Determination of Blood Lead Levels in Adult Bahraini Citizens Prior to the Introduction of Unleaded Gasoline and the Possible Effect of Elevated Blood Lead Levels on the Serum Immunoglobulin IgG

Afnan Mahmood Freije, PhD* Maheen Ghuloom Dairi, PhD*

Objective: This study is designed to investigate the possible effect of environmental lead exposure on the immune system.

Design: Prospective study.

Setting: University of Bahrain, College of Science, Department of Biology, Isa Town Campus.

Method: The concentration of lead (Pb) and the serum immunoglobulin IgG were measured in the blood of 40 human adults by atomic absorption spectrophotometry and ELISA assay in April 2000, soon before the implementation of unleaded gasoline (July, 2000) in the Kingdom of Bahrain.

Result: The blood lead levels of participants were found to be elevated (mean 15.3±5.7 µg/dl), in comparison with the standard international levels, in which blood lead levels of >10 µg/dl are designated as lead poisoning. On the other hand, the serum immunoglobulin IgG was within the normal range (mean 1007.1±147.1 mg/dl). However, no correlation was found between the blood lead levels and the serum IgG levels (p = 0.14).

Conclusion: This study has shown that blood lead levels in 29 (72.5%) of the participations were higher than the internationally accepted level. The study also suggests that elevated blood lead levels did not have an effect on the efficiency of the humoral immunity represented by serum IgG.

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Lead (Pb), an occupational and environmental toxicant; it is an air pollutant generated from human activities such as the industrial burning of fossil fuels. Lead exposure occurs mostly from breathing air or dust, eating contaminated food, drinking contaminated water and ingesting lead from lead-based paint especially by children[1-5]. Relatively, lead from automobile exhaust is considered the major source for environmental lead in many countries including Bahrain[2,6]. However, a growing number of countries have adopted a successful transition to unleaded gasoline, including Bahrain which started its use in July 2000[7]. In Bahrain, tetraethyl lead was added to gasoline at a concentration of 0.84 g/l for Mumtaz (Premium) and 0.53 g/l for Jayyid (Regular)[6].

The definition of lead poisoning (i.e. ≥10 µg/dl) adopted by many international institutes including the CDC (Centers for Disease Control and Prevention, USA)

* Assistant Professor
Department of Biology
College of Science
University of Bahrain
Kingdom of Bahrain

was lowered to a blood lead level of 10 µg/dl in 1991 due to the rapid decline in environmental and occupational lead exposure in most developed countries[8]. Therefore, blood lead level <10 µg/dl is now universally accepted as the low-lead exposure threshold[9,10].

Absorbed lead enters the bloodstream and then travels to various organs and tissues. More than 99% of lead in blood is bound to erythrocytes, whereas more than 95% of absorbed lead is stored predominantly in bones[4,11].

Lead was shown to affect a number of systems and organs, including the central nervous system (the most sensitive), kidneys and the immune system. Lead exposure results in an increased frequency of infection and increased susceptibility to bacteria[12]. However, the effect of lead on serum immunoglobulin levels remains highly controversial[13,14].

Lead poisoning affects the health of humans, as well as cognitive and behavioral development, especially during childhood. Several studies have shown that lead poisoning in children is associated with irreversible decline of intelligence, behavior, neuromotor problems and poor school performance[2,10].

This study is designed to investigate the possible effect of environmental lead exposure on the immune system.

METHOD

Venous blood samples (10 ml) were obtained from 40 adults, ages between 19 and 48 years by a trained phlebotomist using venepuncture methods. The group included 31 women and 9 men with an age range of 19-41 and 20-48 years old respectively. This group volunteered to participate in the present study in response to an announcement which was distributed all over the University of Bahrain during the period of study, without any preference for sex, residential area, etc. by the authors.

Whole blood samples collected in heparinized tubes (5ml) were used directly for lead measurement whereas blood samples (5ml) collected in plain tubes, containing no anti-coagulating factors such as heparin, were centrifuged to obtain the serum and stored at -20^{o}C until IgG analysis.

Whole blood samples were broken up immediately after collection with 16N nitric acid and measurements were made using a graphite-furnace atomic absorption spectrophotometer according to the method of Hernandez-Avila et al (1998)[15]. Standard samples of known lead content (5, 10, 20, and 30µg/dl) were included in each run, as well as blank samples.

Human serum IgG was isolated from samples by DEAE Sephacel column chromatography (2cm x 10cm) and measured by an ELISA double-antibody sandwich method, following the procedure of Kan et al (1983)[16]. Five standard solutions were prepared and used with the ELISA.

