IJN
Iranian Journal of Neonatology
http://ijn.mums.ac.ir

Open Access
Original Article

The Association of Amniotic Fluid Cadmium Levels with the Risk of Preeclampsia, Prematurity and Low Birth Weight

Karim Ebrahim[1]*, Azadeh Ashtarinezhad[2]

1. Department of Toxicology, School of Pharmacy, Shahid Beheshti University of Medical Sciences, Tehran, Iran
2. Department of Occupational Safety and Health, School of Health, Iran University of Medical Sciences, Tehran, Iran

ABSTRACT

***Background*:** Cadmium is an extremely toxic metal with ubiquitous environmental distribution. Cadmium is a potent reproductive and developmental toxic agent, and intensive environmental exposure to this substance has been reported among the residents of Tehran. In this study, we investigated the correlation between amniotic fluid cadmium levels and the risk of preeclampsia, prematurity and low birth weight.

***Methods*:** This study was conducted on 341 hospitalized pregnant women. Amniotic fluid samples were obtained during childbirth, prepared by acid digestion, and cadmium concentration was determined using atomic absorption spectrophotometer equipped with a graphite furnace.

***Results*:** In this study, the mean amniotic fluid cadmium concentration in non-smokers was significantly lower (780± 0.231 µg/l) compared to smokers (13.4±0.91 µg/l). Increased amniotic fluid cadmium levels were associated with the reduction of birth weight, and birth weight was below 2500 g in all the cases with amniotic fluid cadmium levels of >15µg/l. Moreover, preeclampsia was more prevalent among smoking mothers compared to non-smokers. The total incidence rate of preeclampsia (12.6%) was significantly higher in this study compared to similar studies. In addition, no correlation was found between amniotic fluid cadmium levels and the rate of prematurity.

Conclusion: According to the results of this study, cadmium levels of amniotic fluid could be used as sensitive and reliable biomarkers to monitor the reproductive and developmental toxicity of cadmium.

Keywords: Cadmium, Amniotic fluid, Preeclampsia, Prematurity, Low birth weight

Introduction

Cadmium is an extremely toxic metal, which is widely distributed in the environment. This pollutant has no natural sources, and millions of tons of cadmium have been added to the environment through human activities over the past two centuries. Tobacco smoke is one of the most important sources of cadmium exposure in the general population, and cadmium blood levels have been shown to increase by approximately 0.1-0.2 µg/l after smoking one cigarette. Environmental exposure to cadmium most commonly occurs in the form of chronic exposures via drinking water (1, 2).

Cadmium exerts several toxic effects on humans and experimental animals; this substance mainly targets the kidneys, liver and the cardiovascular system (3). Several studies have reported a direct association between cadmium exposure and increased risk of bone fracture, renal dysfunction, hypertension and cancer (4).

Furthermore, cadmium is a potent reproductive and developmental toxic agent; by crossing the placenta and entering the fetus, cadmium is able to cause a wide spectrum of deleterious effects on the reproductive system and fetal development.

On the other hand, high doses of cadmium in animals could lead to congenital developmental disorders and fetal death accompanied by placental necrosis, while low-level exposure throughout gestation might result in retarded fetal growth and low birth weight (LBW) (2, 5).

According to several experimental studies, cadmium toxicity results in numerous adverse effects during the prenatal period; for instance, cadmium exposure was observed to cause early onset of puberty and changes in the mammary gland of female offspring in rats. Moreover, cadmium exposure was found to affect immune system function during the prenatal period (6).

* *Corresponding author*: Karim Ebrahim, Department of Toxicology, School of Pharmacy, Shahid Beheshti University of Medical Sciences, Tehran, Iran. Tel: 00982188653239; E-mail: k.najafabady@sbmu.ac.ir

In a study by Siegers et al. (1983), cadmium concentration in amniotic fluids was determined in 155 pregnant (smokers and non-smokers), and the obtained results indicated that cadmium concentration in the amniotic fluids of smoking pregnant women was approximately three times higher compared to non-smokers. Nevertheless, no correlation was observed between amniotic fluid cadmium levels and other factors such as age, week of gestation, blood pressure and pregnancy-induced complications (7).

