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Blood lead level and correlation with pregnancy-associated anaemia

Jehan Hamadneh[1], Adnan Massadeh[2], Shereen Hamadneh[3], Manal Kassab[4], Nahla S. Al-bayyari[5], Yousef Khader[6], and Wafa Sbuihat[7]

1. Department of Obstetrics and Gynecology, Faculty of Medicine, Jordan University of Science and Technology, Irbid, Jordan
2. Department of Medicinal Chemistry and Pharmacognosy, Faculty of Pharmacy, Jordan University of Science and Technology, Irbid, Jordan
3. Department of Maternal & Child Health, Faculty of Nursing, Al-albayt University, Mafraq, Jordan
4. Department of Maternal & Child Health, Faculty of Nursing, Jordan University of Science and Technology, Jordan
5. Department of Nutrition and Food Technology, Al-Huson University College, Al-Balqa Applied University, Al-Salt, Jordan
6. Department of Community Medicine, Public Health and Family Medicine, Faculty of Medicine, Jordan University of Science and Technology, Jordan University of Science and Technology, Irbid, Jordan
7. Department of Applied Biological Sciences, Faculty of Paramedical Science, Jordan University of Science and Technology, Irbid, Jordan

RESEARCH

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Corresponding Author:
Jehan M. Hamadneh
Department of Obstetrics and Gynecology
Faculty of Medicine, Jordan University of Science and Technology, Irbid, P.O Box: 3030, Jordan.
Tel: +962-799166694
Email: jehan_Hamadneh@yahoo.com; Jmhamadneh@just.edu.jo

ABSTRACT

Background
The most common reason that leads to anaemia is related to the reduction in certain trace elements such as Fe. However, it has been found that an elevation in some other heavy metals such as Pb could also lead to anaemia.

Aims
This research aims to assess the Correlation between Pb blood levels and Fe, Haemoglobin levels during pregnancy among Jordanian women.

Methods
A cross-sectional study was conducted in the Northern area of Jordan. Venous blood samples collected from 167 pregnant women for the determination of haemoglobin (Hb), Fe and Pb levels of which 17 in the first trimester, 19 in second trimesters, 131 in third trimesters. Women were classified into two groups, the first group included cases with blood Pb Levels ≥10µg/dL (high blood Pb level group, n=118) and the second with blood Lead levels less than 10µg/dL (low blood Lead level group, n=49).

Results
About 71.4 per cent of women had anaemia (n=120; Hb<10.5g/dl) and 70.7 per cent of women had a high blood Pb level of ≥10µg/dL). Pb blood levels ranged from 6.45 to 28.0µg/dL. The mean (SD) of blood Pb level was 12.1 (4.1) µg/dL. The mean haemoglobin and Fe levels did not differ significantly between women with low and high levels of Pb. Blood Pb levels were not significantly correlated with haemoglobin levels (r=-0.025; P=0.747) nor with iron levels (r=0.099; P=0.241). After adjusting for important variables, Pb was not significantly associated with haemoglobin (P=0.223) and with iron (P=0.116).

Conclusion
The level of Pb in the blood of pregnant women has no any association with haemoglobin and Fe levels during pregnancy.

Key Words
Lead, haemoglobin, iron

What this study adds:

1. What is known about this subject?
Studies show that Fe doesn't necessarily associate with a change in Pb blood levels among older female children or adolescents.

2. What new information is offered in this study?
Study findings show that Pb blood levels are higher among women with income less than 600 JD, women in the third trimester, and among women who were using multivitamins.

3. What are the implications for research, policy, or practice?
The level of Pb in the blood of pregnant women has no any association with haemoglobin and Fe levels during pregnancy. Further studies are needed to elucidate this relationship.

