Estimation of lead in blood donors: Removal of lead associated with blood transfusion using carboxymethyl cellulose

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Abstract: Lead is well known for centuries for its toxic effects on adults and children through its different routes of exposure. In this study we gave our concern on lead levels of the blood donors to evaluate the possible lead exposure associated with blood transfusion. Twenty four blood donor samples were chosen randomly from Regional Centre for Blood Transfusion Services in Zagazig, Sharqia governorate, Egypt. The whole blood lead concentration was estimated for all samples by using graphite furnace atomic absorption spectroscopy (GFAAS). We have found that 90% of our blood donors had blood lead levels above 10 µg/dl, demonstrating high risk for lead poisoning in children from the blood transfusion process. Different natural filters were applied to remove the lead from blood. Among different materials used including three types of zeolites and natural products, carboxymethyl cellulose (CMC) was found to decrease the level of lead in blood significantly. It is recommended, that blood filtration using natural materials should be considered in blood banks to decrease the lead levels.

Keywords: Lead level, Lead poisoning, Blood donors, Blood transfusion, Carboxymethyl Cellulose.

INTRODUCTION

Lead is a toxic metal that causes significant public health problems adversely affecting multiple body systems, including neurological, hematological, gastrointestinal, cardiovascular and renal [1]. Children are much more sensitive than adults to the neurotoxic effects of lead even at low levels of exposure because the developing central nervous system (CNS) is more vulnerable to toxicants than the mature CNS. [2-3]. Today almost everyone is exposed to environmental lead by inhalation or ingestion through various sources like leaded gasoline, lead smelting and coal combustion, lead-based paints, lead containing pipes and battery recycling [4]. Recently two studies have shown that exposure of premature infants to lead could be also from blood transfusions [5-6].

Blood transfusion is a necessary process to save lives of many people, this why blood coming from different donors is screened for the diseases that are transmissible through transfusion such as Human Immunodeficiency, Hepatitis B, Hepatitis C viruses and Syphilis bacteria [7]. Many blood banks ignore physical examination of the blood donors and non infectious blood tests that is based on local medical problems which can be another cause of contaminated blood [8]. Laboratory investigations are the only reliable way to diagnose lead-exposed individuals since lead poisoned individuals do not show any or specific signs or symptoms [2-9]. The most common laboratory method available to determine blood lead concentrations is atomic absorption spectrometry (AAS) [10].

In Egypt, there is no national examination survey to provide data and estimate the prevalence of elevated blood lead levels however; auto exhaust and lead smelting industry are the major source of environmental lead exposure in greater Cairo and other Egyptian governorates [11].

The aim of the present study was to estimate lead level in random samples of blood donors for the assessment of lead poisoning risk that may result from blood transfusion process. We also aimed to eliminate lead from blood samples using different natural filters such as CMC and rice husk as well as synthesized mordenite, NaY, and ZSM-5 zeolites.

MATERIAL AND METHODS:
Samples collection

This study was conducted on 24 random samples of blood donors attending Regional Centre for Blood Transfusion Services in Zagazig, Sharqia governorate, Egypt. Fresh blood was drawn from each
donor involved in this study under complete aspect precautions. Informed written consent was obtained from each donor prior to blood collection.

**Laboratory investigations**

Five milliliters of blood were taken from each donor by venipuncture and divided into two tubes. The first tube containing ethylene diamine tetra acetic acid (EDTA) was used for the estimation of lead concentration and the different hematological parameters. The blood lead level was then determined by the graphite furnace atomic absorption spectroscopy (GFAAS, Perkin Elmer 4100 ZL) [12]. Complete blood picture analysis was measured using Sysmex automated hematology analyzer (Sysmex K X 21 N, Kobe, Japan). The second plain tube containing a clot activator was incubated at 37 °C for 30 minutes to allow clot formation, and then centrifuged for serum separation. The sample serum was used for routine biochemical kidney functions assays (diamond diagnostics, USA) including creatinine and blood urea nitrogen (BUN) using Robotic touch photometer (ROBONIK ECO PLUS, Mumbai, India).

