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THE IMPACT OF LIFESTYLE FACTORS ON AGE-RELATED DIFFERENCES IN HAIR TRACE ELEMENT CONTENT IN PREGNANT WOMEN IN THE THIRD TRIMESTER[*]

Anatoly V. Skalny[1,2,3,4], Alexey A. Tinkov[3,4]✉, Irina Voronina[5,6], Olga Terekhina[5], Margarita G. Skalnaya[4], Tatiana G. Bohan[5], Lyubov A. Agarkova[7], Yulia Kovas[5,8]

[1]All-Russian Research Institute of Medicinal and Aromatic Plants
Grina 7, Moscow 117216, **Russia**
[2]Institute of Bioelementology, Orenburg State University
Pobedy 13, Orenburg 460018, **Russia**
[3]Laboratory of Biotechnology and Bioelementology, Yaroslavl State University
Sovetskaya 14, Yaroslavl 150000, **Russia**
[4]Peoples' Friendship University of Russia (RUDN University)
Miklukho-Maklaya 6, Moscow 117198, **Russia**
[5]Department of Psychology of Personality, Tomsk State University
Lenin Av. 36, Tomsk 634050, **Russia**
[6]Psychological Institute, Russian Academy of Education
Mokhovaya 9–4, Moscow 125009, **Russia**
[7]Research Institute of Obstetrics, Gynecology and Perinatology, Tomsk National Research Medical Center
of the Russian Academy of Sciences, Kooperativny 5, Tomsk 634050, **Russia**
[8]Department of Psychology, Goldsmiths, University of London
Whitehead Building, New Cross, London SE14 6NW, **United Kingdom**

ABSTRACT

Background. Trace elements play a significant role in the regulation of human reproduction, while advanced age may have a significant impact on trace element metabolism. The objective of the present study was to assess the impact of lifestyle factors on age-related differences in hair trace element content in pregnant women in the third trimester.

Material and methods. A total of 124 pregnant women aged 20–29 ($n = 72$) and 30–39 ($n = 52$) were examined. Scalp hair trace element content was assessed using inductively coupled plasma mass spectrometry at NexION 300D (Perkin Elmer, USA) after microwave digestion.

Results. The results showed that the elder pregnant women had 36% ($p = 0.009$), 14% ($p = 0.045$), and 45% ($p = 0.044$) lower hair Zn, V, and Cd content, and 16% ($p = 0.044$) higher hair B levels – in comparison to the respective younger group values. Multiple regression analysis demonstrated that the age of the women had a significant influence on hair V and Zn levels. B content was also significantly influenced by age at first intercourse, smoking status, and specific dietary habits. None of the lifestyle factors were associated with hair Cd content in pregnant women. Hair V levels were also affected by following a special diet. Interestingly, alcohol intake did not have a significant impact on hair trace element content.

Conclusion. These data indicate that lifestyle factors have a significant influence on age-related changes in hair trace elements during pregnancy that may impact the outcome of pregnancy.

Keywords: pregnancy, age, zinc, boron, lifestyle

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✉tinkov.a.a@gmail.com, phone +79 619 378 198

INTRODUCTION

Trace elements play a significant role in the regulation of human reproduction (Pathak and Kapil, 2004). In particular, essential trace elements like zinc, selenium, iron, and copper are required for normal fetal development. In turn, deficiency of these metals and metalloids may be associated with an adverse pregnancy outcome (Gambling et al., 2003; Singh, 2004). Therefore, the pregnant maternal organism faces increased demand for trace elements (Hovdenak and Haram, 2012). At the same time, maternal overexposure to toxic trace elements and metals may have a negative impact on the birth outcome and further development of the offspring (Silbergeld and Patrick, 2005). Therefore, monitoring trace element status in pregnancy is of particular importance. Hair is successfully used for monitoring trace element status, although certain limitations exist (Chojnacka et al., 2010).