Hypertension is a relatively common medical complication during pregnancy, and preeclampsia is the most severe form of pregnancy-induced hypertension. Preeclampsia occurs in 7-10% of all pregnancies worldwide and is a major cause of maternal and prenatal mortality and morbidity. Cadmium intoxication is believed to induce hypertension, and several studies have indicated that pregnant humans and animals are comparatively more sensitive to the toxic effects of cadmium (1).

In another study, Dawson et al. (1999) observed a significant increase in the cadmium levels of amniotic fluid in patients with preeclampsia (8). In addition, many studies have investigated the correlation between the placental and umbilical cord blood, mother's whole blood level of cadmium and LBW of the neonate; however, adequate information is lacking on the association between the cadmium levels of amniotic fluid and the risk of LBW (2, 9, 10).

Tehran, the capital of Iran, is one of the most air-polluted cities in the world, and several surveys have reported high levels of cadmium exposure in the general population of this city (11). The present study aimed to investigate the association between cadmium levels of amniotic fluid and disorders such as preeclampsia, prematurity and the risk of LBW in five different public hospitals of Tehran.

Method

Subjects and Study Design

This cohort study was conducted on 341 pregnant women hospitalized in five public hospitals in Tehran from September 2011 to December 2012.

All the subjects completed questionnaires including information about age, ethnic origin, employment status, smoking habits, dietary patterns, use of medications and number of previous pregnancies and childbirths.

Other recorded data were the time of birth, weight and height of the neonates. Exclusion criteria were as follows: 1. pre-existing hypertension or history of preeclampsia; 2. liver, renal, cardiovascular or endocrine diseases; 3. use of nutritional supplements, diuretics, or receiving hormonal treatments.

Furthermore, patients with severe disorders such as diabetes, epilepsy, renal disease and Rh-factor disorders were excluded from the study. Written informed consent was provided from all the subjects prior to participation.

Since the majority of reports on smoking pregnant women are ambiguous, and many of these women declare passive exposure to tobacco smoke, this parameter is determined through the assessment of plasma thiocyanate (SCN (-)) levels in pregnant women. According to the literature (12), patients with thiocyanate concentrations of <59 μM/L are considered as non-smokers, while those with thiocyanate concentrations of >60 μM/L are defined as smokers.

The clinical criteria to define a preeclamptic condition was A) systolic blood pressure >140 mmHg and/or diastolic blood pressure >90 mmHg after 20th week of gestation, B) proteinuria >500 mg per 24 hours. Moreover, the birth weight of neonates was recorded and converted to a dichotomous measure of LBW (Yes/No) (<2500 g; LBW).

Based on the cadmium concentration in the amniotic fluid, the subjects were divided into the following groups: 1. low exposure (<1 μg/l), 2. moderate exposure (1-10 μg/l) and 3. high exposure (>10 μg/l). Differences in the prevalence of LBW (<2500 g), preeclampsia and prematurity were also compared between these groups.

Sampling

Samples of amniotic fluids were provided from all the subjects for diagnostic purposes during childbirth and were properly labeled and stored at -20ºC until analysis.

Measurement of Cadmium Concentration in Amniotic Fluids

To determine cadmium concentrations, the samples were prepared via acid digestion for analysis, and 5 ml of 1:1 mixture of Per chloric acid (70%) and nitric acid (65%) were added to 1 ml of amniotic fluids. Afterwards, the samples were heated until drying, reconstituted with nitric acid and distilled water, and analyzed using an atomic absorption spectrophotometer (AAS) (Model 4000, Perkin Elmer Corp, USA), which was equipped with a graphite furnace for the accurate measurement of cadmium.

All measurements were conducted in duplicate, and repeated measures were performed for suspicious results.

Statistical analysis

To examine the normal distribution of data, Shapiro-Wilk test was performed. If necessary, data were analyzed using non-parametric linear regression, Student's t-test and one-way analysis of variance (ANOVA), as well as Dunnett's test for multiple comparisons. The obtained values were expressed in Mean± Standard Error of the mean (SEM), and a P value of <0.05 was considered significant. All the statistical analyses in this study were performed using Graph Pad Prism 5© software.

