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Original Research Article

Relationships between parameters of iron metabolism and serum concentrations of copper and selenium in women with normal and problem pregnancies

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Abstract

Published data show that homeostasis of iron, copper and selenium is tightly interlinked. The aim of our study was to determine the relationships between parameters of iron metabolism and serum concentrations of copper and selenium in women with normal and problem pregnancies. We conducted a prospective case control study including 14 pregnant women: 8 with normal and 6 with problem pregnancies. Haematological parameters haemoglobin, haematocrit, RBC, MCV, MCH, and MCHC; biochemical markers – serum ferritin, serum iron, total iron-binding capacity, and transferrin saturation; serum concentrations of copper and selenium were determined in all women between 16-18 weeks of gestation. The group of problem pregnancies was followed up between 30-32 weeks of gestation. Significantly higher mean transferrin saturation (38.11±3.52% vs. 25.74±2.66%; p=0.0207) and lower mean serum selenium concentration (779.4±37.53 nmol/l vs. 1019.38±29.67; p=0.0004) were found in problem pregnancies at the beginning of second trimester compared to the normal ones. Correlation analysis of laboratory parameters at the beginning of third trimester revealed that serum copper concentrations in problem pregnancies were significantly inversely related with serum iron (r=–0.9045; p=0.0349) and transferrin saturation (r=–0.9882; p=0.0015), whereas serum selenium was positively correlated with serum iron (r=0.9082; p=0.0329) and negatively with serum copper (r=–0.8590; p=0.0284). Dynamical assessment of parameters of iron metabolism and serum concentrations of copper and selenium during pregnancy would allow appropriate trace element supplementation, would optimize its period and duration for the purpose of carrying and delivering healthy children.

Key words: Copper, Dynamical assessment, Iron metabolism, Pregnancy, Selenium,

List of Abbreviations

Fe – iron; **Hb** – haemoglobin; **Cu** – copper; **Se** – selenium; **GPx** – glutathione peroxidase; **IDA** – iron deficiency anaemia; **BMI** – body mass index; **GN** – group of normal pregnancies; **GP** – group of problem pregnancies; **ART** – assisted reproductive technology; **IUI** – intrauterine insemination; **IVF** – in vitro fertilization; **Hct** – haematocrit; **RBC** – red blood cells; **SFe** – serum iron; **TIBC** – total iron-binding capacity; **SatTf** – transferrin saturation; **ECLIA** – electrochemiluminescence immunoassay; **Cybrd1** – cytochrome B reductase 1; **Steap** – six-transmembrane epithelial antigen of the prostate; **DMT1** – Divalent metal transporter1; **ATP7A** – Menkes copper ATP-ase

INTRODUCTION

Over the last years, a significant knowledge was gained in the metabolism of iron (Fe) and other essential trace elements, mechanisms of their homeostatic regulation, and interrelations between trace elements. Iron is required for a variety of physiological processes being an integral component of the oxygen-transporting proteins

haemoglobin (Hb) and myoglobin, and iron-containing enzymes. Iron homeostasis is regulated at cellular and systemic level (Kroot et al., 2011). Due to the fact that there is no physiological mechanism for regulation of iron excretion, maintenance of systemic iron homeostasis is achieved by modulation of the intestinal iron absorption (Andrews, 1999). Under physiological and pathological conditions systemic iron homeostasis is regulated in response to many diverse factors: body iron status, iron requirements for erythropoiesis, oxygen tension, and inflammation (Kroot et al., 2011). In addition, iron concentrations are significantly influenced by interactions with other trace elements, such as copper (Cu) and selenium (Se).

Erythropoiesis is a physiological process, which requires considerable amounts of Fe. Pregnancy as a physiological condition in women is associated with elevated requirements of Fe due to the growth of foetus, placenta, and expansion of the maternal erythroid mass (Koenig et al., 2014). It is estimated that the additional iron requirements throughout the pregnancy correspond to approximately 3 mg Fe/day (Scientific Committee on Food, 2006). Deficiency of Fe in pregnancy has been associated with increased maternal and infant morbidity and mortality, preterm birth, low birth weight, and long-lasting negative effects on children's cognitive development (Stoltzfus et al., 2004).

