Environmental Exposures in the Etiology of Abortion: Placental Toxic and Trace Element Levels

Authors

Emre Baser[1], Demet Aydogan kırmızı[1], Vugar Ali Turksoy[2], Taylan Onat[1], Melike Demir Çaltekin[1], Mustafa Kara[3], Ethem Serdar Yalvac[1]

Affiliations

1 Yozgat Bozok University, Faculty of Medicine, Department of Obstetrics and Gynecology, Yozgat, Turkey
2 Yozgat Bozok University, Faculty of Medicine, Department of Public Health, Yozgat, Turkey
3 Ahi Evran University Training and Research Hospital, Department of Obstetrics and Gynecology, Kırşehir, Turkey

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Correspondence

Dr. Emre Baser
Department of Obstetrics and Gynecology
Faculty of Medicine
Yozgat Bozok University
66100 Yozgat
Turkey
Tel.: +905052749203
emrebasermd@gmail.com

ABSTRACT

Purpose Intensive research has been conducted on the effects of toxic and trace elements on pregnancy. Previous studies indicated a possible relationship between placental levels of these elements and first-trimester abortion; however, their effects on the further gestational weeks are not clear. This study aimed to investigate the effect of changes in the levels of placental trace and toxic elements on second-trimester abortion.

Methods The patient group consisted of 30 women with missed abortion. The control group comprised 60 healthy term and singleton pregnant women who gave birth. Placental samples were obtained from the patients and the healthy controls, and the concentrations of placental elements were measured using inductively coupled plasma mass spectrometry.

Results In the abortion group, placental arsenic, cadmium, mercury, lead, antimony, tin, cobalt, manganese, and selenium levels were significantly higher than those of the control group ($p < 0.05$). Antimony was determined as an independent predictor with an odds ratio of 6.1 in toxic elements ($p = 0.025$), and selenium was determined as an independent predictor with an odds ratio of 2.3 in trace elements ($p = 0.015$).

Conclusion The changes in trace element and toxic element levels, especially an increase in antimony and selenium, in placental tissue due to environmental exposure may play an important role in second-trimester abortion.

Introduction

Spontaneous abortion (SA) can be defined as pregnancy loss that occurs before 20 weeks of gestation or under 500 g [1]. Of clinically defined pregnancies, 10–15 % result in SA and the etiology of this condition has not yet been established [2]. SA is classified as first/second-trimester SA, according to whether it occurs before or after 12 weeks of gestation [3–5]. More than 50 % of the abortions in the first trimester are caused by chromosomal abnormalities [6]. It is believed that this condition occurs as a result of the combination of many causes, including chromosomal abnormalities, abnormal intrauterine environment, endocrine diseases, immune dysfunction, excessive smoking, trauma, severe systemic infections, and nutritional deficiencies [7–9].

The rapid development of industry and the use of large amounts of heavy metals increase exposure to environmental toxins. There is evidence that some harmful chemicals can halt embryonic development [10]. Heavy metals are toxic elements, even in small amounts. They are taken into the body by ingestion, respiration, and absorption through the skin. Most heavy metals cannot be removed from the body without special support. Slowly accumulating, these toxic metals can affect the structure of enzymes, DNA, mitochondria in the body, and directly or indirectly disrupt the membrane structure, permeability, and metabolism of cells. Prenatal life, the most sensitive period of human development, involves high levels of cellular division and differentiation [11]. Therefore, fetuses are very sensitive to teratogens even at low levels of exposure that do not harm their mother [12]. Exposure, especially during organogenesis, can cause permanent structural and anatomic changes, which can subsequently lead to functional consequences [13]. Studies have shown that heavy metals such as lead (Pb), cadmium (Cd), or arsenic (As) are associated with poor pregnancy outcomes such as SA, stillbirth, pregnancy hypertension, low birth weight (LBW), and low Apgar scores [14–18].