Background

Iron deficiency anaemia is the most common cause of anaemia in pregnancy[1] the prevalence of Fe-deficiency anaemia is around 46–66 per cent in developing countries.[2] The most common reason that leads to anaemia is related to the reduction in certain trace elements such as Fe. However, it has been found that an elevation in some other heavy metals such as Pb could also lead to anaemia.[3-7]

Human exposure to Pb occurs through diet, air, drinking water where Pb pipes are being used, ingestion of Pb products,[4] and exposure to old leaded paint chips or pigments of lead-glazed pottery which is known as PICA in pregnancy.[8]

A previous study reported that blood Pb levels ≥10µg/dL is significantly correlated with anaemia, decreasing Fe absorption and negatively affecting hematologic indices.[3,9-11] The explained mechanism of lead-induced anaemia is related to the fact that by increasing Pb level in the blood, the Heme-formation, which is essential Fe metabolic step, will be interfered and this, in turn, will increase the rate of red blood cell destruction and blocking iron absorption.[12] Moreover, it was reported that insufficient Fe might amplify Pb contamination by increasing retention and absorption of Pb in the body,[9] which in turn affects an important Fe dependent metabolic step in Heme-biosynthesis.[12]

The presence of Pb prevents the formation of haemoglobin which causes Pb-induced anaemia (hypochromic microcytic anaemia).[9] Hypochromic microcytic anaemia can result from a significant reduction in Fe level even if no Pb is present, but the presence of Pb causes deterioration of anaemia.

Severe anaemia, typically less than seven can be associated with significant maternal and foetal complications.[11] Anaemia during pregnancy has negative health impact on both maternal and foetus outcomes, such as premature delivery,[2] and intrauterine growth retardation.[2] According to WHO, the lower limits of normal haemoglobin level are 12g/dL in women and 14g/dL in men and normal haemoglobin level up to 11g/dL percent in pregnancy.[8] So any haemoglobin concentration less than 11g/dL during pregnancy by WHO standards should be considered anaemia. In developing countries pregnant women with Hb level <10.5g/dL still tolerate pregnancy, labour and delivery without negative outcomes, so pregnant women with (Hb<10.5g/dL) was assumed anaemic.[8]

The difficulty in establishing a precise diagnosis of lead-induced anaemia status of pregnant women represents a complicating factor in the understanding of the relationship between maternal Fe level and trace elements level such as lead blood levels. This study aimed to determine the correlation of blood Pb levels with Fe levels and haemoglobin levels during pregnancy among Jordanian women.

Method

A cross-sectional study was conducted to determine the correlation of blood lead levels with iron levels and haemoglobin levels during pregnancy in a group of pregnant women attending a teaching hospital in the north of Jordan. Women were randomly selected from the patients' care records irrespective of the trimester of the pregnancy. All included pregnant women were non-alcoholic and no PICA cases. Women with any disease that may affect Pb blood levels (gestational hypertension, gestational diabetes and those who receive blood transfusion) were not included in the study. A total number of 167 pregnant women met the inclusion criteria and were included in this study.

Data were collected using a questionnaire. The first part of the questionnaire included the socio-demographic and relevant characteristics of the participants such as age, education, income, parity, trimester of pregnancy, daily intake of Fe and multivitamins.

A trained researcher collected an aliquot of 3mls of venous blood samples from pregnant women during their visits to the maternal clinics. Blood was transferred into EDTA tube immediately centrifuged (4500 RCF for 10 min) to separate the plasma from whole blood to prevent the transfer of Pb from erythrocytes. Pb in blood was measured by taking 0.5g of Ammonium Di-hydrogen Phosphate ($NH_4H_2PO_4$) which was mixed with 2mls Triton X100 in a 100mL glass bottle at the room temperature. Graphite Furnace Atomic Absorption Spectrometry (GFAAS) (also known as Electrothermal Atomic Absorption Spectroscopy (ETAAS) was used to determine the concentration of Pb at wavelengths of 283.3nm. GFAAS is a type of spectrometry that uses a graphite-coated furnace to vaporize the sample. Briefly, the technique is based on the fact that free atoms will absorb light at frequencies or wavelengths characteristic of the element of interest (hence the name atomic absorption spectrometry).