Copper serves as a cofactor in a number of enzymes involved in iron metabolism and haemoglobin synthesis (Olivares and Uauy, 1996; Collins et al., 2010; Zimmermann, 2007). Copper plays important role in systemic iron homeostasis and is considered vital to the normal haematopoiesis (World Health Organization, 1998). Iron and copper metabolism are intimately linked at the level of major aspects of iron homeostasis – intestinal absorption, mobilization from stores and utilization by the bone marrow. Relationships between iron metabolism and Cu have not been completely elucidated yet. Both high and low serum Cu levels have shown associations with iron deficiency and anaemia (Gropper et al., 2002; Turgut et al., 2007; Wajeunnesa et al., 2009; Shah et al., 2011). In pregnancy, Cu concentration in maternal serum is increased in order to meet elevated copper requirements of foetus (Alebic-Juretic and Frkovic, 2005; Alvarez et al., 2007). *In vivo* animal studies have shown that prenatal copper deficiency is associated with intrauterine growth retardation, and long-term postnatal alterations in antioxidant defense, formation of connective tissue, and production of energy (Vukelic et al., 2012). High Cu levels in maternal serum are also considered detrimental being associated with cerebral disorders in the foetus and increased risk of spontaneous abortions (Alvarez et al., 2007).

Selenium, as a cofactor of selenoproteins, acts in antioxidant defense and redox processes of the organism (Moghadaszadeh and Beggs , 2006), including erythropoiesis and erythrocytes functioning (Chow and Chen, 1980; Nagababu et al., 2003). The total number of known human selenoproteins reaches 25 and they are classified into 3 families: glutathione peroxidases (GPx), thioredoxin reductases, and iodothyronine deiodinases. Selenoproteins play important roles in thyroid hormone metabolism and muscle development and function. These proteins enhance the apoptosis of cancer cells, improve the immune response against infectious diseases, suppress prostaglandin synthesis, and are involved in the normal sperm maturation and motility (Moghadaszadeh and Beggs, 2006). The significance of Se for endocrine and reproductive functions is highlighted by the fact that under conditions of insufficient intake, Se is redistributed into the brain, endocrine and reproductive organs (Beckett and Arthur, 2005; Bermano et al., 1995). Selenium is provided mainly by food. A diet containing 0.1 µg Se/g of food is considered sufficient for normal growth and reproduction (World Health Organization, 1996). Deficiency of Se has been associated with increased risk of thyroid disease, preeclampsia, spontaneous abortions, male infertility (Rayman, 2000; Combs, 2001), and foetal congenital abnormalities (Kilinc et al., 2010). The period of pregnancy is known to be associated with a particularly high risk of Se deficiency. This is presumed to be due to high requirements of Se for the growth of foetus and increased tissue metabolism of the mother (Bivolarska, 2014) leading to decreased maternal serum Se concentrations (Gladyshev, 2006; Alvarez et al., 2007). It is known that GPx and iodothyronine deiodinases play important roles in production of thyroid hormones and exposition of tissues to their physiological effects (Moghadaszadeh and Beggs, 2006). However, according to other authors it is the physiologic haemodilution that predominantly accounts for decreased serum Se levels during pregnancy (Kilinc et al., 2010). Selenium deficiency has been found among pregnant women in Bulgaria (Bivolarska, 2014).

The relationship between Se and iron metabolism still remains unclear. Although a number of studies have shown significantly lower serum Se in subjects with iron deficiency anaemia (IDA) compared to non-anaemic subjects (Yetgin et al., 1992; Gurgoze et al., 2004; Van Nhien, et al., 2006), the results concerning subjects with non-anaemic iron depletion remain controversial (McAnulty et al., 2003). We consider that the findings in subjects with IDA may be explained by the role of selenium in antioxidant defense and normal erythropoiesis. It is of interest to note that iron supplementation has been shown to diminish serum Se levels, while selenium supplementation has not been associated with alterations in iron metabolism (Viita et al., 1989).