Recent studies on the etiology of abortion show that processes related to inflammation and oxidation can be effective in these reactions. These enzymatic reactions take place in the co-factor of trace elements such as manganese (Mn), zinc (Zn), selenium (Se), cobalt (Co), and copper (Cu). They are found in very low levels in serum and tissues (picogram, microgram levels), and also have very important effects in human pregnancy.

There are a limited number of studies examining placental levels of trace and toxic elements in cases of abortion. The main objective of the present study was to compare placental toxicity and essential element levels and to determine their place in the etiology of abortion in healthy patients with second-trimester SA.

Materials and Methods

The participants were selected from among patients attending the Department of Obstetrics and Gynecology of our hospital between March 2018 and December 2018. The study protocol was approved by our hospital Ethics Committee (2018-KAEK-189_2018.02.27-07). A statement of informed consent was obtained from all patients including the parents/legally authorized representative of illiterate participants before the onset of the study.

To detect an effect size of 0.765 at alpha error of 0.05 and statistical power of 0.85, 64 participants were required for our study, based on the result of a similar study [19]. A total of 90 patients, 30 cases and 60 controls, were included in the study. The study group consisted of patients diagnosed as having SA at 12–20 weeks of their gestational period. Abortus was defined as the absence of fetal heartbeat in transvaginal and abdominal ultrasonography (USG) and/or inability to observe the fetus using USG following bleeding/pain in a previously known patient [20]. These patients had an SA using medical therapy. Curettage devices were not used so as to prevent heavy metal contamination. All materials were kept in sterile plastic containers. The control group comprised healthy, singleton patients at 37 weeks of gestation and over with no complications. Patients aged between 19 and 35 years were included in the study. Multiple pregnancy, fetal anomaly, ectopic pregnancy, molar hydatidiform, uterine anatomical disorders (benign tumors, endometrial polyp, congenital uterine anomalies), smoking habit, tooth filling, infectious diseases including toxoplasmosis, cytomegalovirus and herpes (TORCH), deep anemia, coagulation disorders, autoimmune diseases, hypertension, and endocrinologic and chronic systemic diseases were regarded as the exclusion criteria.

Complete blood count, routine biochemistry, toxicology, coagulation, thyroid function tests, TORCH, and other laboratory parameters of the patients were taken under record. Information regarding age, body mass index (BMI), gravidity, parity, chronic and systemic diseases, drugs, amalgam fillings, and passive smoking status was gathered and recorded through questionnaires completed in face-to-face interviews.

Toxicologic analysis

The placental tissues were separately labeled and stored at −20 °C until the analysis preparation phase and then transferred onto a glass table. The tissues on the glass tray, together with the table, were pre-dried at 75 °C for 24 h in an incubator. Tissue samples collected from the incubator were weighed to determine dry matter using a precision scale. A minimum of 0.2 g placental dry tissues for each sample for toxicological analysis was taken and placed in Teflon tubes in a microwave oven (Start D – Microwave Digestion System; Milestone Srl, Sorisole, Italy). Five milliliters of nitric acid (HN03 65 % Suprapur; Fisher Scientific, Waltham, MA, USA) and 5 mL of ultrapure water were added to the placental samples and digested in the microwave. Then samples were transferred to 50-mL polypropylene tubes and the total volume was filled to 20 mL with deionized water. Samples were stored at +4 °C in the polypropylene tubes before analysis. The metal levels of the digested samples in the microwave oven were determined using inductively coupled plasma mass spectrometry (ICP-MS) (Thermo Scientific ICAP Qc). Method validations were performed using certified reference material (Seronorm Trace Elements Whole Blood L-2; Sero, Bilingstad, Norway). CRM was measured five times on the same day and on different days. The average of the repeated measurements was used for the validation of this method whereby the relative standard deviation (RSD) values did not exceed 5 %. The r2 values of the calibration curves of all metals were calculated as 0.9998, 0.9998, and 0.9993, respectively. The interval of calibration was set to 0.5–1000 µg/L for all metals [21].