Results

In total, 341 pregnant women with the mean age of 33.6±5.1 years were enrolled in this study. Approximately 83.2% of the studied subjects were within the age range of 20-35 years, 2% were under 18 years of age, and 9.7% were over 35 years. In addition, the majority of the subjects (79.1%) were multiparous, and others (21.9%) were nulliparous.

No significant difference was observed between multiparous and nulliparous patients in terms of the incidence of LBW, preeclampsia and prematurity (data not shown). Furthermore, other demographic features had no significant effects on the occurrence of LBW, preeclampsia and prematurity. In this study, all the patients had been residents of Tehran within the past three years, and no one had experienced occupational exposure to cadmium.

For the measurement of cadmium concentration in amniotic fluids by AAS, limit of detection (LOD) and limit of quantification (LOQ) were estimated to be 28 ng/l and 71 ng/l, respectively; within-day and between-day and 11.2%, respectively. In addition, the concentration of SCN (-) in 46 subjects was >59 coefficient of variation in this method were 8.5% therefore, these subjects were considered as smokers.

The mean cadmium concentration of amniotic fluid in non-smokers (780± 0.231 ng/l) was significantly lower compared to smokers (13.4± 0.91 µg/l). As shown in Figure 1, a significant correlation was observed between the amniotic fluid cadmium concentrations and SCN (-) levels in subjects with smoking habits (r^2=0.882).

Moreover, mean of birth weight in smokers and non-smokers was 3187±241 g and 3445±829 g, respectively; however, no significant difference was observed between these two variables according to the student's t-test (P<0.05).

In the group of smoking mothers, 7 neonates (15.2%) and in the non-smoker group 37 neonates (12.5%) had LBW (≤2500 g). The analysis of birth weight and amniotic fluid cadmium levels via non-parametric linear regression was indicative of a correlation between the reduction of birth weight and an increase in the cadmium levels of amniotic fluids (Figure 2).

On the other hand, the prevalence of preeclampsia among the smoking mothers was 26% (N=12), while it was 10.5% (N=31) among non-smokers, and a significant difference was observed between these two variables according to the student's t-test (P<0.05).

The total incidence rate of preeclampsia in all the subjects of this study was significantly higher (12.6%) than the results of similar studies in this regard. Moreover, no significant difference was observed in terms of the prevalence of prematurity between smokers and non-smokers in the present study.

In this study, the subjects were divided into three groups based on the level of cadmium concentration in amniotic fluids, and the differences in the incidence of LBW (<2500 g), preeclampsia and prematurity were investigated using ANOVA.

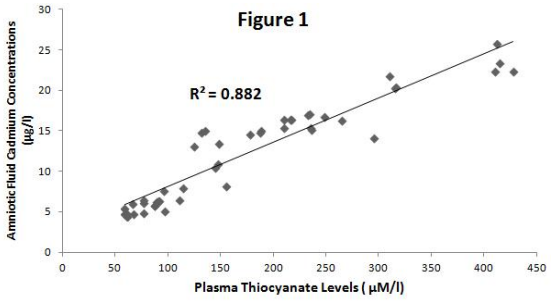

Figure 1. The correlation between amniotic fluid cadmium concentrations and plasma thiocyanate levels

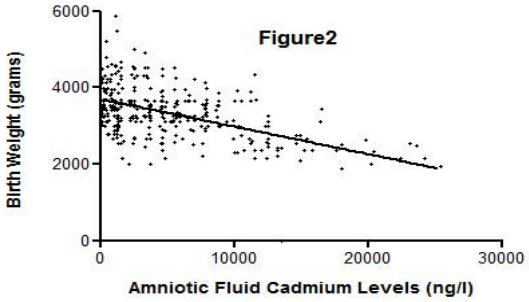

Figure 2. The relationship between birth weight and amniotic fluid cadmium levels. A significant decrease in birth weight with the increased levels of amniotic fluid cadmium can be observed

Table 1. Exposure status and its relations with incidence of low birth weight, preeclampsia and prematurity