Existing data demonstrate that the risk of an adverse outcome increases with the age of pregnant women (Hoffman et al., 2007). In particular, it has been proposed that the lowest rate of adverse pregnancy outcomes was observed in women aged 20–29 years (Andersen et al., 2000). It has also been demonstrated that age has a significant influence on trace element content in different biological substrates (Clark et al., 2007; Skalnaya et al., 2016). However, the data are rather contradictory. It is therefore important to investigate whether age has a significant influence on hair trace element content in women.

The objective of the present study was to assess the association between age and hair trace element content in pregnant women in the third trimester, as well to reveal the impact of lifestyle factors on these parameters.

MATERIALS AND METHODS

The protocol of the study was approved by the Ethics Committee for Interdisciplinary Investigations at Tomsk State University/Psychological Institute of the Russian Academy of Education. The study was carried out in agreement with the principles of the Declaration of Helsinki of the World Medical Association (1964) and its later amendments. All the women took part in the investigation on a voluntary basis. All participants signed the informed consent form before the investigation and were informed about the procedures performed.

A total of 124 pregnant women were examined in the third trimester of pregnancy. All examinees were divided into two groups according to their age. Group 1 consisted of 72 pregnant women aged 20–29 years, whereas group 2 consisted of 52 women aged 30–39 years. The groups consisted of pregnant women living in the Siberian Federal District of the Russian Federation (Tomsk, Novosibirsk, and Barnaul) in similar proportions. Age and normal healthy pregnancy were used as the inclusion criteria. The following exclusion criteria were used in the study: 1) former occupational exposure to heavy metals or other chemicals, 2) the presence of metal implants, 3) the use of hormonal replacement therapy.

Prepregnancy values of anthropometric parameters (height, weight, body mass index) were recorded. In addition, all examinees filled in a questionnaire in order to obtain information on obstetric anamnesis, as well as lifestyle parameters (Table 1). All women used vitamin/mineral supplements from different manufacturers, therefore, this parameter was not included into descriptive statistics and multiple linear regression models. Data on iron supplementation is specified in Table 1.

Proximal parts (1–1.5 cm) of occipital scalp hair strands (0.05–0.1 g) were collected in the third trimester of pregnancy. These hair samples were washed with acetone, rinsed three times with distilled deionized water and dried in air. Microwave digestion of the samples was performed in a BerghofSpeedwave4 DAP-40 (Berghof Products and Instruments, Germany) system at 170–180°C for 20 minutes. After cooling, the resulting solutions were transferred into test-tubes with the addition of polypropylene with the subsequent addition of distilled deionized water to a total volume of 15 ml. Hair trace element content [μg/g dry weight] was assessed using inductively coupled plasma mass spectrometry at NexION 300D (PerkinElmer Inc., Shelton, CT, USA). Internal standards containing 10 μg/l yttrium-89 and rhodium-103 prepared from Yttrium (Y) and Rhodium (Rh) Pure Single-Element Standards (PerkinElmer Inc., Shelton, CT, USA) were used for internal online standardization. The certified reference material of human hair GBW09101 (Shanghai, China)

Table 1. Anthropometric data, obstetric anamnesis, and lifestyle parameters of the women

