



Original Research Article

Antioxidant status in women who have had a miscarriage



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ABSTRACT

Purpose: During normal pregnancy there is an oxido-reductive balance between action of pro-oxidative factors. The aim of this study was to evaluate the total antioxidant status (TAS) and glutathione peroxidase (GSH-Px) activity, and the content of selenium (Se), zinc (Zn), copper (Cu) and manganese (Mn) in women who have had a miscarriage.

Patients and methods: The study group consisted of 83 women who had had miscarriages. The control group included 35 women in the first trimester of pregnancy, and 35 pregnant women after childbirth. **Results:** TAS activity and Cu concentration in serum in women who experienced a miscarriage were significantly lower, but Mn level – higher, than in women in the first trimester of pregnancy. The content of Se, Cu and Mn in placental tissue in patients who have had a miscarriage was significantly higher, while Zn content was lower than in pregnant women at full-term delivery.

Conclusions: Our findings, provided in the research, enable us to claim that the total antioxidative status is significantly lower in women who have had a miscarriage. Low level of Zn but high of Mn in the examined biological material may be indicative to the incidence of miscarriage.

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1. Introduction

Miscarriage is the spontaneous end of a pregnancy at a stage where the embryo or fetus is incapable of surviving independently [1]. During normal pregnancy there is an oxido-reductive balance between activity of pro-oxidative factors, like free radicals, and competence of antioxidative systems, in which Se, Zn, Mn and Cu are present and which are ingredients of enzymes in the first line of defense taking part in expelling free radicals. There are two types of antioxidants in the human body: enzymatic antioxidants (superoxide dismutase, catalase, glutathione peroxidase and glutathione reductase) and non-enzymatic antioxidants are influenced by dietary intake (vitamin C, vitamin E, Se, Zn, Mn, Cu, taurine, hypotaurine, glutathione, β -carotene and carotene) [2,3].

An important increase of constitutional oxidative stress markers is connected with advanced and irreversible stadium of

abortion which does not appear in mild pregnancy disorders. There is a lot of literature on the effects of oxidative stress in the female reproduction with involvement in the pathophysiology of miscarriage [4–6].

Deficiencies of some antioxidant activities are associate with the trace elements: Se, Zn, Cu and Mn can result in poor pregnancy outcomes, including: fetal growth restriction, preeclampsia and are associate with increased risk of civilization diseases in adulthood [7–10].

The aim of this study was to evaluate the activity of total antioxidant status (TAS) and glutathione peroxidase (GSH-Px), the content of selenium (Se), zinc (Zn), copper (Cu) and manganese (Mn) in women who have had a miscarriage.

2. Patients and methods

2.1. Study groups

The study material was the venous blood and placental tissue fragments collected during surgical procedures from 83 women who have had a miscarriage aged 18–44 (30 years; BMI 22.43 kg/m²; 8.9 week of pregnancy). We obtained samples from patients of

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Table 1
TAS and GSH-Px activity, Se, Zn, Cu and Mn concentration in serum.

No.	Parameters and trace elements	Reference range*	Control group (A) n=35 Me (Q ₁ –Q ₃)	Women who have had a miscarriage n=83 Me (Q ₁ –Q ₃)	p
1	TAS (mmol/L)	1.30–1.77	2.925 (2.680–3.066)	2.434 (2.264–2.540)	<0.00002
2	GSH-Px (U/L)	4171–10,881	4308.2 (3445.9–5219.7)	6724.1 (5172.1–8276.1)	<0.002
3	Se (μg/L)	70–140	51.61 (46.76–55.05)	57.57 (48.68–64.98)	<0.04
4	Zn (mg/L)	0.6–1.2	0.587 (0.553–0.796)	0.571 (0.522–0.664)	ns
5	Cu (mg/L)	0.8–1.2	1.461 (1.249–1.628)	1.107 (0.975–1.370)	<0.002
6	Mn (μg/L)	0.4–1.2	0.817 (0.492–1.106)	1.291 (0.687–1.760)	<0.008

Me – median; Q₁ – lower quartile; Q₃ – upper quartile; TAS – total antioxidant status; GSH-Px – glutathione peroxidase; A – control group (women in the first trimester of pregnancy).

p < 0.05 – statistical significance; ns – not significant.