Standards and reagents

Nitric acid (Suprapur, 65 %; Merck KGaA, Darmstadt, Germany) was used for the sample and standard reference material digestion. Ultra-pure water (Direct-Q; MilliporeSigma, Burlington, MA, USA) was used for dilution standard (6020 Cal, Denge 24; Inorganic Ventures, Christiansburg, VA, USA; 19E Multi Element Standard; ChemLab, Zedelgem, Belgium) and sample preparation. CRM (Seronorm Trace Elements Whole Blood L-2) was used for validation.

Statistical analysis

Statistical analysis was performed using the SPSS software (version 20; IBM Corp. Armonk, NY, USA). The data are expressed as mean (SD) and in percentiles. The distribution of the variable data was

determined using visual (histograms, probability plots) and analytical methods (Kolmogorov-Smirnov and Shapiro-Wilk's test). The Mann-Whitney U test was used for non-parametric numerical data and Student's t-test was used for parametric numerical data. Categorical data were compared through the use of the Chi-square test or Fisher's exact test. Multivariate logistic regression analysis was performed to predict SA. Spearman's correlation test was used to evaluate the difference between age, BMI, parity, and toxic elements levels. A p-value less than 0.05 was considered significant. The study population was determined using the G * Power program [22].

Results

A total of 55 patients with SA were admitted to our hospital between the indicated periods ▶**Fig. 3**. Of these patients, 30 met the inclusion criteria. The control group consisted of 60 pregnant women who had singleton births after a term pregnancy and met the criteria for inclusion in the study. In the study group, the mean week of gestational loss was 15. ± 2.5 according to the last menstrual period (LMP) and 13.9 ± 2.3 according to USG. In the control group, the gestational age was 38.5 ± 1.5 weeks for LMP and 37.4 ± 1.6 years for USG. The mean hemoglobin and hematocrit values of the patients were 12.1 ± 1.5 and 37.8 ± 11.7 g/dL, respectively, and there was no significant difference between the two parameters (P < 0.05) (▶**Table 1**).

Sociodemographic and lifestyle characteristics

The mean age of all patients was 29.4 ± 5.2 years, the mean BMI was 27.3 ± 4.6 kg/m^2, the mean gravidity was 3.7 ± 1.8, the mean parity was 2.2 ± 1.1, and the mean number of stillbirths was 0.5 ± 0.9. There were no significant differences between the groups in terms of these characteristics (P < 0.05). Of the patients, 72.4 % lived in urban areas and there were no significant differences between the groups (▶**Table 1**). As to the educational status of the women participating in the study, 4.4 % of the participants were illiterate, 63.3 % were primary or secondary school graduates and 32.2 % were high school graduates. There were no statistically significant differences between the 2 groups in terms of education level. In addition, no statistically significant differences were determined between toxic and trace element levels according to education level (data not shown). The toxic element levels of the patients according to age, BMI, and parity values were evaluated using correlation analysis and no significant correlations were found (P > 0.05) (▶**Table 4**).