Parameter	Age		*P* value
	20–29 (*n* = 72)	30–39 (*n* = 52)	
Age, years	26.43 ±2.13	33.31 ±3.07	<0.001*
Married, % (*n*)	97% (69/72)	83% (43/52)	0.256
Cohabiting, % (*n*)	3% (3/72)	17% (9/52)	0.104
Years married	2.85 ±1.73	5.68 ±4.06	<0.001*
College, % (*n*)	7% (5/72)	21% (11/52)	0.133
University, % (*n*)	92% (66/72)	77% (40/52)	0.221
Other, % (*n*)	1% (1/72)	2% (1/52)	0.435
Income, ×1000 RUR	55.2 ±25.6	61.8 ±31.3	0.219
Height, cm	165.51 ±6.53	164.71 ±6.11	0.435
Weight, kg	61.56 ±10.78	65.69 ±16.47	0.604
BMI	22.43 ±3.46	24.18 ±5.72	0.364
Age at menarche, years	13.06 ±1.39	13.22 ±1.22	0.636
Age at first sex, years	17.70 ±2.46	18.14 ±2.87	0.973
Frequency of sex, per week	2.25 ±0.76	2.29 ±1.08	0.662
First pregnancy, % (*n*)	50% (36/72)	21% (11/52)	0.007*
Having a miscarriage, % (*n*)	13% (9/72)	21% (11/52)	0.394
Number of previous miscarriages	0.15 ±0.36	0.24 ±0.51	0.643
Having a termination, % (*n*)	24% (17/72)	33% (17/52)	0.361
Number previous terminations	0.35 ±0.73	0.63 ±0.98	0.199
Planning the pregnancy, % (*n*)	86% (62/72)	73% (38/52)	0.276
Alcohol consumption a year before, % (*n*)	85% (61/72)	75% (39/52)	0.438
Frequency of alcohol consumption, per week	2.97 ±1.32	2.67 ±1.61	0.586
Alcohol consumption during pregnancy, % (*n*)	19% (14/72)	23% (12/52)	0.703
Smoking a year before, % (*n*)	31% (22/72)	31% (16/52)	0.941
Number of cigarettes, per day	1.03 ±1.64	0.94 ±1.53	0.919
Smoking during pregnancy, % (*n*)	3% (2/72)	15% (8/51)	0.307
Special diet (vegetarianism)	7% (5/72)	17% (9/52)	0.293
Consumption of fruit and vegetables, per week	4.58 ±1.10	4.84 ±1.10	0.703
Fe supplementation, % (*n*)	31% (22/72)	42% (22/52)	0.154

*Significant group difference at $p < 0.05$ (Mann-Whitney U-test).

was used for laboratory quality control with the recovery rate >90%.

Statistica 10.0 (Statsoft, Tulsa, OK, USA) software was used for statistical treatment of the data. Data distribution was not Gaussian for all the trace elements as assessed by the Shapiro-Wilk test. Group values were expressed as a median and the respective 25–75 percentile boundaries. The significance of group differences was assessed using the Mann-Whitney U-test. Multiple linear regression analysis was used in order to assess the impact of various factors on hair trace element levels that were significantly different between the groups. A level of significance of $p < 0.05$ was set for all statistical analyses performed.

RESULTS

The results showed that among all the essential trace elements studied (Table 2) only hair Zn and V content were significantly lower in the older age group

Table 2. Hair trace element content in pregnant women aged 20–29 (group 1) and 30–39 (group 2) years, µg/g

Element	Group		*P* values
	1 (*n* = 72)	2 (*n* = 52)	
Al	4 (2.4–6.9)	3.8 (2.2–6.1)	0.345
As	0.011 (0.006–0.016)	0.008 (0.006–0.015)	0.245
B	0.308 (0.238–0.425)	0.358 (0.277–0.471)	0.044*
Ba	3.3 (2.2–5.6)	4.2 (2.5–6.8)	0.257
Cd	0.011 (0.005–0.016)	0.006 (0.003–0.014)	0.044*
Co	0.019 (0.01–0.044)	0.018 (0.012–0.046)	0.735
Cr	0.091 (0.051–0.265)	0.082 (0.056–0.162)	0.798
Cu	16.4 (12.1–26.5)	16.4 (11.4–29.4)	0.894
Fe	17.7 (11–26.5)	14.3 (10.3–23.1)	0.395
Hg	0.291 (0.188–0.449)	0.286 (0.178–0.446)	0.692
I	0.344 (0.255–0.615)	0.417 (0.277–0.646)	0.341
Li	0.01 (0.006–0.013)	0.008 (0.003–0.014)	0.225
Mn	1.08 (0.7–1.98)	1.26 (0.57–2.3)	0.688
Ni	0.299 (0.206–0.44)	0.296 (0.18–0.418)	0.727
Pb	0.378 (0.255–0.679)	0.34 (0.206–0.498)	0.115
Se	0.362 (0.286–0.445)	0.383 (0.286–0.508)	0.529
Si	33 (20–48)	41 (29–54)	0.129
Sn	0.162 (0.083–0.588)	0.142 (0.078– 0.507)	0.586
Sr	7.1 (4.7–11.7)	8.4 (5.7– 13)	0.276
V	0.011 (0.006–0.018)	0.007 (0.005–0.012)	0.009*
Zn	252 (203–350)	218 (185–292)	0.035*