The digestion of samples was done by mixing of 0.1mL of a mixture with 0.05 mL of sample, whereas to determine Fe concentrations samples were diluted 1:3 with deionized water. Flame Atomic Absorption Spectrometer (FAAS) was used to determine the concentration of Fe at wavelengths of 248.3nm with a slit width of 0.5 nm. Both Flame and Graphite manufactured by Unicam Atomic Absorption, United Kingdom, model (SOLAAR M5).

Pregnant women were divided into two groups using the CDC cut off point of 10µg/d [11,12,13] High blood Pb Levels ≥10µg/dL (n=118) and 2) low Pb levels <10 µg/dL(n=49).

Data were analysed using the Statistical Package for Social Sciences (SPSS) version 20. Pearson correlation was used to correlate between blood Pb level, Fe, and Haemoglobin. The differences between means were analyzed using independent t-test. Multiple Linear Regression analysis was used to analyse the association between Pb (independent variable) and other dependent variables including Haemoglobin and Fe. For statistical analysis, a *p*-value of <0.05 was considered statistically significant.

Results

Blood samples were taken from 167 pregnant women (17 (10.2 per cent) in the first trimester, 19 (11.4 per cent) in second trimesters, and 131 (78.4 per cent) in third trimesters). A total of 92 (55.8 per cent) women aged less than 30 years and 93 (57.8 per cent) had a bachelor degree or higher degrees (Table 1). About 71.4 per cent of women had anaemia (n=120; Hb<10.5g/dl), and 70.7 per cent of women had a high blood Pb level of ≥10µg/dL. Pb blood levels ranged from 6.45 to 28.0µg/dL. The **(**mean±SD) of blood Pb level was 12.1 (4.1)µg/dL.

Table 2 shows the mean (±SD) of haemoglobin, Pb, and Fe levels and the analysis of the differences according to the studied characteristics using independent t-test. The mean of blood Pb level differed significantly according to income, trimester, and multivitamin use, being higher among women with income less than 600 JD, women in the third trimester, and among women who were using multivitamins. The mean of blood Haemoglobin level differed significantly according to income, trimester, and multivitamin use, being higher among women with income less than 600 JD, and women in the first trimester (Table 2).

Table 3 shows the means of haemoglobin and iron among women with low and high levels of Pb. Using the independent test, the mean haemoglobin and Fe levels did not differ significantly between women with low and high levels of Pb. Blood Pb levels were not significantly correlated with haemoglobin levels (r=-0.025; P=0.747) nor with Fe levels (r=0.099; P=0.241).

Table 4 shows the multiple linear regression of the association between Pb and other parameters including haemoglobin, and Fe. After adjusting for important variables, Pb was not significantly associated with Haemoglobin (P=0.223) and with Fe (P=0.116). Haemoglobin level was correlated significantly with gestational age (*P*<0.005) (Table 4).

Discussion

Pb exposure remains a public health problem among childbearing age women and their developing foetus. High Pb level during pregnancy can adversely affect maternal, and foetus health.[12,14] Previous Pb exposures during other life stages will be stored, and then bone Pb stores are mobilized during periods of pregnancy and breastfeeding stage.[15,16] This, in turn, can result in lasting adverse health effects on both the mothers and their infants.[15,16]

This study aimed to determine the correlation of blood lead levels with iron levels and haemoglobin levels during pregnancy among a sample of Jordanian women. Study findings show that Pb blood levels are higher among women with income less than 600 JD, women in the third trimester, and among women who were using multivitamins. The association between high Pb blood levels among low-incomes mothers could be related to the fact that low-income pregnant women could hardly maintain a healthy lifestyle that includes regular physical activity and having

the adequate nutrition that increases heavy-metal excretion.[17-20] The reason for increasing the level of Pb during third trimester is related to the fact that Pb stored in the skeleton released and mobilized during pregnancy especially the third trimester. Another study finding supports this finding.[21] Pregnant women on multivitamins show a high Pb concentration because vitamins supplementation is one of many other materials that are containing Pb.[22]