Adequate availability of Fe, Cu and Se and maintenance of their balance within the body is essential for the health of pregnant women, developing foetus and

newborn child. Due to the higher requirements for micronutrients during pregnancy, pregnant women are at an increased risk of trace element deficiencies (Langley-Evans, 2009; Bivolarska, 2014). Consequences can be deleterious to the health of both the mother and developing foetus.

The aim of our study was to determine the relationships between parameters of iron metabolism and serum concentrations of Cu and Se in pregnant women with normal and problem pregnancies.

MATERIALS AND METHODS

Study population

The present study was approved by the Institutional Ethical Committee of Medical University – Pleven (Bulgaria) in accordance with the ethical criteria for studies involving human beings.

We conducted a prospective case control study between October, 2015 and January, 2016. A total of 14 pregnant women with a mean age 29.95 ± 4.88 (median 30.0, range 22.0-39.0) were included in the study. They were recruited for the study at time of registration among pregnant women who had periodic check-ups of their pregnancies in two outpatient centers. All of women were enrolled in the study between 16-18 weeks of gestation (in the beginning of second trimester). The women participated voluntarily. Informed consent was obtained prior to participation. A detailed questionnaire was filled including data of course and outcome of preceding pregnancies, any history of chronic illness, reproductive failures, and administration of medications, iron, trace element, and vitamin supplementation. Anthropometric parameters body weight and height were measured by a standard methodology and used for assessment of body mass index (BMI) – BMI (kg/m^2) = body weight/$height^2$. Complete physical examination was performed with measurement of blood pressure according to the indirect method with a manual sphygmomanometer. Two groups of women were defined in the study population: the group of normal pregnancies (GN) consisting of 8 women and the group of problem pregnancies (GP) including 6 women. As normal pregnancies were defined singleton pregnancies with BMI 20-25 kg/m^2 and arterial pressure less than 140/90 mmHg at time of registration. Exclusion criteria for GN involved pre-existing cardiovascular, respiratory and renal disorders, systemic autoimmune diseases, chronic inflammatory conditions, diabetes mellitus or impaired glucose tolerance, obesity (BMI ≥30 kg/m^2 at time of registration), previous pregnancy with gestational diabetes or pre-eclampsia, preceding reproductive failures, and multiple gestation.

The group GP included pregnancies associated with a high risk. Reproductive failures (infertility, spontaneous abortions) preceded the current pregnancy in all women from group GP except for one with pre-existing valvular heart disorder. Furthermore, two of these women had pre-existing autoimmune thyroid disease (Hashimoto thyroiditis), and one – chronic hepatitis. In the women with preceding reproductive failures, current pregnancy was achieved by assisted reproductive technology (ART) procedures: intrauterine insemination (IUI) and conventional in vitro fertilization (IVF). One woman had multiple pregnancy and the others had singleton pregnancies.

Measurements of parameters of iron metabolism, serum concentrations of Cu and Se

Iron metabolism was characterized by haematological parameters and biochemical markers which assess various aspects of iron metabolism (Centers for Disease Control and Prevention, 1998). Haematological parameters: Hb, haematocrit (Hct), red blood cells (RBC), erythrocyte indices – MCV, MCH, and MCHC were determined. Biochemical markers serum ferritin, serum iron (SFe), total iron-binding capacity (TIBC), and transferrin saturation (SatTf) were used to characterize iron metabolism. Percent SatTf was calculated as a ratio of SFe and TIBC – SatTf(%)=SFe/TIBC×100. Trace elements Cu and Se were determined quantitatively in serum.

Taking into account the diurnal variations in the concentrations of trace elements and biochemical markers of iron metabolism we collected venous blood samples in the morning at fasting state. Haematological indicators were measured from whole-blood samples collected in vacutainer tubes with EDTA as an anticoagulant. Commercially available vacutainers with clot activator to accelerate the blood clotting and polymer gel separating serum from the clot were used for biochemical tests and trace element determinations. Blood samples were immediately transported to our laboratory, where were allowed to clot for two hours at room temperature. After centrifugation at 3000 rpm for 10 minutes, serum was separated from the clot. Serum samples were put in cryogenic storage vials and stored at –70°C until analysis.