Data presented as Median (25–75).
*Group difference is significant at $p < 0.05$.

in comparison to the respective younger group values by 36% and 14%. No significant effect of age on hair Co, Cr, Cu, Fe, I, Li, Mn, Se, and Si content was found. Similarly, among all the toxic trace elements studied only hair B and Cd values were significantly different between the groups. In particular, pregnant women from the older age group were characterized by a significant 16% increase in hair B levels compared to the younger group's values. Conversely, a significant age-associated 45% decrease in hair Cd content was observed in the pregnant women. At the same time, the values of hair Al, As, Ba, Hg, Ni, Pb, Sn, and Sr level were not significantly different between the groups.

Multiple regression analysis (Table 3) demonstrated that hair B content was significantly predicted by age, age at first intercourse, smoking a year before pregnancy, the number of cigarettes per day in a year before pregnancy, and following a specific dietary habits (vegetarianism). None of the lifestyle factors were associated with hair Cd content in pregnant women. It is also notable that alcohol intake both a year before and during pregnancy did not have a significant impact on hair B, Cd, V, and Zn content.

The results of multiple linear regression analysis demonstrated that the models significantly accounted for 28% and 11% of hair B and V content variance in the pregnant women. The absence of the model

Table 3. Multiple linear regression models with hair B, Cd, V, and Zn content in pregnant women as dependent variables

Parameter	B		Cd		V		Zn	
	β	*p*	*β*	*p*	*β*	*p*	*β*	*p*
Age	0.268	0.015*	–0.165	0.208	–0.235	0.048*	–0.243	0.048*
Height, cm	–0.024	0.957	0.692	0.193	0.198	0.687	–0.729	0.179
Weight, kg	0.313	0.791	–1.792	0.212	–0.455	0.731	1.965	0.179
Body mass index	–0.278	0.802	1.716	0.200	0.437	0.722	–1.831	0.177
Age at menarche	–0.165	0.067	–0.110	0.303	0.047	0.635	0.096	0.357
Age at first intercourse	0.393	<0.001*	0.061	0.595	0.184	0.085	0.214	0.056
First pregnancy	0.179	0.097	–0.030	0.819	0.188	0.123	–0.153	0.230
Having a miscarriage	0.107	0.606	–0.032	0.898	–0.041	0.861	–0.140	0.572
Number of previous miscarriages	–0.158	0.437	–0.002	0.993	–0.012	0.958	–0.008	0.974
Having a termination	0.092	0.585	0.136	0.507	–0.047	0.805	–0.125	0.536
Number of previous terminations	0.032	0.851	–0.048	0.818	–0.002	0.990	0.073	0.723
Planning the pregnancy	0.043	0.617	0.006	0.957	0.065	0.497	–0.008	0.937
Smoking a year before	0.580	0.038*	0.039	0.908	–0.088	0.777	–0.526	0.112
Number of cigarettes	–0.482	0.044*	–0.029	0.928	0.074	0.804	0.530	0.099
Smoking during pregnancy	0.069	0.490	0.195	0.107	0.036	0.751	–0.003	0.980
Alcohol consumption a year before	–0.123	0.621	–0.305	0.302	–0.168	0.526	–0.148	0.607
Frequency of alcohol consumption	–0.167	0.494	0.250	0.384	0.348	0.182	0.135	0.631
Alcohol consumption during pregnancy	–0.037	0.694	–0.071	0.532	0.009	0.933	0.099	0.376
Consumption of fruit and vegetables	0.045	0.595	0.023	0.823	0.038	0.697	0.052	0.609

