

THE EFFECT OF ALCOHOL CONSUMPTION ON MATERNAL AND CORD BLOOD ELECTROLYTE AND TRACE ELEMENT LEVELS*

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ABSTRACT

Background. Earlier studies demonstrated that alcoholism significantly alters electrolyte and trace element homeostasis. However, the existing data on the interplay between maternal alcohol consumption and fetal trace element status are contradictory. Therefore, the primary objective of the present study was to assess the influence of alcohol consumption on maternal and cord blood trace elements.

Material and methods. A total of 30 pregnant women (15 women consuming alcohol and 15 controls) were examined. Assessment of electrolyte and trace elements concentration in maternal (1 and 3 trimesters) and umbilical cord blood was performed using inductively-coupled plasma mass spectrometry.

Results. In the first trimester of pregnancy alcohol consumption is associated with increased whole blood Ca and Na levels. In the third trimester of pregnancy, women consuming alcohol are characterized by significantly increased Co levels. Conversely, the level of Co and Mn in the cord blood of offspring maternally exposed to ethanol is decreased. A significant correlation between first trimester blood and cord blood concentrations of K was revealed both in control women and those consuming alcohol. In the third trimester of pregnancy in the control women, a significant correlation with cord blood was detected for Fe, Mg, P, and Pb. Oppositely, in the third trimester in women consuming alcohol we detected a close association between maternal whole blood and cord blood levels for Ca, Cd, and Pb.

Conclusion. The data obtained demonstrate that maternal alcohol consumption results in fetal Co and Mn deficiency.

Key words: alcohol, cobalt, manganese, pregnancy, fetal alcohol syndrome

*The current investigation is supported by the Russian Foundation for Basic Research within project №15-04-08621.

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INTRODUCTION

Alcoholism or alcohol dependence syndrome (F10.2-ICD-10) is a serious public health concern (Degenhardt et al., 2008). Special attention is given to alcoholism in pregnant women (Ethen et al., 2009). Particularly, maternal perinatal alcohol consumption is closely associated with fetal neurophysiologic disturbances (Riley et al., 2011).

The existing data demonstrate that alcoholism significantly alters electrolyte and trace element homeostasis. An examination of alcoholic cirrhosis patients revealed significantly decreased serum and hepatic zinc content in parallel with elevated copper levels (Zarski et al., 1985). A significant association between alcohol dependence syndrome and excessive iron, copper and nickel was detected (Cook et al., 1991).

Taking into account the role of essential trace elements in embryogenesis (Fall et al., 2003) as well as the embryotoxic effect of toxic heavy metals (Thompson and Bannigan, 2008), one can suppose that fetal alcohol syndrome may be at least partially mediated by an alcohol-induced trace element imbalance. Moreover, it has been noted that altered trace elements status may be associated with a poor developmental outcome in children (Pathak and Kapil, 2004).

Despite the presence of multiple investigations of the interplay between alcoholism and trace element status, the existing data are contradictory. Therefore, the primary objective of the present study was to assess the influence of chronic alcohol consumption on maternal and cord blood trace elements.

MATERIAL AND METHODS

All procedures performed in the present study were in accordance with the ethical standards of the institutional and national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Informed consent was obtained from all individual participants included in the study.

A total of 30 pregnant women were enrolled in the current study. Alcohol consumption was investigated using a questionnaire. Only women with a moderate and high alcohol intake (self-reported: more than one drink per day) who continued drinking during

pregnancy ($n = 15$) were examined in the alcohol group, according to a previously published classification of alcohol consumption (Serwin et al., 2002). 15 age-matched pregnant women who do not consume alcohol were included in the control group. The mean age in the alcohol and control groups was 26 ± 6 and 24 ± 4 years old, respectively. Only women with normal pregnancy (as assessed in the maternity hospital) were investigated in both groups. In order to avoid the influence of side factors on electrolyte and trace element status, the following exclusion criteria were used: metal implants (including dental amalgam fillings), endocrine disorders, acute traumas, inflammatory and infectious diseases, occupational exposure to heavy metals (current and former), using dietary mineral supplements, current or former smoking, excessive fish consumption, and vegetarian diet. Blood samples from pregnant women were collected in the first (3rd month) and third trimesters (8th month). Fasting maternal blood and umbilical cord blood samples were collected using «S-Monovette» (Sarstedt, Nümbrecht, Germany) and trace element-free equipment in maternity welfare centre and maternity hospital, respectively.

The samples were degraded in a Berghof Speed-wave 4 microwave digestion system (Berghof Products & Instruments, Germany) at 180°C for 20 minutes in concentrated HNO₃.