Although Pb was found to be high in most of the pregnant women in the current study, haemoglobin level doesn't correlate with blood Pb levels. Our finding is consistent with other studies.[23,24] However, findings show that there is a decrease in haemoglobin content with increasing blood Pb levels.[25-28] These differences in the results could be related to the difference in sample size and in blood lead levels recorded in these studies. Also, the differences in age of other study participants' which included children could cause the differences in results.

The noticeably high blood Pb levels which were present in the participants of the current study could be due to high contamination from environmental pollution and the past lead exposure that may be mobilized during pregnancy from endogenous bone Pb stores that result in elevation in maternal blood levels. It has been reported that during pregnancy the increased blood Pb is either related to endogenous (bone saved), bone Pb stores are mobilized during periods of pregnancy, or from previous lead pollution.[33]

Anaemia is a health problem among pregnant women, which is associated with a decrease in some trace elements as Fe and an increase in heavy metals as lead. Concerning Fe levels in pregnant women and its relation with blood Pb level, our findings using Pearson correlation, showed no significant correlation between Pb levels and Fe levels ($r=0.099$, $P=0.241$). This indicates that Fe does not necessarily associate with a change in Pb blood levels as some previous studies reported.[8;29,31-33]

Our study finding is similar to many other findings that found no association between Fe deficiency and increased blood Pb concentrations.[34-37] However, a study conducted on young children showed an association between Fe levels and blood Pb levels after taking iron supplementation.[35-39] Furthermore, an association between Fe supplementation and blood Pb levels in infants with very low blood lead concentrations have been observed.[40] It is possible that the difference in study findings is related to differences in the age distribution of the study subjects, the assumptions used, or the degrees of Pb exposure. Studies show that Fe doesn't necessarily associate with a change in Pb blood levels among older female children or adolescents.[38,40]

However, and similar conclusion to our findings was reached among adolescent women owing to the overshadowing effects of oestrogen on Pb levels.[34,37] Also, it has been concluded that pregnant women have lower blood Pb concentrations than men and there is no association between high blood Pb levels and Fe deficiency as oestrogen promotes bone mineralization and redistributes blood Pb into bone in women.[35,38]

Conclusion

Previous research findings showed an inverse correlation between blood Pb levels with Fe and haemoglobin levels during pregnancy. However, our finding showed that the level of Pb in the blood of pregnant women has no any association with haemoglobin and Fe levels during pregnancy. This finding is inconsistent with other research results which showed that blood Pb level is negatively affected the haematological parameters which may eventually lead to Fe deficiency anaemia during pregnancy. In conclusion, blood Fe level can be well correlated with haemoglobin concentration. The decrease in blood Fe level leads to the decrease in haemoglobin concentration. However, haemoglobin and Fe levels did not differ significantly between women with low and high levels of Pb which indicates that the level of Pb in the blood of pregnant women has no any association with haemoglobin and Fe levels. Further studies are needed to elucidate this relationship.

References

1. Ahmad N, Kalakoti P, Bano R et al. The prevalence of anaemia and associated factors in pregnant women in a rural Indian community. Australasian Medical J. 2010;3(5):276–280.
2. WHO guideline: Calcium supplementation in pregnant women. Geneva, World Health Organization, 2012.
3. Scholl T. Maternal iron Status: Relation to fetal growth, length of gestation and the neonate's iron endowment. Nutr Rev. 2011;69(Suppl 1):S23–S29.
4. Hegazy AA, Zaher MM, Abd El-hafez MA, et al. Relation between anemia and blood levels of lead, copper, zinc and iron among children. BMC Research Notes. 2010;3:133–141.
5. European Food Safety Authority (EFSA) Panel on Contaminants in the Food Chain (CONTAM). Scientific Opinion on Lead in Food. EFSA J. 2010;8:1570–1577.