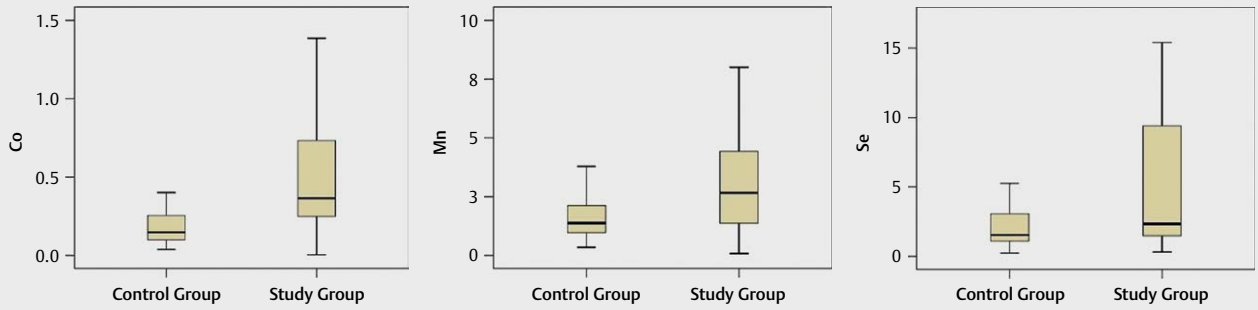
Toxic element and trace element values

Representative placental accumulation of toxic and trace elements are shown in ▶**Fig. 2**. In the SA group, placental aluminum (Al), As, Cd, Hg, Pb, Sb, and Sn levels were significantly higher than in the control group (P < 0.001). However, no significant differences were found for vanadium (V) (P = 0.992) (▶**Table 2**). In the SA group, placental Mn (4.2 ± 5.8, 2.1 ± 2.9, respectively), Co (0.9 ± 1.85, 0.3 ± 0.4, res-

▶**Table 1** Demographic characteristics and some laboratory data.

	All patients	Control group	Case group	p
	n: 90	n: 60	n: 30	
Age (years)	29.4 ± 5.2	26.9 ± 4.4	28.1 ± 5.0	0.077
BMI (kg/m^2)	27.3 ± 4.6	25.7 ± 6.0	28.2 ± 5.2	0.249
Gravidity	3.7 ± 1.8	3.5 ± 1.1	3.7 ± 1.8	0.723
Parity	2.2 ± 1.1	2.2 ± 1.0	2.1 ± 1.1	0.572
Number of stillbirths	0.5 ± 0.9	0.1 ± 0.4	0.7 ± 2.6	0.621
Educational status (%)				0.125
illiterate	4 (4.4)	1 (1.7)	3 (10.0)	
Primary and secondary school	57 (63.3)	39 (65.0)	18 (60.0)	
High school	29 (32.2)	20 (33.3)	9 (30.0)	
Place of residence (%)				0.816
Urban	63 (72.4)	43 (71.7)	20 (74.1)	
Rural	24 (27.6)	17 (28.3)	7 (25.9)	
GW according to USG measurements	13.9 ± 2.3	37.4 ± 1.6	13.9 ± 2.3	< 0.001
GW according to LMP	31.1 ± 11.0	38.5 ± 1.5	15.4 ± 2.5	< 0.001
Hemoglobin (g/dL)	12.1 ± 1.5	12.1 ± 1.6	12.0 ± 1.3	0.894
Hematocrit (%)	37.8 (11.7)	38.4 (14.2)	36.7 (3.2)	0.867

Note: Unless otherwise specified, results are presented as mean (SD). BMI: body mass index; GW: gestational weeks; LMP: last menstrual period.



▶ **Fig. 1** Trace elements that are significantly higher in the abortion group.

▶ **Table 2** Levels of toxic and trace elements in the placenta.

Toxic Elements	Control group		Case group		
	Mean	SD	Mean	SD	p
Aluminum (µg/L)	0.05	0.06	0.40	0.97	<0.001
Arsenic (µg/L)	0.37	0.51	2.08	1.96	<0.001
Chromium (µg/L)	1.41	2.34	0.62	0.64	0.141
Cadmium (µg/L)	0.08	0.14	0.48	0.65	<0.001
Mercury (µg/L)	0.09	0.12	0.41	0.51	<0.001
Lead (µg/L)	0.49	1.17	2.93	2.28	<0.001
Antimony (µg/L)	0.05	0.04	0.70	0.89	<0.001
Tin (µg/L)	1.52	2.91	2.78	2.86	0.001
Vanadium (µg/L)	0.70	0.83	0.62	0.79	0.918
Trace Elements					
Cobalt (µg/L)	0.25	0.34	0.97	1.85	<0.001
Copper (µg/L)	25.80	21.33	42.28	55.91	0.614
Manganese (µg/L)	2.06	2.90	4.20	5.78	0.009
Selenium (µg/L)	2.12	1.39	5.01	4.88	0.018