Statistical analysis was done using Excel for Windows (version 1997) in order to calculate the Pearson correlation coefficient and to evaluate the degree of association of lead blood levels and serum IgG levels.

RESULT

The blood lead levels and the serum IgG levels of the participants are summarized in Table 1 and 2. The mean blood lead level was 15.3±5.7 µg/dl, ranging from 5 to 28 µg/dl, only two persons had levels greater than 25 µg/dl. The blood lead levels of 29 samples (72.5%) of which 7 (17.5%) were males and 22 (55.5%) were females. Their blood lead level was higher than 10 µg/dl, which is the internationally agreed diagnostic criterion of lead poisoning in adults. Although blood lead levels were elevated in two male smokers (21-22 µg/dl), the number of smokers was too small (5%) to be taken into consideration. Serum concentrations of IgG ranged from 745.7 to 1301.4 mg/dl, with a mean level of 1007.1±147.1 mg/dl. No correlation was observed between the blood lead levels and the serum IgG levels (p = 0.14).

Table 1: Blood Lead Levels in Relation to Age and Sex

No of individuals	Age (years)	No of males	Males %	No of females	Females %	Lead in blood µg/dl in males	Lead in blood µg/dl in females
34	19-25	7	17.5(5*)	28	70	12-22	5-28
0	26-30	-	-	-	-	-	-
0	31-35	-	-	-	-	-	-
4	36-40	1	2.5	2	5	11	16-26
1	41-45	-	-	1	2.5	6	-
1	46-50	1	2.5	-	-	13	-

Total % of blood lead level >10 µg/dl = 72.5 %
Total % of males with blood lead level >10 µg/dl = 17.5 %
Total % of females with blood lead level >10 µg/dl = 55.5 %
Total % of males with blood lead level ≤10 µg/dl = 0 %
Total % of females with blood lead level ≤10 µg/dl = 22.5 %
Total % of non-smoking individuals = 95%
Total % of smoking individuals (2 males) = 5 %
n = 40

Table 2: Blood Lead Levels in Relation to Area of Residence

Area	No. of individuals	Mean lead level µg/dl± SD	Range µg/dl
Manama & Capital Area	7	13.43±7.81	5-26
Muharraq	1	16±0	
Isa Town	6	15.17±4.71	10-22
Hamad Town	7	16.71±6.99	10-28
Riffa	2	10.5±0.71	11-10
Sitra and Central Area	6	11.83±3.6	12-18
Northern Area	11	18.55±3.8	12-23

Table 2 summarizes the lead level in relation to area of residence. No significant difference was observed between various areas of residence and blood lead levels (p = 0.22).

DISCUSSION

In most developed countries, a recent decline in environmental and occupational lead exposure is mainly due to the removal of lead from gasoline. It should be noted that communities living near lead, copper, zinc and battery recycling smelters have been reported to be severely poisoned by these metals from emissions of dust and particulates[10]. The control of lead exposure in the workplaces in Bahrain, lead levels in smelters or refineries' workers, if ever conducted, were never published and always considered as company private and privileged information. On the other hand, communities located near industrial areas have never been screened. However, preventive measures have been adopted in July 2000, by removing lead from gasoline[7].

Depending on the degree of lead exposure, the health consequences could range from undetectable effects to irreversible decline of intelligence, behaviour and poor school performance among children[14,17]. Several studies on children have revealed the loss of one to two IQ points when blood lead level was ranging between 10-20 µg/dl or 5-10 µg/g range in tooth lead level[18,19]. Blood lead level as low as 5 µg/dl in adults causes an increase of blood pressure, especially systolic[20,21]. Despite inadequate evidence for lead carcinogenicity in human, lead has been classified as a possible human carcinogen (Group 2B) by the International Agency for Research on Cancer (IARC) based on sufficient evidence that lead is carcinogenic in experimental animals[22]. Furthermore, the study of Vaglenov et al showed a clear genotoxic dose-related effect to the lead levels in blood associated with occupational lead exposure[5].

Plasma is the component of blood through which lead is free to cross the cell membrane and cause organ toxicity. However, the predominant endogenous storage site for lead is the skeleton, in which more than 95% of the total amount of lead is stored. Therefore, blood lead levels serve as an indicator of recent environmental exposure but not long-term exposure[4,11].