Assessment of electrolyte and trace elements concentration in maternal whole blood and umbilical cord blood was performed using inductively-coupled plasma mass spectrometry at NexION 300D (PerkinElmer Inc., Shelton, USA). Laboratory quality control was performed using the certified ClinChek® reference materials levels 1 and 2 (Recipe, Munich, Germany). The procedure was performed both before and after a series of analyses. The recovery rates for all electrolytes and trace elements exceeded 80%.

All data were stored in MS Excel calculation sheet (Microsoft Corp, USA). Statistical treatment of this data was performed using Statistica 10 (Statsoft, Tulsa, OK, US). The Shapiro-Wilk test revealed Gaussian distribution of data on electrolyte and essential trace element concentrations in whole blood. These data were expressed as a mean and the respective standard deviations (SD). Group comparison was performed using one-way ANOVA. In contrast, the values of

toxic trace elements content were not normally distributed and were expressed as median and 25 and 75 percentile boundaries. The Mann-Whitney U-test was used for group comparison of these data. Correlation analysis was performed using Spearman's coefficient of rank correlation. The level of significance was set as $p < 0.05$ for all analyses.

RESULTS

The data obtained demonstrate that alcohol consumption significantly altered maternal whole blood electrolyte content (Table 1). In particular, a significant increase of 10% and 9% in whole blood Ca and Na, respectively, was detected in women consuming excessive amounts of alcohol in the first trimester of pregnancy. At the same time, no significant changes

in whole blood trace elements levels were detected. In contrast to the first trimester, no significant group difference in whole blood electrolytes was revealed in the third trimester (Table 1). However, women consuming alcohol were characterized by a significant 17% increase in whole blood Co levels as compared to the control values. No marked difference in the concentration of other trace elements was detected. When compared to the values obtained in the first trimester, no significant changes were detected for control women in the third trimester. At the same time, in women consuming alcohol whole blood Mn significantly increased by 39% from the first to the third trimester of pregnancy. In contrast, the concentration of Ni in 3rd trimester was characterized by a significant 32% decrease compared to the respective first trimester values.

Table 1. Whole blood electrolyte and trace elements concentration in pregnant women in 1 and 3 trimesters, $\mu\text{g/ml}$

Element	1 trimester		3 trimester	
	control	alcohol	control	alcohol
Ca	59.782 \pm 6.950	65.619 \pm 5.155*	57.152 \pm 5.465	60.638 \pm 7.136
K	1 941.546 \pm 627.051	2 081.867 \pm 459.492	1 867.775 \pm 352.597	1 832.507 \pm 461.462
Mg	30.826 \pm 3.991	30.950 \pm 2.258	29.152 \pm 2.833	30.531 \pm 3.044
Na	2 187.471 \pm 217.986	2 381.633 \pm 173.413*	2 189.786 \pm 165.929	2 274.933 \pm 225.432
P	333.029 \pm 38.892	337.083 \pm 32.313	325.468 \pm 40.072	342.180 \pm 31.592
Co	0.0012 \pm 0.0003	0.0012 \pm 0.0002	0.0012 \pm 0.0002	0.0014 \pm 0.0003*
Cu	1.315 \pm 0.188	1.405 \pm 0.264	1.410 \pm 0.168	1.367 \pm 0.185
Fe	412.204 \pm 85.347	392.580 \pm 40.898	385.882 \pm 89.002	402.157 \pm 63.610
Mn	0.022 \pm 0.007	0.018 \pm 0.005	0.024 \pm 0.006	0.025 \pm 0.008**
Se	0.151 \pm 0.029	0.161 \pm 0.031	0.157 \pm 0.029	0.158 \pm 0.031
Zn	5.542 \pm 0.925	5.423 \pm 0.957	5.622 \pm 0.940	5.887 \pm 1.068
Cd	0.0003 (0.0002–0.0004)	0.0003 (0.0002–0.0011)	0.0003 (0.0002–0.0004)	0.0003 (0.0002–0.0007)
Ni	0.0085 (0.0056–0.0103)	0.0090 (0.0061–0.0130)	0.0053 (0.0047–0.0071)	0.0061 (0.0050–0.0067)**
Pb	0.016 (0.014–0.022)	0.013 (0.012–0.017)	0.015 (0.011–0.025)	0.017 (0.015–0.026)

Data expressed as mean \pm SD (electrolytes and essential trace elements) and median (25–75) (toxic trace elements).

*Significant difference in comparison to the control values at $p < 0.05$.

**Significant difference in comparison to the 1 trimester values at $p > 0.05$.

p value is indicated in accordance to one-way ANOVA (minerals and essential trace elements) and Mann-Whitney U-test (toxic trace elements).

These data demonstrate that the cord blood electrolyte level was rather stable (Table 2). Despite a 7% decrease in cord blood Ca levels in the alcohol-exposed group, these changes were only slightly significant as compared to the control values. At the same time, maternal alcohol consumption resulted in a significant 7% and 32% decrease in cord blood Co and Mn levels in comparison to the control values. The level of other trace elements was not affected by maternal alcohol consumption.