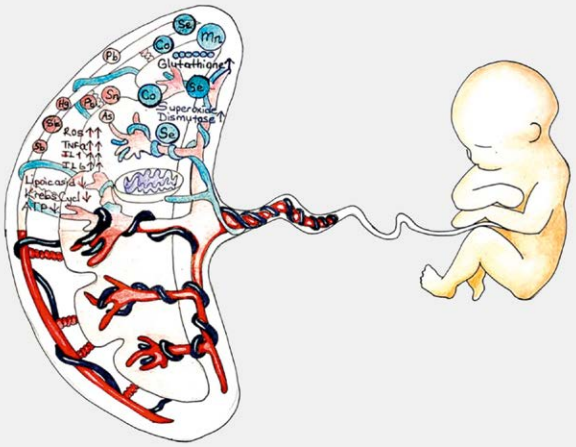
SD: standard deviation

pectively), and Se (5.0 ± 4.9, 2.1 ± 1.4, respectively) levels were significantly higher than in the control group ($p = 0.009$, $p < 0.001$, $p = 0.018$, respectively). By contrast, Cr (0.6 ± 0.6, 1.4 ± 2.34, respectively) and Cu (42.3 ± 55.9, 25.8 ± 21.3, respectively) levels were not significantly different ($p = 0.141$, $p = 0.614$, respectively) (▶**Fig. 1**). To predict SA, significant toxic elements and trace elements were evaluated using multivariate logistic regression analysis. According to this analysis, Sb was determined as an independent predictor with an odds ratio (OR) of 6.1 in toxic elements ($P = 0.025$), and Se was determined as an independent predictor with an OR of 2.3 in trace elements ($P = 0.015$) (▶**Table 3**).

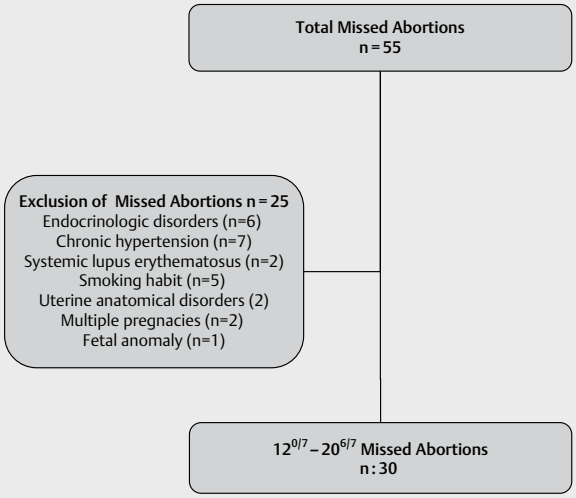
Discussion

In the present study, Al, As, Cd, Hg, Pb, Sb, and Sn levels of placental toxic elements, and Co, Mn, and Se levels of placental trace elements were significantly higher in the SA group compared with those of the control group. To our knowledge, this is the first study to assess placental heavy metal and trace element levels in second-trimester abortion.

The placenta shows histologic changes during pregnancy to ensure fetal-maternal exchange of nutrients. Cytotrophoblasts form a continuous layer up to 12 weeks. By the second trimester, this layer becomes a discontinuous structure [13]. The passage of endogenous and exogenous substances is associated with these exchange and transport mechanisms [13]. During pregnancy, many environmental toxins including heavy metals produce different degrees of fetal exposure due to this changing structure in the 12th



▶ **Fig. 2** Illustration of toxic and trace element accumulation in the placenta (Source: Çiğdem Aydoğan)



▶ **Fig. 3** Flowchart: Details of sample size distribution.

▶ **Table 3** Model of multivariate logistic regression of potential factors on spontaneous abortion.