Exposure status	Low (<1 µg/l)	Moderate (1-10 µg/l)	High (>10 µg/l)
Total number	112 (32.8 %)	173 (50.7%)	56 (16.4%)
Mean birth weight	3531±426*	3311±424*	3102±752*
Number of low birth weight cases(≤2500g)	11 - (9.8% of cases in this group) -(26.1% of total low birth weight cases)	18* - (10.4% of cases in this group) -(42.8% of total low birth weight cases)	13* - (23.2% of cases in this group) -(30.9% of total low birth weight cases)
Preeclamptic cases	11 - (9.8% of cases in this group) - (25.5% of total preeclamptic cases)	20* - (11.5 % of cases in this group) - (46.5% of total preeclamptic cases)	12* - (21.4% of cases in this group) - (32.5% of total preeclamptic cases)
Number ofprematurity cases	4 (36% of total prematurity cases)	5 (45% of total prematurity cases)	2 (19% of total prematurity cases)

* There is significant difference between groups, determined by ANOVA test, (P<0.05)

As depicted in Table 1, the mean birth weight in each exposure group was significantly different from other exposure groups and an increase in exposure intensity lead to a reduction in the mean of birth weight. In addition, the incidence of LBW in the moderate- and high-exposure groups was significantly higher compared to the low-exposure group.

Furthermore, 21.4% of the pregnant women in the high-exposure group were found to experience preeclamptic conditions during their pregnancy; this rate was significantly higher than the other exposure groups. However, no significant association was observed between the rate of prematurity and level of exposure in the three groups.

Discussion

Cadmium is a toxic metal, which is added to the environment through human activities such as combustion of fossil fuels, leach of landfill wastes and runoff from agricultural land and mining. Moreover, cadmium is a by-product of electroplating, manufacture of Nickel-cadmium batteries, pigments, stabilizers and alloys (3).

Dietary intake of cadmium in European countries has been estimated to be between 10-30µg per day (14); accordingly, consumption of certain foods such as shellfish, offal and rice could increase the risk of cadmium intake (15). Another significant human route of cadmium intake is smoking, with an estimated 0.2-1.0µg of cadmium per smoking one cigarette (16).

Pregnancy is a critical period in terms of cadmium toxicity, and several adverse outcomes such as preeclampsia, LBW, prematurity, stillbirth and developmental effects have been attributed to acute, sub-acute and chronic cadmium exposure during pregnancy (3, 14, 17). Cadmium accumulates in the placenta interacting with the transport of micro-nutrients and may play a key role in the occurrence of intrauterine growth restriction (18).

Furthermore, cadmium has been shown to inhibit the activity of 11-β-hydroxysteroid dehydrogenase enzyme, while decreasing 11-β-HSD2 mRNA and protein in cultured human trophoblast cells. Reduced 11-β-HSD2 activity in the placenta is directly associated with the reduction of intrauterine growth (19).

For the first time, Siegers et al. (1983) determined amniotic fluid cadmium levels in 155 pregnant women (128 non-smokers and 27 smokers). According to their findings, levels of cadmium concentration were noticeably higher in the amniotic fluids of smokers (mean: 7.29 ± 2.39 µg/l) compared to non-smokers (mean: 2.58 ± 1.36 ng/l).

On the other hand, a significant correlation was observed between cadmium levels and daily consumption of cigarettes in the present study. These values were significantly higher in our study groups compared to previous studies in this regard (780± 0.231 ng/l in non-smokers vs. 12.4± 0.91 µg/l in smokers).

With respect to the findings of previous studies about environmental contamination of cadmium in Tehran and the daily uptake of this substance among the residents of this capital city (11, 20), the remarkable increase in the cadmium concentration of amniotic fluids of pregnant women was not unexpected.

In one study, Menai et al. (2012) measured maternal blood cadmium levels in 901 women during pregnancy investigating the potential effects of maternal environmental cadmium exposure on the birth weight and fetal growth restriction of the neonates. According to their results, maternal cadmium levels were associated with LBW in the offspring of women with smoking habits during pregnancy.

These findings confirmed the effects of cadmium toxicity on fetal growth through the probable accumulation and transition of this substance to the placenta (21). As previously mentioned, no significant difference was observed

between the mean of birth weight in smokers and non-smokers in the current study, which could be due to the small sample size of the smoker group, or the high variations in the birth weight of the neonates.