6. Soylak MS, Sarac M, Mendil D. Determination of trace metals in mushroom samples from Kayseri, Turkey. Food Chemistry. 2005;92:649–452.
7. Volesky B, Holan ZR. Biosorption of heavy metals. Biotechnol. Prog. 1995;11(30):235–250.
8. Alomary A, Al-Momani IF, Massadeh AM. Lead and cadmium in human teeth from Jordan by atomic absorption spectrometry: Some factors influencing their concentration. Science of the Total Environment. 2006;369:69–75.
9. Gulson B, Mizon K, Korsch M, et al. Low Blood Lead Levels Do Not Appear to Be Further Reduced by Dietary Supplements. Environ Health Perspect. 2006;114(8):1186–1192.
10. Centers for Disease Control Prevention (CDC). (2007). CDC's Advisory Committee on Childhood Lead Poisoning Prevention Interpreting and managing blood lead levels <10microg/dL in children and reducing childhood exposures to lead: recommendations of CDC's Advisory Committee on Childhood Lead Poisoning Prevention. MMWR Recomm. 56:1–16.
11. Melku M, Addis Z, Alem M, et al. Prevalence and Predictors of Maternal Anemia during Pregnancy in Gondar, Northwest Ethiopia: An Institutional Based Cross-Sectional Study Anemia. 2014;2014:108–593.
12. Wells EM, Navas-Acien A, Herbstman JB, et al. Low-level lead exposure and elevations in blood pressure during pregnancy. Environ Health Perspect. 2011;119(5):664–669.
13. Shah F, Kazi TG, Afridi HI, et al. Temperature controlled ionic liquid-dispersive liquid phase microextraction for determination of trace lead level in blood samples prior to analysis by flame atomic absorption spectrometry with multivariate optimization. Microchemical J. 2012;101(14):5–10.
14. Ettinger AS. Wengrovitz AG. Guidelines for the identification and management of lead exposure in pregnant and lactating women. U.S. Department of Health and Human Services Atlanta, GA. 2010.
15. Grandjean P, Bellinger D, Bergman A, et al. The faroes statement: Human health effects of developmental exposure to chemicals in our environment. Basic Clin Pharmacol Toxicol. 2008;102:73–5.
16. Bellinger DC, Teratogen update: Lead and pregnancy. Birth Defects Res A Clin Mol Teratol. 2005;73(6):409–20.
17. Anderson K, Nisenblat V, Norman R. Lifestyle factors in people seeking infertility treatment - A review. Aust N Z J Obstet Gynaecol. 2010;50(1):8–20.
18. Genuis SJ, Birkholz D, Rodushkin I, et al. Blood, urine, and sweat (BUS) study: monitoring and elimination of bioaccumulated toxic elements. Arch Environ Contam Toxicol. 2011;61(2):344–57.
19. Sears ME, Kerr KJ, Bray RI Z. Arsenic, cadmium, lead, and mercury in sweat: a systematic review. J Environ Public Health. 2012:184745.
20. Hertz-Picciotto I, Schramm M, Watt-Morse M, et al. Patterns and determinants of blood lead during pregnancy. Am J Epidemiol. 2010;152(9):829–837.
21. Klitzman S, Sharma A, Nicaj L. Lead poisoning among pregnant women in New York City: Risk factors and screening practices. J Urban Health. 2012;79(2):225–237.
22. Jong WC, Soon KK. Relationships of lead, copper, zinc, and cadmium levels versus hematopoiesis and iron parameters in healthy adolescents. Ann Clin Lab Sci. 2005;35(4):428–434.
23. Froom P, Kristal-Bonch E, Benbassat J. Lead exposure in battery –factory workers is not associated with anemia. J Occup Environ Med. 199;41:120–123.
24. Counter SA, Buchanan LH, Ortega F, et al. Blood lead and hemoglobin levels in Andean children with chronic lead intoxication. Neurotoxicology. 2000;21:301–308.
25. Tripathi RM, Raghunath R, Mahapatra S, et al. Blood lead and its effect on Cd, Cu, Zn, Fe and hemoglobin levels of children. Sci Total Environ. 2001;277(1-3):161–168.
26. Zogai D, Krasniqi S, Elezaj I, et al. Correlation between blood lead level and hemoglobin level in Mitrovica children. Med Arch. 2014;68(5):324–328.
27. Mahmoud A, Amin Nada S, Hasan M. Maternal blood lead, sociodemographic and clinical risk factors of unfavourable pregnancy outcomes: A case control study among pregnant women in Egypt. J Appl Sci Res. 2012;8(12):5829–5845.
28. Sim P, Lee H, Kim Y. Effects of iron therapy on blood lead concentrations in infants. J Trace Elem Med Biol. 2014;28(1):56–9.
29. Sim P, Kim Y, Lee H, et al. Iron deficiency increases blood lead levels in boys and pre-menarche girls surveyed in KNHANES 2010-2011. Environ Res. 2014;130:1–6.
30. Wright RO, Tsaih SW, Schwartz J, et al. Association between iron deficiency and blood lead level in a longitudinal analysis of children followed in an urban primary care clinic. J Pediatr. 2003;142:9–14.
31. Wolf AW, Jimenez E, Lozoff B. Effects of iron therapy on infant blood lead levels. J Pediatr. 2003;143:789–795.
32. Kim Y, Lobdell DT, Wright CW, et al. Blood metal concentrations of manganese, lead, and cadmium in relation to serum ferritin levels in Ohio residents. Biol Trace Elem Res. 2015;165:1–9.
33. Rosado JL, Lopez P, Kordas K, et al. Iron and/or zinc supplementation did not reduce blood lead