Toxic Elements					
	B	**SE**	**OR**	**95 % CI**	**p**
Al	50.9	29.2	1.2	0.2–1.3	0.082
As	1.5	1.0	1.5	0.6–34.2	0.137
Cd	0.6	9.6	0.5	0.1–78.0	0.943
Hg	2.8	1.6	1.7	0.7–36.5	0.076
Pb	0.1	0.5	1.1	0.4–3.0	0.856
Sb	25.6	11.4	6.1	0.3–17.0	0.025
Sn	1.3	0.7	0.3	0.2–1.3	0.068
Trace Elements					
	B	**SE**	**OR**	**95 % CI**	**p**
Co	0.8	0.7	1.4	0.6–8.1	0.212
Mn	−0.1	0.1	0.9	0.7–1.1	0.409
Se	0.4	0.1	2.3	1.1–2.9	0.015
Cu	0.0	0.0	1.0	0.9–1.0	0.253

Al: aluminum; As: arsenic; Cd: cadmium; Hg: mercury; Pb: lead; Sb: antimony; Sn: tin; B: standardized regression coefficient; SE: standard error; OR: odds ratio; CI: confidence interval.

week of the placenta. This change in the placenta may play a role in the etiology of second-trimester abortion. Therefore, this study was planned to include patients who were past their 12 weeks of gestation.

Toxic elements

Rapid industrialization and urbanization are the most important factors that increase chronic heavy metal exposure. In particular, chronic exposure to Pb, Cd, and Hg is a global concern. In addition, the widespread use of Al makes exposure inevitable [23]. Pb, Cd, Hg, and Al are usually taken into the body by inhalation and through foods. These toxic elements show their toxic effects by blocking antioxidant systems [24, 25]. These effects are mainly caused by thiol-containing antioxidant enzymes and glutathione peroxidase. The placental passage of these heavy metals is not fully understood, but existing transport systems are thought to be effective. Metallothionein (MT) is the protein that plays the most important role in intracellular metal binding and placental involvement of heavy metals. There is evidence that Hg, Pb, and Cd can induce MT synthesis in the placenta [26–28]. MT regulates metal homeostasis and has antioxidant properties. Cd is the heavy metal that induces MT production at the highest rate. Therefore, the placental involvement of Cd is higher. Defective embryogenesis, premature birth, and LBW caused by Cd exposure may result from an interaction with MT [29]. Diet and smoking are the main sources of Cd exposure. Based on the literature, it is thought that the intense consumption of rice and wheat is the cause of Cd transition, especially in Asian societies [30]. Considering that our patients were non-smokers, it can be assumed that foods were the main source of Cd. The high wheat consumption in Turkey explains this situation significantly. Pb easily passes through the placenta by passive diffusion. Therefore, it is known that even low-dose Pb exposure may prevent fetal growth and potentially cause pregnancy complications such as LBW, abortion, and pre-term labor [31]. In the literature, it has been stated that Pb and Hg levels might vary and this variability could be related to lifestyle, age, BMI, and parity. However, we found no significant relationships between parity, educational level, BMI, and age and Pb, Cd, and Hg levels.

Arsenic was one of the most toxic elements listed in the substance priority list in 2015 by the Agency for Toxic Substances and Disease Registry. The toxicity generated by As is related to the biotransformation it undergoes in the body. This transformation is a methylation process [32]. Both the inorganic and methylated As forms easily pass through the placenta. Studies report increased As

▶ **Table 4** Change in toxic element levels according to age, BMI, and parity.

	Age (years)		BMI (kg/m²)		Parity	
	r	p	r	p	r	p
Al	0.163	0.124	0.060	0.575	0.036	0.740
Cr	−0.011	0.918	−0.071	0.505	0.037	0.730
As	0.125	0.239	0.049	0.647	−0.045	0.678
Cd	0.131	0.217	−0.015	0.885	0.060	0.579
Sb	0.165	0.119	0.000	1.000	−0.023	0.827
Hg	0.102	0.338	0.002	0.989	0.102	0.341
Pb	0.203	0.055	0.038	0.725	0.040	0.708

Al: aluminum; As: arsenic; Cd: cadmium; Cr: chromium; Hg: mercury; Pb: lead; Sb: antimony; BMI: body mass index.