Correlation analysis (Table 3) revealed a significant direct relationship between whole blood Cd and Se in the 1st and 3rd trimesters of pregnancy in the control examinees. As was the case with the control-group women, a close relationship between whole blood Cd levels in trimesters 1 and 3 was detected in persons with excessive alcohol consumption. Despite the absence of any significant group difference in the

whole blood zinc concentration, a direct relationship between the 1st and 3rd trimester values was detected in alcoholic women.

A significant association between cord blood and maternal whole blood electrolyte content was also revealed (Table 3). In particular, first trimester whole blood K levels in the control women were significantly associated with those in cord blood. In the third trimester of pregnancy, a significant correlation with cord blood was detected for Fe, Mg, P, and Pb whole blood concentrations. Likewise, whole blood K levels in the first trimester of pregnancy in women consuming excessive amounts of alcohol were directly associated with cord blood electrolyte content. Conversely, in the third trimester we detected a close association between maternal whole blood and cord blood levels for Ca, Cd, and Pb.

Table 2. The level of electrolytes and trace elements in umbilical cord blood of alcohol-exposed and non-exposed newborns, µg/ml

Element	Control	Alcohol
Ca	61.623 ±6.335	56.897 ±7.064
K	3 230.679 ±1 527.103	2 880.200 ±1 232.053
Mg	33.476 ±3.163	32.156 ±3.266
Na	1 970.286 ±152.351	1 926.500 ±260.733
P	428.304 ±67.820	396.140 ±64.413
Co	0.0016 ±0.0006	0.0013 ±0.0006*
Cu	0.566 ±0.161	0.638 ±0.282
Fe	536.843 ±53.216	506.083 ±124.674
Mn	0.059 ±0.027	0.040 ±0.011*
Se	0.162 ±0.026	0.162 ±0.027
Zn	2.360 ±0.591	2.720 ±1.151
Cd	0.0001 (0.0000–0.0002)	0.0001 (0.0000–0.0002)
Ni	0.0119 (0.0045–0.0157)	0.0057 (0.0042–0.0101)
Pb	0.014 (0.010–0.017)	0.014 (0.012–0.018)

Data expressed as mean ±SD (electrolytes and essential trace elements) and median (25–75) (toxic trace elements).

*Significant difference in comparison to the control values at $p < 0.05$ in accordance to one-way ANOVA (minerals and essential trace elements) and Mann-Whitney U-test (toxic trace elements).

Table 3. Correlation between whole blood electrolyte and trace elements concentration in women at 1 and 3 trimesters and maternal and cord blood, µg/ml

Element	1 vs 3 trimester		1 trimester vs cord		3 trimester vs cord		
	control	alcohol	control	alcohol	control	alcohol	
Ca	<i>r</i>	0.104	0.174	0.403	0.209	-0.173	0.684
	<i>p</i>	0.725	0.535	0.153	0.454	0.555	0.005*
K	<i>r</i>	0.050	0.375	0.660	0.694	0.129	0.315
	<i>p</i>	0.865	0.168	0.010*	0.004*	0.660	0.252
Mg	<i>r</i>	0.242	0.494	0.413	-0.173	0.563	0.229
	<i>p</i>	0.405	0.062	0.142	0.537	0.036*	0.412
Na	<i>r</i>	-0.003	-0.067	0.373	0.351	-0.058	0.296
	<i>p</i>	0.991	0.812	0.189	0.200	0.845	0.284
P	<i>r</i>	0.357	0.227	0.446	-0.188	0.648	0.107
	<i>p</i>	0.210	0.417	0.110	0.503	0.012*	0.705
Cd	<i>r</i>	0.843	0.718	-0.180	0.415	0.042	0.721
	<i>p</i>	<0.001*	0.003*	0.537	0.124	0.888	0.002*
Co	<i>r</i>	0.471	0.419	-0.096	-0.169	-0.198	0.307
	<i>p</i>	0.090	0.120	0.744	0.548	0.498	0.266
Cu	<i>r</i>	-0.044	-0.036	-0.357	0.369	-0.635	-0.306
	<i>p</i>	0.882	0.900	0.210	0.175	0.015*	0.268
Fe	<i>r</i>	0.545	0.269	0.361	0.116	0.634	0.484
	<i>p</i>	0.044*	0.333	0.205	0.680	0.015*	0.067
Mn	<i>r</i>	0.579	0.237	0.226	0.087	0.386	-0.173
	<i>p</i>	0.030*	0.396	0.436	0.757	0.172	0.537
Ni	<i>r</i>	-0.060	-0.160	-0.208	-0.438	0.077	0.099
	<i>p</i>	0.838	0.569	0.475	0.102	0.793	0.727
Pb	<i>r</i>	0.427	-0.161	0.682	-0.136	0.737	0.809
	<i>p</i>	0.128	0.566	0.007*	0.630	0.003*	<0.001*
Se	<i>r</i>	0.573	-0.224	0.012	0.002	-0.274	0.240
	<i>p</i>	0.032*	0.421	0.968	0.994	0.344	0.389
Zn	<i>r</i>	0.136	0.610	0.012	-0.245	-0.335	-0.175
	<i>p</i>	0.643	0.016*	0.968	0.378	0.242	0.534

*Correlation is significant at $p < 0.05$.