Haematological parameters were determined on haematological analyzer MICROS. Serum ferritin was measured by electrochemiluminescence immunoassay (ECLIA) on Cobas e 411 immunoassay analyzer. Serum Fe and TIBC were determined by FerroZine method. Serum concentrations of trace elements Cu and Se were measured by electrothermal atomic absorption spectrophotometry on atomic absorption spectrophotometer Perkin-Elmer Zeeman 5000.

Table 1. Results of investigated laboratory parameters in pregnant women of GP and GN as means ± standard error (SE) at time of registration (between 16-18 weeks of pregnancy)

Parameter	GP n=6	GN n=8	p-value
Hb (g/L)	121.2±3.01	119.0±2.38	0.5774
Hct (L/L)	0.34±0.01	0.35±0.01	0.6352
RBC ($\times 10^{12}$/L)	3.81±0.13	3.89±0.11	0.6744
MCV (fl)	88.75±1.52	89.75±2.16	0.7241
MCH (pg)	31.15±1.21	30.9±1.71	0.9105
MCHC (g/L)	341.0±4.12	344.5±5.04	0.6281
Ferritin (µg/L)	57.56±14.09	41.18±10.65	0.3783
SFe (µmol/L)	21.37±2.45	17.66±1.85	0.2568
TIBC (µmol/L)	56.31±3.38	68.3±2.55	**0.0196**
SatTf (%)	38.11±3.52	25.74±2.66	**0.0207**
Serum Cu (µmol/L)	34.48±2.59	36.33±2.05	0.5872
Serum Se (nmol/L)	779.4±37.53	1019.38±29.67	**0.0004**

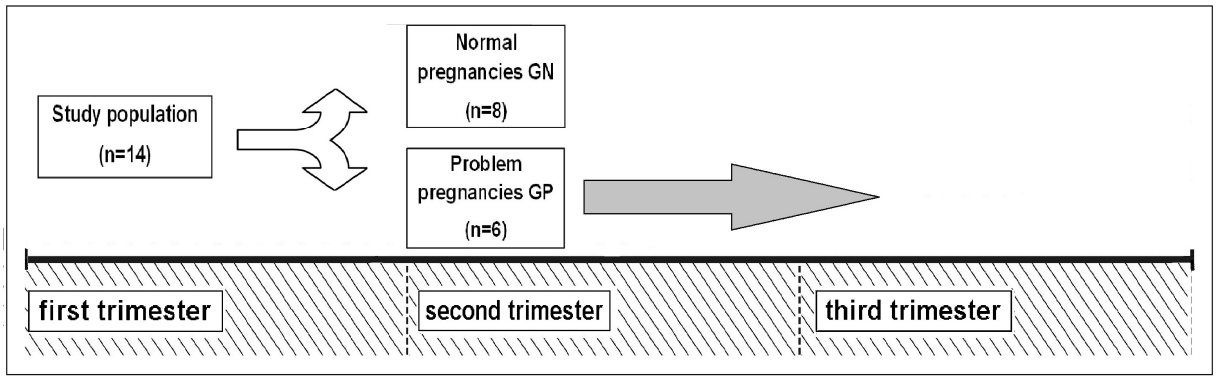


Figure 1. Study profile

Statistical analysis

Data were processed by statistical package STATGRAPHICS Centurion XVI. The mean values of anthropometric, haematological and biochemical parameters were compared between the groups of normal and problem pregnancies using analysis of variance (One-Way ANOVA). Student's t-test was used to test whether there were any significant differences between the mean age characteristics of studied groups. Relationships between numerical variables were assessed using a correlation analysis with calculation of the correlation coefficients by Pearson's test. The potential effects of outliers were assessed in all analyses. In all the analyses, as a level of statistical significance, below which the null hypothesis was rejected, a value of 0.05 ($p<0.05$) was used.

RESULTS

The group of normal pregnancies consisted of 8 women with a mean age 31.09 years ±4.85 (median 31.0, range 22.0-39.0). The group of problem pregnancies included 6 women with a mean age 28.38 years ±4.78 (median 27.5, range 23.0-36.0). We found no significant difference between the mean ages of women from two groups ($p=0.2418$).