**Preparation of filter materials**

One gram of poly vinyl alcohol (PVA) (Fluka, MW 125,000) dissolved in 100 ml distilled water was added to 0.1 g of carboxymethyl cellulose (CMC) (SD Fine chem. Limited, high viscosity 98%, India), dry raw rice husk (RH) after being thoroughly washed with distilled water, mordenite [13], zeolite NaY (Mobil Company Si/Al = 5.6, BET = 850 m²/g) or zeolite ZSM-5 [14] with heating and stirring until drying. After that, lead and hemoglobin levels were measured before and after filtration of blood. A selected blood sample referred to as # 23 was used to finally test the uptake of lead over the different prepared materials as mentioned before. Filtration with PVA was used as a negative control.

**Statistics**

Graphic presentation of the results and statistical analysis were performed using Prism software (GraphPad Software Inc., San Diego, CA, USA). Quantitative results were expressed as mean ± standard deviation (SD).

**RESULTS**

**Lead concentration of the blood donor samples**

Lead concentration was measured for 24 whole blood samples selected randomly from Regional Centre for Blood Transfusion Services. All the samples tested showed high levels of lead in the blood ranging from 7 to 60 µg/dL (Fig.1). The Results demonstrate that Lead levels of 11 samples are above 10 µg/dL with a mean value ± S.D equal to (17.5±1.78) while, lead levels in another 11 samples are above 20 µg/dL with a mean value ± S.D equal to (27.7±4.14). Only two out of the 24 tested samples have lead level below 10 µg/dL with a mean value ± S.D equal to (6.85±0.21) (Table 1).

![Fig-1: lead levels of the blood donor samples](image)

**Table 1: Percentage of studied samples with lower and higher lead values**

<table>
<thead>
<tr>
<th>Blood Samples</th>
<th>lead level &lt;10 µg/dl</th>
<th>lead level &gt;10 µg/dl</th>
<th>lead level &gt;20 µg/dl</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number</td>
<td>2</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>%</td>
<td>8.4</td>
<td>45.8</td>
<td>45.8</td>
</tr>
<tr>
<td>Mean ±SD</td>
<td>6.85±0.21</td>
<td>17.5±1.78</td>
<td>27.7±4.14</td>
</tr>
</tbody>
</table>

**Kidney function and hemoglobin levels of blood donors samples**

High levels of lead in blood were shown to be associated with diseases such as kidney failure and anemia. For that, creatinine, BUN and hemoglobin levels were tested to make sure that high lead levels present in almost all tested samples is not linked with such diseases which normally prevent blood donation. The mean results of creatinine, BUN and hemoglobin for all samples lie in the normal ranges demonstrating that the blood donors were not linked to diseases such renal failure of anemia (Table 2). However, only sample number one with lead concentration 60 µg/dL show moderate anemia with hemoglobin concentration equal to 8.7 g/dL.
Removals of lead in blood by filtration

The presence of high lead levels in the blood donor samples causes lead poisoning if blood is transferred especially to the children. Therefore, we have tried to remove lead ions from the blood sample #23 using different filters. The materials used such as mordenite, zeolite NaY, zeolite ZSM-5, and rice husk showed no effect on the removal of lead from the tested blood sample when compared to the negative control polyvinyl chloride (PVA). On the contrary, a significant decrease in the lead level was achieved when CMC was used for filtration giving 60% reduction in lead concentration, i.e. from 23 to 14 μg/dl (Figure 2). The Hemoglobin level in the filtered blood sample was in normal range for all used materials.

DISCUSSION

This study was designed to estimate lead levels of random blood donors attended Regional Centre for Blood Transfusion Services to explore the possible effects of these levels on the blood transfusion process especially in countries suffering from environmental lead pollution like Egypt. The results of this study showed that 90% of the samples having lead levels greater than 10 µg/dL of them 45% with lead levels greater than 20 µg/dL. The mean blood lead level of all samples tested in this study was 17 µg/dL, while in Los Angeles USA, Morocco, and Brescia Italy blood banks were 1, 8.7, and 14.8 µg/dL, respectively [15-16]. Studies done in different Egyptian governorate have shown that the control of lead is not efficient, so that the level of lead in drinking water, air, or soil in some sporadic areas is still high [17-18-19].