One of the important findings regarding birth weight is that an increase in the cadmium levels of amniotic fluids could lead to a reduction in birth weight. In the present study, birth weight was below 2500 g in almost all the cases with amniotic fluid cadmium levels of >15 µg/l; therefore, it could be concluded that amniotic fluid cadmium levels of >15 µg/ml are significant risk factors for fetal growth and could considerably affect the pregnancy outcome.

In another study, Dawson et al. (1999) demonstrated that amniotic fluid levels of lead, calcium, magnesium, zinc and selenium between 33-40 weeks of gestation could cause significant differences between normal and preeclamptic pregnancies (8).

The observed changes in the levels of calcium and cadmium were consistent with the results of animal and human studies, in which sub-acute or chronic cadmium exposures were found to be associated with depression. The observed changes in the levels of calcium and cadmium were consistent with the results of animal and human studies, in which sub-acute or chronic cadmium exposures were found to be associated with calcium intake depression. Cadmium interference with calcium homeostasis and might results in physical syndromes such as to preeclampsia (22, 23).

In a study by Kosannovic et al. (2002), high concentrations of cadmium were detected in the umbilical cord blood and amniotic fluids of preeclamptic pregnant women (1); these findings were confirmed by several other studies (24).

In the present study, a significant difference was observed in the incidence of preeclampsia between smokers and non-smokers; the total incidence rate in all the subjects was reported to be 12.6%, which was significantly higher than the rates reported in the normal population (7-10%). Moreover, it was indicated that 21.4% of the subjects in the high-exposure group (amniotic fluid cadmium levels of >10 µg/l) experienced preeclamptic conditions during their pregnancy, which was significantly different from other exposure groups and confirmed the pivotal role of cadmium toxicity in pregnancy-induced hypertension.

Although several studies have been conducted on the effects of cadmium toxicity on pregnancy outcome (e.g. premature labor) (25, 26) no correlation was observed between the cadmium levels of amniotic fluids and rate of prematurity in the current study.

Conclusion

Amniotic fluid cadmium levels could be used as sensitive and reliable biomarkers of reproductive and developmental toxicity of cadmium. Therefore, determination of cadmium toxicity during pregnancy could predict the potential effects of this substance on fetal development and offer constructive solutions on environmental exposure control, quitting smoking and use of chelation therapy if necessary.

Acknowledgment: We thank Foroug Forouzandeh for technical assistance. We are also indebted to Dr. Fatemeh Nadafi for reading the manuscript.

References

1. Kosanovic M, Jokanovic M, Jevremovic M, Dobric S, Bokonjic D. Maternal and fetal cadmium and selenium status in normotensive and hypertensive pregnancy. Biol Trace Elem Res. 2002; 89:97-103.
2. Ronco AM, Urrutia M, Montenegro M, Llanos MN. Cadmium exposure during pregnancy reduces birth weight and increases maternal and foetal glucocorticoids. Toxicol Lett. 2009; 188: 186-91.
3. Satarug S, Garrett SH, Sens MA, Sens DA. Cadmium, environmental exposure, and health outcomes. Ciência & Saúde Coletiva. 2011; 16: 2587-602.
4. Satarug S, Baker JR, Urbenjapol S, Haswell-Elkins M, Reilly PE, Williams DJ, et al. A global perspective on cadmium pollution and toxicity in non-occupationally exposed population. Toxicol Lett. 2003; 137:65-83.
5. Fréry N, Nessmann C, Girard F, Lafond J, Moreau T, Blot P, et al. Environmental exposure to cadmium and human birthweight. Toxicology. 1993; 79:109-18.
6. Kippler M, Hoque AM, Raqib R, Ohrvik H, Ekström EC, Vahter M. Accumulation of cadmium in human placenta interacts with the transport of micronutrients to the fetus. Toxicol Lett. 2010; 192:162-8.
7. Siegers CP, Jungblut JR, Klink F, Oberheuser F. Effect of smoking on cadmium and lead concentrations in human amniotic fluid. Toxicol Lett. 1983; 19:327-31.
8. Dawson EB, Evans DR, Nosovitch J. Third-trimester amniotic fluid metal levels associated with preeclampsia. Arch Environ Health. 1999; 54:412-5.
9. Llanos MN, Ronco AM. Fetal growth restriction is related to placental levels of cadmium, lead and arsenic but not with antioxidant activities. Reprod Toxicol. 2009; 27:88-92.