* – reference range [24].

the Department of Perinatology of the Medical University of Białystok Clinical Hospital and Division of Obstetrics and Pathology of Pregnancy SPZOZ District Hospital in Białystok in the years 2007–2011.

The control group included 35 pregnant women aged 18–31 (in age 28 years; BMI 21.10 kg/m²; 9.4 week of pregnancy) who gave birth to healthy children. Blood was collected from women in the first trimester of pregnancy (A). Placental tissue fragments were collected from 35 pregnant women after the birth of children (B).

The study was approved by the local Medical Ethics Committee (No. R-I-002/348/2007). All women gave informed and written consent to participate in the study.

2.2. Research methodology

TAS and GSH-Px activity was measured by using the ready-made sets of tests by Randox Laboratories-Us Ltd., USA and Spectrophotometer UV–VIS. ABTS[®] (2,2'-Azino-di-[3-ethyl-benzthiazoline sulphonate]) is incubated with a peroxidase (metmyoglobin) and H₂O₂ to produce the radical cation ABTS^{®*}. This has a relatively stable blue–green color, which is measured at 600 nm. Antioxidants in the added sample cause suppression of this color production to a degree which is proportional to their concentration.

Glutathione Peroxidase (GPX) catalyzes the oxidation of Glutathione (GSH) by Cumene Hydroperoxide. This method is based on that of Paglia and Valentine [11]. In the presence of Glutathione Reductase (GR) and NADPH the oxidized Glutathione (GSSG) is immediately converted to the reduced form with a concomitant oxidation of NADPH to NADP⁺. The decrease in absorbance at 340 nm is measured.

The content of microelements was estimated using the atomic absorption spectrometry (AAS) method, a flameless technique with electrothermal atomization in a graphite tube (Se, Cu, Mn) and flame atomization (Zn) with Zeeman's background correction on a Z-5000 spectrometer from Hitachi, Japan. Certified reference material: Seronorm Trace Elements Serum 0608414, Sero-As Norway and Community Bureau of Reference Bovine Muscle

BCR 184, Belgium as control for analysis were used to test the accuracy of the method.

2.3. Statistical analysis

The Statistica v. 9.0 software (StatSoft, Inc.) was applied for data computation to determine statistical significance which was set at p < 0.05 by using Student's *t*-test for dependent samples. Due to the small number of one of the groups it was not possible to evaluate the normality of distribution and, therefore we selected nonparametric methods (test of Mann–Whitney *U* and Spearman's rank correlation coefficient). To compare the two groups we used the test of Mann–Whitney *U*, and to determine the association between two variables Spearman's rank correlations were used.

3. Results

3.1. TAS and GSH-Px activity, Se, Zn, Cu and Mn concentrations in serum

TAS and GSH-Px activity, Se, Zn, Cu and Mn concentrations in serum of women in the first trimester of pregnancy (control group – A) and women who have had a miscarriage are presented in Table 1. TAS activity, Zn and Cu concentration in serum in women who experienced a miscarriage was lower than in women in the first trimester of pregnancy (A), but GSH-Px activity, Se and Mn level was higher in experimental group. Se and Zn concentration in both groups (experimental and control) was below the reference range [12].

3.2. The content of Se, Zn, Cu and Mn in placental tissue

The content of Se, Zn, Cu and Mn in placental tissue of women after childbirth and women who have had a miscarriage (B) are presented in Table 2. Se, Cu and Mn content in placental tissue in patients who have had a miscarriage was higher than in pregnant women at full-term delivery (B). Only Zn content in placental

Table 2
Se, Zn, Cu and Mn content in placental tissue.

No.	Trace elements	Control group (B) n=35 Me (Q ₁ –Q ₃)	Women who have had a miscarriage n=83 Me (Q ₁ –Q ₃)	p
1	Se (ng/g)	64.79 (58.76–75.68)	108.9 (91.16–139.1)	<0.00001
2	Zn (μg/g)	9.167 (8.095–10.38)	7.682 (6.366–9.277)	<0.0007
3	Cu (μg/g)	0.990 (0.860–1.098)	1.419 (1.169–1.881)	< 0.00002
4	Mn (ng/g)	89.86 (73.38–123.9)	127.1 (90.47–174.9)	<0.003

Me – median; Q₁ – lower quartile; Q₃ – upper quartile; B – control group (pregnant women after childbirth).

p < 0.05 – statistical significance.

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