concentrations in children in a randomized, placebo-controlled trial. J Nutr. 2006;136:2378–2383.
34. Bárány E, Bergdahl IA, Bratteby LE, et al. Iron status influences trace element levels in human blood and serum. Environ Res. 2005;98(2):215–23.
35. Serwint JR, Damokosh AI, Berger OG, et al. No difference in iron status between children with low and moderate lead exposure. J Pediatr. 1999;135:108–110.
36. Hammad TA, Sexton M, Langenberg P. Relationship between blood lead and dietary iron intake in preschool children: a cross-sectional study. Ann Epidemiol. 1996;6(1):30–3.
37. Park JH, Park S, Kim Y. Iron deficiency is not associated with increased blood cadmium in infants. Ann Occup Environ Med. 2014;26:3.
38. Choi JW, Kim SK. Association between blood lead concentrations and body iron status in children. Arch Dis Child. 2003;88:791–792.
39. Choi JW, Kim SK. Relationships of lead, copper, zinc, and cadmium levels versus hematopoiesis and iron parameters in healthy adolescents. Ann Clin Lab Sci. 2005;35:428–434.
40. Barany E, Bergdahl IA, Bratteby LE, et al. Iron status influences trace element levels in human blood and serum. Environ Res. 2005;98:215–223.

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PEER REVIEW

Not commissioned. Externally peer reviewed.

CONFLICTS OF INTEREST

The authors declare that they have no competing interests.

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ETHICS COMMITTEE APPROVAL

The IRB approval of this research was sought by the Ethics Committee of Jordan University of Science and Technology [GM760]. The research protocol was approved by the institutional Ethics Committee (IRB) of the King Abdulla University Hospital (KAUH) at Jordan University of Science and Technology. All women signed an informed consent form agreeing to participate in this study. All participants were assured that the data they provide will not be traced back to them in reports, presentations and other forms of dissemination. The use of pseudonyms names ensured that none of the participants would be identifiable and remain anonymous.