methylation during pregnancy [16]. Therefore, the amount of As in the blood is found to be lower in the following weeks of gestation. Although it is known that there is As accumulation in the placenta, there are limited studies on the type and results of accumulation. Animal experiments indicate the presence of embryo toxicity and teratogenic effects; however, the kinetics and toxicity between species make it difficult to determine the effects on humans [33]. Nevertheless, available data show that abortion, stillbirth, pre-term labor, and birth defects are increased [15, 16, 34]. In the present study, inorganic and methylated As was evaluated as total, and As levels were found to be significantly higher in the study group. This can be associated with decreased blood levels due to increased methylation and increased placental involvement, especially in the later gestational weeks.

Sn is a heavy metal commonly used in the plastics and communications industries and various agricultural applications. Although it is present in the blood [35, 36], serum [37], and liver [38], there is no systematic study on tissue distribution. In animal experiments, some Sn derivatives have been shown to show a wide range of toxicity [39]. The effects include neurotoxicity, reproductive and developmental toxicity, hematologic effects, and immunotoxicity [39, 40]. Placental retention has been reported to be high in animal experiments [41]. However, information on placental elimination is not clear. Its effects in humans and its place in the etiology of abortion has not yet been clarified. Sn levels in the present study were found to be high and statistically significant in the SA group.

The idea of avoiding fetal toxicity caused by heavy metal exposure has led to attempts to reduce maternal exposure. However, these applications have produced different exposures and toxicities. Sb exposure has been brought up by attempts to avoid high-toxicity Pb. The development of 'Pb-free' materials has increased the use of Sb, and consequently bismuth (Bi) [42]. Sb is an element that is particularly used in the pharmaceutical field. Therefore, water and food contaminations are common. It is also taken into the body through inhalation. Sb is thought to produce endocrine effects by mimicking physiologic estrogen effects through estrogen receptors [43]. In a study, BMI was found to be a risk factor independent of age and gravity for Sb gestational diabetes [44]. Although studies indicated increased gestational diabetes and insulin resistance in pregnant women with high urinary Sb excretion, there are no studies that evaluated their relationship with the second-trimester abortion [44]. In the present study, no correlation was found between placental Sb levels and age, BMI, and parity. In multivariate logistic regression analysis, it was found that the toxic element that caused the highest increase in the risk of SA was Sb with an OR of 6.1.

Trace elements

Oxidative stress is regarded as a promoter for many complications in pregnancy such as defective embryogenesis, embryopathy, recurrent pregnancy loss, spontaneous abortion, fetal growth restriction, intrauterine fetal death, preeclampsia, and LBW [45]. Recent studies on the etiology of abortion have suggested that processes related to inflammation and oxidation could be effective. It is known that total antioxidant capacity decreases in women with recurrent pregnancy loss in the first trimester [46]. Glutathione peroxidase, thioredoxin reductase, and selenoprotein superoxide dismutase enzymes play a role in the oxidative process [47]. Trace elements such as Mn, Zn, Se, Co, and Cu act as co-factors in these enzymatic reactions. Decreased maternal serum levels of Se, Mn, and Cu were associated with abortion [10, 48, 49]. The role of trace elements in the etiology of abortion may be due to their relationship with antioxidant capacity.

Co is a trace element whose maternal concentration may vary during pregnancy. It is found bound to Vitamin B12 [50]. The effect of Co on pregnancy and placental functions is not clear. It has been reported that Co levels in pregnancy were negatively associated with pregnancy hypertension [51]. In a study on the relationship of Co with habitual abortions, no relationship was found [48]. Although it is well known that Co induces hypoxia by inhibiting proline hydroxylase in biologic systems, exact mechanisms for different toxicities in various organ systems have been studied. Increased use of Co, especially in orthopedic surgery and artificial manu-

facturing, has caused cytotoxic, genotoxic, and immunologic effects in patients, and has led to the development of conditions such as pseudo-tumor, an aseptic lymphocytic vasculitis associated with high inflammation [52]. In our study, the detection of higher levels of Co in the SA group can be explained with the formation of a hypoxic and inflammatory process in the placental environment.