All the women participating in this study were examined with respect to investigated characteristics, anthropometric and laboratory parameters at the beginning of second trimester – between 16-18 weeks of pregnancy. The mean values of anthropometric and haematological parameters, biochemical markers of iron metabolism, and serum trace element concentrations were compared between the study groups (Table 1).

In accordance with the findings, group of problem pregnancies was followed up in the beginning of third trimester – between 30-32 weeks of gestation, as shown on Figure 1.

There were no significant differences in body weight ($p=0.2006$) and BMI (0.6292) between women with normal and problem pregnancies at time of registration. Mean body weight of women with normal pregnancies was 70.13±3.79 kg with BMI 23.45±0.82 kg/m^2 and the respective values were 61.8±4.8 kg for body weight and

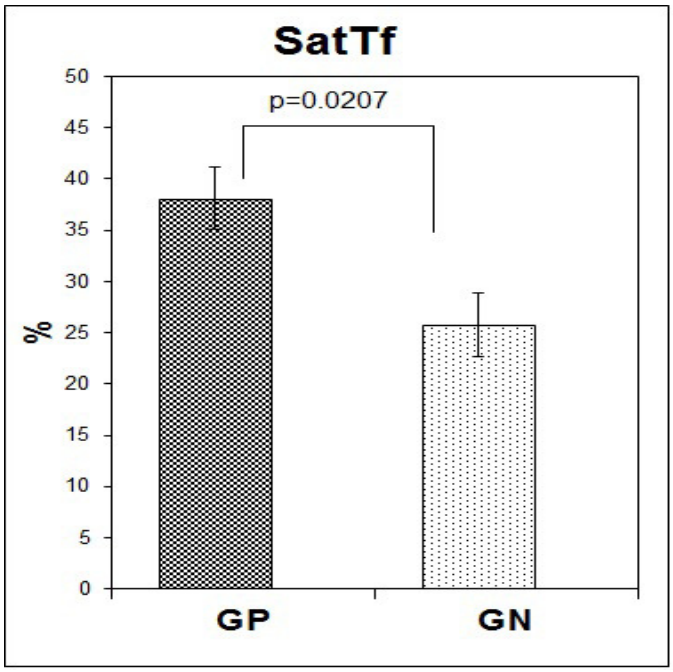


Figure 2. Percent SatTf of pregnant women with problem and normal pregnancies at the beginning of second trimester as means ± SE

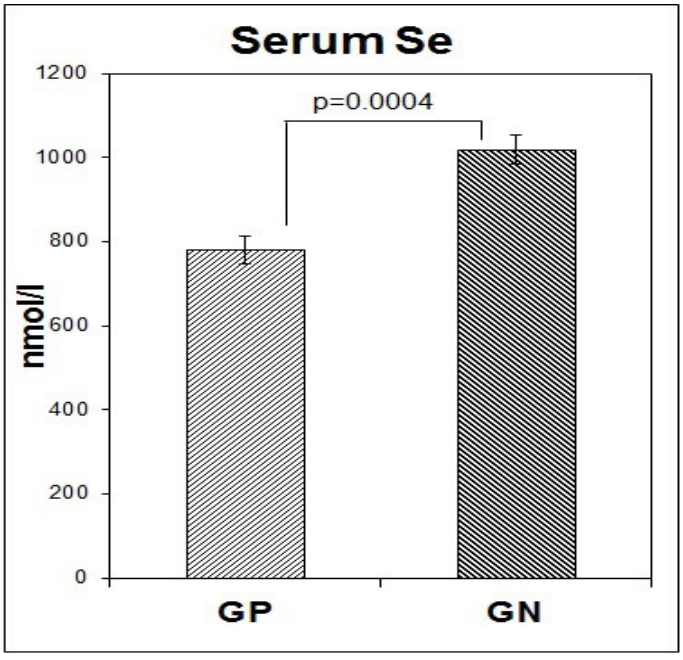
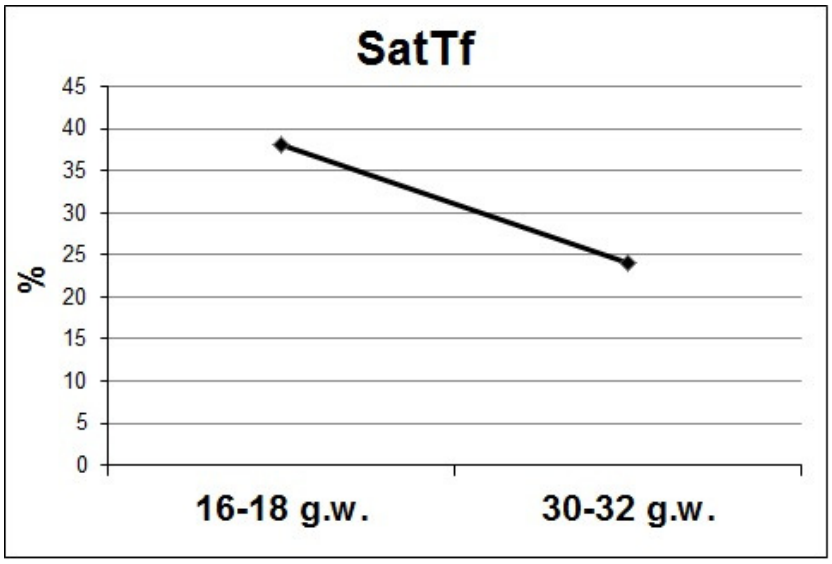