Table 1: The socio-demographic and relevant characteristics of the participants

Age	**n**	**%**
<30	92	55.8
≥30	73	44.2
Education		
<bachelor degree	68	42.2
≥bachelor degree	93	57.8
Income (JDs; 1 JD = 1.04 US $)		
< 600	101	60.8
≥600	65	39.2
Parity		
Primiparous	37	22.4
02-Mar	87	52.7
>3	41	24.8
Trimester		
First	17	10.2
Second	19	11.4
Third	131	78.4
Iron intake/Day	87	52.4
Multivitamins Intake	68	41

Table 2: The means of haemoglobin, lead, and iron according to the studied characteristics

	Haemoglobin level (g/dl)			Lead level (µg/dL)			Iron level (mg/L)		
	Mean	**SD**	**P value**	**Mean**	**SD**	**P value**	**Mean**	**SD**	**P value**
Age			0.271			0.602			0.618
<30	10.8	1.7		12.1	3.3		2.3	2.7	
≥30	10.5	1.5		12.4	4.1		2.1	2.6	
Education			0.899			0.667			0.793
<bachelor	10.6	1.6		12.4	3.9		2.1	2.5	
≥bachelor	10.6	1.6		12.1	3.5		2.3	2.8	
Income			0.020			0.048			0.889
< 600	10.9*	1.7		11.8*	3.5		2.3	2.8	
≥600	10.3	1.4		12.9	3.7		2.2	2.4	
Parity			0.508			0.246			0.867
Primiparous	10.9	1.8		12	3		2.4	1.4	
≤3	10.6	1.6		12.6	3.9		2.1	2.8	
>3	10.5	1.6		11.6	3.5		2.4	3.1	
Trimester			<0.005			0.04			0.316
First	12.2*	2.1		10.3*	1.9		2.1	1.4	
Second	11.4	2		11.6	2.6		1.4	0.7	
Third	10.3	1.3		12.5	3.8		2.4	3	
Multivitamins			0.041			0.048			0.441
NO	10.9*	1.7		11.7*	3.4		2.4	3	
YES	10.3	1.4		12.9	3.8		2	2	
*Statistically significant (independent t test)									

Table 3: The differences in the means of haemoglobin and iron between low and high levels of lead

	Lead (µg/dL)				
	<10		**≥10**		
	(n=49)		**(n=118)**		
	Mean	**SD**	**Mean**	**SD**	**P-value ***
Haemoglobin level (g/dl)	10.9	1.7	10.5	1.6	0.149
Iron level (mg/L)	2.4	3.3	2.2	2.2	0.624
*Independent t test					

Table 4: Multiple linear regression of the association between lead and other parameters including haemoglobin and Iron

Dependent Variable	Independent variables	Coefficient (B)	SE (B)	95% confidence interval		P-value
Hb						
	Age in years	0	0.02	-0.05	0.05	0.897
	Education	0.15	0.23	-0.31	0.62	0.516
	Income	-0.43	0.25	-0.92	0.06	0.082
	Parity	-0.07	0.19	-0.45	0.31	0.712
	Gestational age	-0.36	0.06	-0.48	-0.25	<0.005
	Iron intake	-0.08	0.28	-0.63	0.47	0.767
	Multivitamins use	-0.42	0.24	-0.9	0.06	0.083
	Lead	0.04	0.03	-0.02	0.1	0.223
Iron						
	Age in years	-0.05	0.05	-0.16	0.06	0.377
	Education	0.36	0.52	-0.67	1.39	0.489
	Income	-0.17	0.56	-1.27	0.93	0.76
	Parity	0.23	0.43	-0.62	1.09	0.588
	Gestational age	0.16	0.13	-0.09	0.42	0.209
	Iron intake	-0.58	0.62	-1.8	0.64	0.346
	Multivitamins use	-0.9	0.54	-1.98	0.17	0.100
	Lead	0.13	0.08	-0.03	0.29	0.116