In Bahrain, previous studies on umbilical cord blood levels of newborns has shown high lead levels (9.34±3.86 µg/dl), with 57% of the babies having $\geq$ 10 µg/dl, 7% $\geq$15 µg/dl, and 2% $\geq$20 µg/dl, in comparison to nearby regions in which unleaded gasoline is used, such as Saudi Arabia (4.14 µg/dl)[23]. These values were close to levels in the USA in the early seventies and Australia in the early eighties[23]. Lead levels in deciduous teeth of children of Bahrain also showed toxic concentrations in 35% of those studied[24]. Madany et al has also reported high lead levels in some local fish and shellfish in Bahrain, and high lead concentrations in ambient air[25]. Elevated air and blood lead levels, with a definite correlation between the lead in air concentration and the volume of traffic, were also reported in Bahrain by Vreeland and Ekarath in 1990[6]. In their study, blood lead levels in 77 adults ranged between 5-36.6 µg/dl (mean 15.74±5.84 µg/dl), and high concentrations of lead in air were recorded in sites characterized by high traffic density (average of lead in the air 0.834 and 1.04 µg m^{-3}). Lead was also measured in one of the most consumed fish species in Bahrain (Grouper fish *Epinephelus. coioides*) in 1996, in which lead levels (mean 10.8 µg.g^{-1}, range 4.3-15.2 µg.g^{-1}) were higher than fish lead limits adopted world wide (0.5-6 µg.g^{-1})[26]. In 1997, Akhter and Al-Jowder measured lead in the sediments of 19 coastal stations in Bahrain[27]. They also reported high levels of lead (111 mg/kg) which was attributed to pollution from land-based industrial and urban sources, namely automobiles that contribute to the overall pollution in the coastal area.

The results of the present study have shown that the mean blood lead level in Bahrain was (15.3±5.7 µg/dl) in April 2000. In Bahrain, lead from automobile exhausts is considered the main source of lead exposure[6]. Other sources such as water, food and paint were not investigated in the present study. However, lead water pipes are no longer in use, except possibly in very old houses[6]. The association of cigarette smoking with elevated blood lead levels is well documented[28]. However, the small number of smokers in the present study did not allow us to draw a definite conclusion about the correlation between high blood lead levels and cigarette smoking. There was no correlation between the area of residence and blood lead levels in the present study, most probably due to the small number of samples. However, a study by Vreeland and Ekarath in 1990[6] showed a significant correlation between vehicular traffic density and lead concentration in air and blood samples taken in Bahrain. They reported an almost twofold (182%) increase of lead in air between the Budaiya area (rural) and Manama city which correlated with an increase in blood lead levels of 28.5-30.4%.

Blood lead levels in adults in Bahrain showed no significant differences between the years 1990 and 2000, most probably due to the use of leaded gasoline. In USA, the blood lead levels dropped from 15.8 µg/dl in 1976 to 9.2 µg/dl in 1980, then there was a further drop to 5.94 µg/dl in 1991 and finally to 2 µg/dl in 1999[29]. However, in Korea blood lead levels persisted high (32 µg/dl) during the years 1997-1999, which were considered normal in China (<40 µg/dl) in 1998[30,31]. The blood lead level in Bahrain is comparable to the lead level in the USA during the mid 1970s. The decline in the concentration of blood lead level from 15.8 µg/dl in 1976 to 2 µg/dl in 1999 in the USA was associated with the introduction of unleaded gasoline and the vehicle emission regulation during 1976[29].

The mean serum IgG in the present study was 1007.1±147.1 mg/dl and ranged from 745.7 to 1301.4 mg/dl. Serum IgG levels were within the normal range (751-1560 mg/dl), and (650-1640 mg/dl for male age 20-50 and 690-1700 mg/dl for females age 20-50)[32,33]. No significant correlation between blood lead levels and serum IgG concentrations was observed in the present study. Environmental pollutants such as lead could potentially cause adverse health effects through an interaction with the immune system in several ways. Lead has been reported to be an international immuno-toxic agent[34,35]. However, conflicting results on the effects of lead on the immune system in humans have been reported in several studies[36,37]. Some reports have shown suppressive effects on some immunological parameters such as T-helper cells, serum immunoglobulins (IgG, IgM, IgA), C3 and C4 complement protein levels in workers with high lead exposures[34,38,39]. On the other hand, some reports showed no significant changes[40].

The findings of this study provide important and useful information, particularly if such a study is repeated in order to determine the effect of using unleaded gasoline and, therefore, establishing a new blood lead level in Bahrain. Such an investigation is underway by the authors on a larger group sample (200 adults).

CONCLUSION

The result of this study revealed that blood lead level in 72.5% of the participants was higher than the internationally accepted level, most probably due to the use of leaded gasoline in automobiles. These findings also suggest that elevated blood lead levels did not have an effect on the efficiency of the humoral immunity represented by serum IgG. However, further investigations are

required in order to reveal the possible relationship between elevated blood lead levels and the immune system.

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