Figure 3. Serum Se concentrations of pregnant women with problem and normal pregnancies at the beginning of second trimester as means ± SE in nmol/l

22.82±0.97 kg/m^2 for BMI for group of problem pregnancies.

Comparison of laboratory parameters between groups at the beginning of second trimester revealed significantly higher mean SatTf in problem pregnancies than the normal ones (Figure 2), while TIBC was significantly lower in problem pregnancies. Serum concentrations of Se were found to be significantly lower in group GP versus group GN (Figure 3). No significant differences were observed for the mean values of haematological



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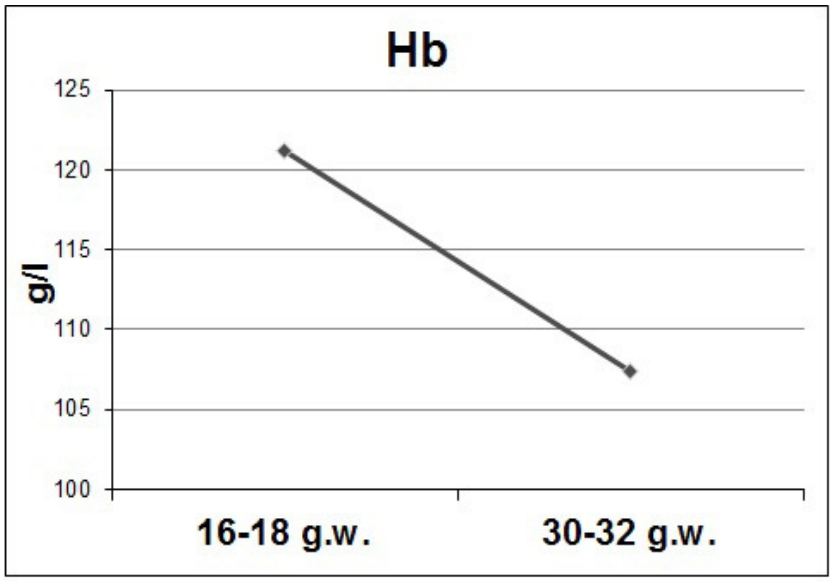

B

*Abbreviation: g. w. – gestational weeks

Figure 4. Dynamical changes of SatTf (A) and Hb concentration (B) throughout the gestation in women with problem pregnancies

parameters, serum ferritin and SFe ($p \geq 0.05$). Serum Cu concentrations also showed no significant differences between women with normal and problem pregnancies.