DISCUSSION

Our data demonstrate that excessive alcohol consumption affects concentrations of maternal and cord blood electrolytes and trace elements. At the same time, in the first trimester of pregnancy, the most prominent changes were detected in whole blood electrolyte levels (Ca and Na), whereas in the third trimester trace element levels were affected (Co and Mn). Moreover, maternal whole blood trace element levels in the third trimester were significantly associated with cord blood concentrations.

The existing data demonstrate that alcohol intake results in decreased serum Ca levels (Jorde et al., 2001) and intestinal Na transport (Mekhjjan et al., 1975), in contrast to our findings. It is hypothesized that the interaction between pregnancy and alcohol consumption may have resulted in the increase observed.

The most interesting data were obtained for whole blood cobalt. In particular, the whole blood cobalt concentration in the third trimester of pregnancy in women with excessive alcohol consumption was significantly elevated in comparison to the control values. Conversely, maternal ethanol consumption was associated with decreased umbilical cord blood Co. The existing data on Co status in relation to alcohol consumption are limited. However, Co plays a structural role in B₁₂ vitamin and the level of the latter is closely related to Co status. The most recent review demonstrated that changes in B₁₂ levels are multidirectional, ranging from decreased concentrations to elevation of vitamin concentrations (Fragasso, 2013). The increase observed in whole blood Co concentrations in alcohol-abusing women is generally in agreement with the increased plasma B₁₂ levels in alcohol abuse patients (Cylwik et al., 2010) and alcoholic liver disease patients (Baker et al., 1998). It has been proposed that alcoholic liver disease results in B₁₂ redistribution and decreased vitamin stores (Baker et al., 1998). Taking into account the association between increased blood B₁₂ levels and decreased vitamin stores, one can suppose that the decrease observed in cord blood B₁₂ concentration may be indicative of decreased vitamin transfer to the fetus. Cobalamin deficiency is associated with a variety of clinical signs, including anemia and neurological disorders (Carmel, 2000). In turn, cobalt

itself stimulates erythropoietin production through stabilization of hypoxia-induced factor and mimicking hypoxic conditions (Simonsen et al., 2012).

Another important finding of the study is decreased umbilical cord blood Mn content after maternal ethanol consumption. The majority of studies demonstrated a significant association between ethanol and Mn exposure and certain biological functions (Bouchard et al., 2003). At the same time, the influence of alcohol consumption on Mn status has not been subject to sufficient research. In particular, earlier experimental studies demonstrated a positive influence of alcohol consumption on tissue Mn concentrations (Shukla et al., 1978). Conversely, we have detected a significant decrease in the cord blood Mn concentration in newborns maternally exposed to ethanol. It is also notable that Mn deficiency is significantly associated with fetal intrauterine growth retardation (Wood, 2009).

Surprisingly, we have failed to detect a significant change in iron, copper, zinc, and selenium levels, in contradiction to earlier indications (Arinola, 2008; Cook et al., 1991; González-Reimers et al., 2010; Skalny, 1990; Skalny and Skosyeva, 1987; Zarski et al., 1985). Earlier studies also demonstrated that certain alcoholic drinks may be contaminated with toxic trace elements (Sherlock et al., 1986). At the same time, we did not observe a significant increase in heavy metal levels in either maternal or cord blood. However, a significant correlation for Cd was detected only for alcohol-consuming women and their children. It is supposed that this observation may be associated with ethanol-induced changes in placental development and impaired placental transfer of certain compounds (Burd et al., 2007).

Therefore, the data demonstrate obtained:

- in the first trimester of pregnancy, alcohol consumption is associated with increased whole blood Ca and K levels as compared to the control values
- in the third trimester of pregnancy, women consuming alcohol are characterized by significantly increased Co levels
- the level of Co and Mn in cord blood of offspring maternally exposed to ethanol is decreased when compared to the control values.

The data obtained demonstrate that altered trace element and electrolyte status during pregnancy may be associated with the low trace element status of the

newborn. These changes may at least partially contribute to the development of fetal alcohol syndrome. However, further studies are required to assess the intimate mechanisms of ethanol-induced alteration of trace element and electrolyte levels, as well as the role of these micronutrients in fetal alcohol spectrum disorder development.

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