Se is an important trace element that plays a role in redox hemostasis. The enzymes involved in redox hemostasis form a comprehensive antioxidant defense against the harmful effects of cell metabolism products. The most well-known enzymes here are glutathione peroxidases containing the Se atom, thioredoxin reductases, and selenoprotein P. In our study, placental Se levels were higher in the abortion group. The results of the studies on the relationship of Se with abortion vary. Some studies reported that low serum Se levels were associated with abortion [49, 53, 54]; however, there are also studies indicating no such relationship [55, 56]. Studies have reported that serum Se levels were lower in pregnant women compared with those in non-pregnant women. Researchers have associated this result with the growing Se need for the fetus [57–59]. When we evaluated the studies on placental Se levels, it was seen that results varied substantially. It was found to be 1.05 mg/g in Saudi Arabia [60], 0.81 mg/g in Spain [61], 0.15 mg/g in Croatia [62], and 0.64 mg/g in Poland [63]. Given the uneven environmental distribution of Se, it is not surprising that the placental content differs substantially on a global basis. This variability probably reflects differences in dietary exposure. Another reason was attributed to low socioeconomic status [64]. It was thought that abortion cases with low Se levels could also develop owing to a decrease in Se-based antioxidant capacity, but the exact mechanism is not known.

Mn is an essential element as well as being a potentially toxic element. It contributes to physiologic functions including bone formation, energy metabolism, reproduction, activation of certain enzymes, and protection against oxidative stress [65]. Dietary intake is sufficient in the general population; therefore, deficiency is uncommon. However, excessive intake is associated with neurotoxicity. Many studies have shown that high-dose exposure to Mn in childhood is associated with poor cognitive functions and hyperactive behavior [66–69]. Mn is a component of Mn superoxide dismutase (Mn-SOD), which protects cells against oxidative stress. Therefore, insufficient Mn-SOD activity may lead to the excessive generation of reactive oxygen species (ROS) in placental mitochondria. This can potentially cause placental cell damage, limiting the transport of nutrients and oxygen to the fetus [70]. Previous studies have shown that both low and high Mn levels decrease calcium (Ca) pumping activity, possibly due to lipid peroxidation in the cell membrane produced by ROS [71]. This mechanism reduces Ca in the placental membrane and deteriorates fetal bone mineralization and cell growth [72–74]. The deterioration of fetal development due to Mn occurs through these mechanisms. However, the mechanism of the formation of abortion is not clear. When evaluating Mn exposure, the maternal blood Mn level at birth is not a sufficient indicator of Mn exposure during pregnancy, because the biologic half-life of Mn in the blood is about 30 days [75]. Therefore, placental Mn levels at any stage of pregnancy offer a much better evaluation. The Mn levels in our study were found to be high and statistically significant in the SA group.

We are exposed to many environmental toxins and heavy metals at all times, regardless of occupation. The toxicodynamic and toxicokinetic properties of these substances vary for each individual. The homogeneity of our data increases because our working group consisted of non-smoking housewives who mostly lived in the city. However, since each of the heavy metals undergoes different processes in the body, it is a limitation of our study that other biologic materials such as blood and amnion other than the placenta were not evaluated. Another limitation is the lack of a genetic examination of abortion materials.

This study showed that many placental toxic elements and Sb, independently of these toxic elements, might increase the risk of second-trimester abortion. Furthermore, trace elements Mn, Se, and Co were found to be associated with abortion. Evaluating all toxic and trace element levels in the present study according to reference levels in the blood, we found that all were below the biologic exposure index. However, there is currently no internationally accepted index for placental heavy metal levels. It is therefore unclear at what levels these elements may have effects. Therefore, the present study may play a leading role in understanding the concentrations and distribution of toxic and trace elements in the placenta. Further studies may help to reveal the adverse effects of these metals on the fetus and life-long health.

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Conflict of Interest

The authors declare that they have no conflict of interest.

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