We followed up the dynamics of investigated haematological and biochemical parameters in women of group GP in the beginning of third trimester and results were compared to the values obtained previously. Dynamical assessment (Figure 4) revealed a significant decline in SatTf from a value of 38.11±4.22% at the beginning of second trimester to 24.14±3.77% at the beginning of third trimester ($p=0.0428$). This was accompanied by a decrease in the Hb concentration from 121.2±2.97 g/l to 107.33±2.71 g/l ($p=0.0073$). Other haematological parameters, serum ferritin, SFe, TIBC, and serum concentrations of Cu and Se did not show any significant changes with progression of the pregnancy ($p \geq 0.05$).

Correlation analysis of laboratory parameters in group GP at the beginning of third trimester revealed that serum Cu concentrations were significantly inversely related with SFe ($r=-0.9045$; $p=0.0349$) and SatTf ($r=-0.9882$; $p=0.0015$), whereas serum Se was positively correlated with SFe ($r=0.9082$; $p=0.0329$). A strong negative relationship was obtained between serum levels of Cu and Se ($r=-0.8590$; $p=0.0284$).

DISCUSSION

The current literature demonstrates that high-risk pregnancies, and especially those associated with inflammatory processes, exhibit significant alterations in iron homeostasis (Gangopadhyay et al., 2011; Koenig et al., 2014). Inflammation has been shown to induce synthesis of hepcidin, a newly discovered peptide hormone recognized as a key regulator of systemic iron homeostasis. Increased hepcidin suppresses iron absorption from the gastrointestinal tract and iron mobilization from the reticuloendothelial system leading to diminished iron availability for the erythropoiesis (Kroot et al., 2011).

Contrary to chronic inflammatory characteristic of the pathological conditions observed in the group of problem pregnancies, we found significantly higher SatTf in problem pregnancies compared to normal pregnancies at the beginning of second trimester. These findings might be explained by the fact that iron and multiple micronutrient supplements were more commonly administered in group GP than group GN. Our observations showed that in the beginning of second trimester 40% of women in GP received iron supplements and all women – multiple micronutrients, while for the same period 12.5% of women in group GN received iron supplements and none – multiple micronutrients. Results in a study of Rettmer et al. (1999) indicate that SatTf increases dramatically in setting of iron supplementation.

Serum concentrations of Cu are one of the most widely used parameters for evaluation of copper status (Olivares and Uauy, 1996). In our study, comparison of serum Cu concentrations between problem and normal pregnancies at the beginning of second trimester did not show any statistically significant differences. Other authors have reported similar results concerning different pathological conditions in comparison to normal pregnancies during the second trimester (Alebic-Juretic and Frkovic, 2005), while others have found significantly lower serum Cu concentrations in pathological pregnancies compared to the normal course of pregnancy (Vukelic et al., 2012).

Serum Se concentration is the most widely used indicator of selenium status (Vandecasteele and Block, 1994). Published data show that Se is distributed in the serum as follows: 53±6% associated with selenoprotein P; 39±6% – as GPx, and 9±4% associated with albumin (Harrison et al., 1996). In 1997 Rayman suggested as a criterion for adequate Se intake a serum concentration of around 100 nmol Se/ml (Combs, 2001). The question of Se deficiency resulting from low dietary intake is of particular interest since the Balkan region is known to have a low soil Se concentration which determines a lower selenium intake by the food and drinking water (Bivolarska, 2014). The alarming fact is that the mean serum level of Se determined for Bulgarian population (45±6 µg/l) is lower compared to the Western European populations (70-120 µg/l) (Combs, 2001). We report on significantly lower serum Se concentration in the group of high-risk as compared to normal pregnancies. This result is consistent with the findings in studies of women suffering first-trimester or recurrent spontaneous abortions. Lower serum Se concentrations in these groups than in women with normal pregnancies have been probably linked to reduced antioxidant defense by the low levels of Se-dependent GPx 1 (Rayman, 2000). As we already stated, most of the pregnancies in group GP were preceded by reproductive failures including spontaneous abortions.

