-a in exposed and controls

Groups	Exposed	Control
Mean	162.78 ng/L	148.27ng/L
S.D.	23.57	24.91
P-value 0.015		

Table 3:level biomarker oxidative stress in exposed and controls

	Exposed Mean± SD	Control Mean± SD	P-value	
GPx pg/ml	1273.0±423.08	1527.3 ±373.57	0.011	S
MDA µmol/L	1.34±1.02	0.73±0.7	0.007	S

This study's results demonstrated the existence of a significant correlation between occupational air pollution exposure to oil and gas activity in the environment and Hb, as shown in table 1.There was an increase in the level of hemoglobin(Hb) in the exposed compared to the control group, and this is the case agreed with other studies[12]. Hb is a major component of red blood cells that has a key role in oxygen and carbon dioxide transport in the body [13]. Some of the gases that pollute the air contribute to a rise in the blood hemoglobin level, such as CO. This gas enters the bloodstream and binds to the protein Hb. This can reduce the blood's ability to deliver oxygen to the body's tissues. As a means of compensation, the body can increase the erythropoiesis process, potentially increasing Hb production[14].

The current study showed that the level of white blood cell (WBC) was a significant increase in exposed workers compared with control. WBC are cells of the immune system involved in defending the body against both infectious diseases and foreign materials. Inhaling dirty air can trigger the releases of WBC into the bloodstream and can result in inflammation. Through this result, exposure to air pollutants may increase the number of WBC, this case agreed with another study [15].

The mean of LYM in exposed workers is significantly high as compared to controls. LYM are the type of WBC that is of fundamental importance in the immune system because the lymphocytes are the cells that determine the specificity of the immune response to infectious microorganisms and other foreign substances [16]. This result indicates that exposure to air pollutants affects LYM levels, and this is the case agreed with another study [17].

The serum level of TNF-a a significant increase $p<0.05$ in exposed workers with serum median levels (162.78±23.5 ng/L) compared to control group serum median levels(148.27±24.9ng/L), as shown in table 2.

In this study, the exposed workers showed a significant increase of pro-inflammatory biomarker TNF-a due to exposure to air pollution. The inhalation of toxic environmental gases and particles impacts the defense systems of the lung. Lung macrophages play a critically important role in the recognition and processing of any inhaled foreign material such as pathogens or toxic gases and particles. Alveolar macrophages and lung epithelial cells are the predominant cells that process and remove inhaled air pollutants from the lung. Cooperatively, they produce pro-inflammatory mediators when exposed to air pollutants [7]. Cytokines are mainly secreted by T helper cells (Th) and macrophages, they regulate the immune and inflammatory response and are primarily involved in the events of pathogenesis in air pollution-related diseases. They may also have the potential to be used as indicators for assessing the harmful effects of air pollution [18]. TNF-a is a major pro-inflammatory cytokine produced by macrophages and the generation and development of inflammatory reactions and related diseases[19]. The air pollutants can stimulate pro-inflammatory cytokine production by macrophages [20] Studies indicate that inhalation of air pollutants can result directly or indirectly in the formation of ROS, as a result, macrophages are activated and released large amounts of TNF-a [21]. Alveolar macrophages exposed to atmospheric particles increase both their phagocytic activity and increase their production of pro-inflammatory mediators such as TNF-a [22].

In this study, the Level of GPx significantly decreases in exposed workers $p<0.05$ with mean level (1273.0±423.08pg/ml) compared to control group mean levels (1527.3±373.57pg/ml), as shown in table 3.

GPx is an antioxidant that is responsible for protecting cells from damage due to free radicals like hydrogen and lipid peroxides [23]. Antioxidant enzymes are proteins involved in the catalytic transformation of reactive oxygen species(ROS) and their by-products into stable nontoxic molecules, therefore, representing the most important defense mechanism against oxidative stress-induced cell damage. Antioxidants in the lungs are the first line of protection against ROS. Individual responsiveness to air contaminants could be influenced by the composition and quantity of antioxidants in respiratory tract lining fluids [24]. After air pollutants entering the lung, faces first the extracellular antioxidant mechanisms present in the respiratory tract lining fluid, which contains enzymatic antioxidants such as GPx [25]. The result of this study might suggest that the decrease of GPx levels in refinery workers exposed to pollutants might be a part of the antioxidant status to protecting tissues from the effects of free radicals. The enzyme GPx plays an important role in regulating the level of different peroxides by accelerating the conversion of reduced glutathione (GSH) to oxidized glutathione (GSSG) after removing the different peroxides (such as hydrogen peroxide, lipid peroxides, and organic peroxides). GPx is part of the cell protection system against oxidative stress. And its products, thereby reducing cell damage caused by an increase in free radicals. Therefore, the exposure of the body to oxidative damage leads to a significant decrease in the level GPx. A rise in free radicals triggers an increase in the use of endogenous antioxidants, resulting in a decrease in endogenous antioxidant levels in the body [26].

MDA is one of the end-products of the peroxidation of membrane lipids caused by ROS formation. It is considered a good marker of oxidative stress [27]. In this study, the Level of MDA significantly increases P<0.01 in exposed workers with a mean level (1.34±1.02µmol/L) compared to control group mean levels (0.73±0.7µmol/L) as shown in table 3.

This study showed a highly significant difference among the exposed group compared to the control group regarding the level MDA which was higher among the exposed group. This is in agreement with other studies which illustrated that air pollutants exposure has been associated with an increase in the overall formation of MDA[28]. Elevated MDA level was observed in exposed workers study, indicating that lipid damages were induced in subjects occupationally exposed to air pollutions [29]. Excessive formation of free radicals increases the process of lipid peroxidation, as evidenced by elevated levels of MDA, the end product of lipid peroxidation, in serum and tissues of exposed subjects [30].

Conclusions

The exposure to air pollutants in refineries can cause markered alterations in hematological parameters and suggestive as useful tools to serve as a marker for biological control or monitoring of residents for the level of exposure to air pollutants. Inhalation of air pollutants can stimulate the immune system. Increase exposure to air pollutants can lead to oxidative stress for refinery workers. A decrease in antioxidant GPx in refinery workers' serum and an Increase in level MDA in refinery workers serum that considers good biomarkers to oxidative stress.

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