In recent years, the focus in lead poisoning has shifted away from adults exposed to high doses in industrial settings to low exposure doses from different sources with adverse health effects on adults and specially children. Center for Disease Control and prevention (CDC) has revised its limit of acceptable blood lead level from 60 µg/dL to 10 µg/dL based on neurological toxicity studies [20], however lead level less than 10 µg/dl cannot be considered safe [21]. The presence of anemia [4], cognitive deficits [22], intellectual impairments [23], or disturbances in fine motor function [24] were demonstrated in children with blood lead levels below 10 µg/dL. The blood lead concentrations measured in this study are significantly higher than those measured in the previous studies, indicates that children are at extreme risk to lead poisoning from blood transfusion.

Lead exposure from blood transfusions in premature infants was reported by Bearer et al., where it was found that increases in post-transfusion blood lead concentration were linear with doses higher than 1.5 µg/dL of donor blood [5]. Recently, Gehrie et al also support that transfusion of packed red blood cells is a source of lead exposure and even show the possible release of lead in plasma from erythrocytes after few minutes of storage [6].

Lead has long been known to alter hematological system [25] and kidney function [26] therefore we have tested the level of hemoglobin, creatinine, and BUN to stand on the health status of those blood donors. Our results show that those blood donors did not have any diseases related to lead toxicity that could prevent them from blood donation. Until now laboratory tests in our blood banks which ensure safety

### Table 2: Mean values of creatinine, BUN and hemoglobin of the studied groups

<table>
<thead>
<tr>
<th>Blood Samples</th>
<th>Creatinine (mg/dl)</th>
<th>BUN (mg/dl)</th>
<th>Hb (g/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean ±SD</td>
<td>0.75±0.21</td>
<td>26±3.8</td>
<td>13.7±1.3</td>
</tr>
</tbody>
</table>

Fig-2: Removal of Lead using different filters
from the diseases that are transmissible through transfusion do not include blood lead testing. Additionally, Clinical symptoms of lead poisoning are rarely seen below 60 μg/dl [2], for these reasons blood donated by persons exposed to lead will end up in our country's blood banks.

The exclusion of blood samples with lead level above the normal average is the only consideration that has been taken until now to prevent blood lead exposure [6]. However, to date no good man-made substitute for human blood is present, besides blood is very difficult to obtain and expensive to analyze. Blood exclusion cannot be applicable in our case because almost all our blood samples contain high lead level. For that, different prepared natural filters were used as a new method to remove lead in blood samples. It was found that blood filtration using rice husk or different zeolites (mordenite, NaY & ZSM-5) have no effect on lead in blood. But when comparing between polyvinyl alcohol (PVA) used as a negative control and CMC we have found a significant decrease of blood lead level from 23 to 14 μg/dL. This means that the removal of lead in blood depends on the chelating-sites binding-lead. CMC, a typical derivative of cellulose, is an anionic polyelectrolyte soluble to water. Our recent study showed that CMC possesses adsorption bands at 1622 and 1432 cm⁻¹ assigned to ν(COΟ) and ν(COΟ) carboxylate vibration stretching, respectively [27]. Besides, peaks at 3410, 3356 and 3316 cm⁻¹ were allocated to free OH stretching vibration and inter- and intra-molecular hydrogen bonds, respectively, in CMC matrix. These function groups served as chelating sites for lead in blood. Since the hemoglobin level was not altered by filtration, CMC might be administered easily, affordable and safe for blood.

CONCLUSION
This study contributes in giving information on lead levels among blood donors. Based on our analysis, lead concentration was above accepted levels in almost all samples tested. Also, for the first time, we succeeded to reduce lead levels in the blood using natural CMC filter which may be useful in preventing lead exposure from blood transfusion. Lead test should be considered in our national blood banks and in countries that suffer from environmental lead exposure.

REFERENCES
11. Rizk FSH, Khoder MIM; Decreased Lead Concentration in Cairo Atmosphere Due to Use of Unleaded Gasoline. CEJOEM, 2001; 7(1): 53-59.