significance for Cd and Zn indicates that the lifestyle factors used as independent variables do not have any substantial influence on hair Cd and Zn. It is possible that other factors that were not included in the present study (e.g., environmental, occupational) have a stronger influence on these trace elements. The age of the examinees and following a special diet had a significant influence on hair V levels. Although the model for hair Zn content was not significant, the age of women had a significant impact on this parameter.

DISCUSSION

The results suggest that older age in pregnant women is associated with increased hair B, and decreased hair Cd, V, and Zn. These results are consistent with an earlier investigation that demonstrated a significant decrease in hair Cd and V levels in women aged 20–30 years in comparison to older women (30–40 years; Skalnaya et al., 2016). Similarly, both hair Zn and Cd were characterized by a significant decrease in adolescents in comparison to children (Peña-Fernández et al., 2016). At the same time, no significant group difference in hair Zn levels was detected (Skalnaya et al., 2016). Another study using similar age groups found higher levels of hair Cd in older women (Khalique et al., 2005). Some previous studies did not find age-related differences in hair Zn levels (Khalique et al., 2005; Skalnaya et al., 2016). Another study in female Koreans aged 20–49 and above 50 years old revealed a significant age-associated decrease in hair Zn content, but an increase in Cd levels (Hong et al., 2009). It is clear that more research is needed to clarify the sources of inconsistencies in the literature. Hypothetically, the inconsistencies observed may result from a different rate of pregnancy in the groups enrolled in different studies as pregnancy significantly affects trace element metabolism. At the same time, geographical and national factors may also underpin existing contradictions.

Human studies regarding age-related changes in hair boron content are quite limited. Animal research has demonstrated a significant age-related increase in hair B levels in rats, but an opposite pattern of age-related decrease in hair B levels was observed in marmosets. The same study found a significant decrease in hair Cd levels in elder primates, as well as the absence of significant group differences in hair V and Zn levels between younger and older ones (Ambeskovic et al., 2013).

Altered trace element status in pregnancy may have a significant impact on pregnancy outcome. In particular, zinc requirements are significantly increased in pregnancy (Fung et al., 1997). Lower zinc intake in pregnancy was associated with preterm birth (Scholl et al., 1993).

The results of the multiple regressions are only partly consistent with previous research. For example, in contrast to previous studies (King, 2000), we did not find any significant influence of smoking, alcohol intake and iron supplementation on zinc status.

A limited number of human studies demonstrated the interrelation between boron exposure and pregnancy outcome. In particular, in a cohort of boron workers a higher number of delays in pregnancy and lower total number of live births was detected in comparison to unexposed women (Chang et al., 2006). Similarly, a recent study demonstrated that increased boron levels in the organism are associated with lower birth weight (Igra et al., 2016).

The effects of low vanadium status in pregnant women are not studied, whereas animal studies demonstrated that low vanadium is associated with increased infant mortality (Valko and Moncol, 2009). It is also expected that vanadium deficiency during pregnancy will result in diabetes (Wu et al., 2012). At the same time, it should be mentioned that a greater number of studies demonstrated the association between vanadium exposure and impaired reproductive function and fetal growth (Apostoli and Catalani, 2010).

Generally, the results of the current study demonstrate that increased age is associated with significant change in several hair trace element content in pregnant women. However, much research is still needed to replicate the results and to identify sources of inconsistencies in the literature. Gaining better insights into age-related changes in trace elements levels may help to prevent adverse pregnancy outcomes.

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