10. Piasek M, Blanusa M, Kostial K, Laskey JW. Placental cadmium and progesterone concentrations in cigarette smokers. Reprod Toxicol. 2001; 15:673-81.
11. Farzin L, Amiri M, Shams H, Ahmadi Faghih MA, Moassesi ME. Blood levels of lead, cadmium, and mercury in residents of Tehran. Biol Trace Elem Res. 2008; 123:14-26.
12. Ellingsen DG, Thomassen Y, Aaseth J, Alexander J. Cadmium and selenium in blood and urine related to smoking habits and previous exposure to mercury vapour. J Appl Toxicol. 1997; 17:337-43.
13. Díaz E, Halhali A, Luna C, Díaz L, Avila E, Larrea F. Newborn birth weight correlates with placental zinc, umbilical insulin-like growth factor I, and leptin levels in preeclampsia. Arch Med Res. 2002; 33:40-7.
14. Thompson J, Bannigan J. Cadmium: toxic effects on the reproductive system and the embryo. Reprod Toxicol. 2008; 25(3):304-15.
15. Rivai IF, Koyama H, Suzuki S. Cadmium content in rice and its daily intake in various countries. Bull Environ Contam Toxicol. 1990; 44:910-6.
16. Satarug S, Moore MR. Adverse health effects of chronic exposure to low-level cadmium in foodstuffs and cigarette smoke. Environ Health Perspect. 2004; 112:1099–103.
17. Nishijo M, Nakagawa H, Honda R, Tanebe K, Saito S, Teranishi H, et al. (2002) Effects of maternal exposure to cadmium on pregnancy outcome and breast milk. Occup Environ Med. 2002; 59:394–97.
18. Yang Q, Wen SW, Smith GN, Chen Y, Krewski D, Chen XK, et al. Maternal cigarette smoking and the risk of pregnancy-induced hypertension and eclampsia. International journal of epidemiolog. 2006; 35: 288-93.
19. Murphy VE, Smith R, Giles WB, Clifton VL. Endocrine regulation of human fetal growth: the role of the mother, placenta, and fetus. Endocr Rev. 2006; 27:141-69.
20. Shariatpanahi M, Anderson AC. Accumulation of cadmium, mercury and lead by vegetables following long-term land application of wastewater. Sci Total Environ. 1986; 52:41-7.
21. Menai M, Heude B, Slama R, Forhan A, Sahuquillo J, Charles MA, et al. Association between maternal blood cadmium during pregnancy and birth weight and the risk of fetal growth restriction: The EDEN mother-child cohort study. Reprod Toxicol. 2012; 34:622-7.
22. Godt J, Scheidig F, Grosse-Siestrup C, Esche V, Brandenburg P, Reich A, et al. The toxicity of cadmium and resulting hazards for human health. J Occup Med Toxicol. 2006; 1: 22.
23. Martelli A, Rousselet E, Dycke C, Bouron A, Moulis JM. Cadmium toxicity in animal cells by interference with essential metals. Biochimie. 2006; 88:1807-14.
24. Vigeh M, Yokoyama K, Ramezanzadeh F, Dahaghin M, Sakai T, Morita Y, et al. Lead and other trace metals in preeclampsia: a case–control study in Tehran, Iran. Environ Res. 2006; 100:268-75.
25. Tabacova S, Baird DD, Balabaeva L, Lolova D, Petrov I. Placental arsenic and cadmium in relation to lipid peroxides and glutathione levels in maternal-infant pairs from a copper smelter area. Placenta. 1994; 15:873-81.
26. Zhang YL, Zhao YC, Wang JX, Zhu HD, Liu QF, Fan YG, et al. Effect of environmental exposure to cadmium on pregnancy outcome and fetal growth: a study on healthy pregnant women in China. J Environ Sci Health A Tox Hazard Subst Environ Eng. 2004; 39:2507-15.