As expected, we observed a significant decline in the Hb concentration and SatTf with the progress of pregnancy. Similar to our findings, dynamical assessment of indicators of iron metabolism in other studies have revealed progressive reduction in the levels of Hb, serum ferritin, and SatTf during the course of pregnancy (World Health Organization, Centers for Disease Control and Prevention, 2007; Bivolarska, 2014). These changes have been explained by the increased requirements of Fe due to the foetal growth and by the physiologic hemodilution as a characteristic of advanced stages of pregnancy. Lack of changes in serum ferritin levels in our study was probably due to confounding effects of inflammatory processes, since we followed up only the group of problem pregnancies. Although serum ferritin is considered the best indicator of iron stores, as an acute-phase reactant, its concentrations may be elevated in the presence of inflammation, hyperthyroidism, or liver disease (Zimmermann, 2008).

In contrast to studies of normal pregnancies, we did not find statistically significant changes of serum Cu concentrations during the problem pregnancies. In other studies investigating changes of serum Cu during the normal pregnancy, a significant rise has been found reaching its maximum level in the third trimester of pregnancy (Alebic-Juretic and Frkovic, 2005; Alvarez et al., 2007; Vukelic et al., 2012). This has been attributed to the increase of ceruloplasmin resulting from elevated oestrogen levels during pregnancy (Baig et al., 2003). These findings are in contrast with our results showing lack of dynamical changes of serum Cu over the gestation which is probably due to the chronic inflammatory processes frequently observed in the group GP. It is well-known the role of serum Cu as an acute-phase reactant showing increased levels in inflammatory conditions (Olivares and Uauy, 1996).

Similar to the serum Cu concentrations, we did not find any significant changes of serum Se with advance of pregnancy. Progressive decline in serum Se levels has been reported during the normal course of pregnancy (Kantola et al., 2004; Alvarez et al., 2007). A significant peak of the mean serum Se values in the second trimester has been observed by other authors, as well as lack of significant changes of serum Se over the pregnancy in different groups of pregnant women

(Bivolarska, 2014). We consider that lack of changes in serum Se with the progress of pregnancy in group GP may result from the relatively low levels of serum Se in this group at the beginning of observation period.

We speculate that the negative correlation between biochemical markers of iron metabolism and serum Cu concentration in our study may be explained by metabolic interactions between Fe and Cu. Inverse relationships between iron and copper homeostasis have been demonstrated in a number of studies. The molecular base of these interactions is possibly at the level of intestinal absorption of Fe and Cu. Both metals are absorbed in the upper small intestine where they are interconnected by intestinal transport mechanisms. The metalloreductase enzymes on the apical membrane of enterocytes: cytochrome B reductase 1 (Cybrd1), cytochrome b (558) reductase, and Steap (six-transmembrane epithelial antigen of the prostate) proteins all function as cupric reductases in addition to their ferric reductase activity (Collins et al., 2010). Divalent metal transporter 1 (DMT1), which is required for iron transfer across the apical membrane (Andrews, 1999; Nadadur et al., 2008), appears to be involved in the transport of Cu (Arredondo et al., 2003) although this issue remains disputable. It has been found that Fe and Cu exhibit competitive inhibitions for their cellular uptake possibly through a DMT1-dependent mechanism (Arredondo, 2004; Arredondo, 2006). Competitive interactions between Fe and Cu for their intestinal absorption might be associated with reciprocal alterations in their bioavailability (Arredondo, 2006). *In vivo* animal studies have found evidence of enhanced copper absorption during iron deficiency, as suggested by the increased intestinal expression of cupric reductase Cybrd1, DMT1, copper exporter Menkes copper ATP-ase (ATP7A), and metallothioneins, and copper accumulation in the gut wall and liver (Collins et al. 2010).

The mechanism explaining positive correlation between SFe and serum Se found in our study is not known. A significant proportion of total Se in serum is present in the form of GPx (Harrison et al., 1996). It is considered that Fe is involved in pretranslational regulation of this enzyme (Gropper et al., 2009). In contrast to our result, a study on non-pregnant women has not found any relationship between normal levels of serum Se and biochemical markers of iron metabolism (Viita et al., 1989), neither has a relation been found in subjects with IDA (Yetgin et al., 1992; Gurgoze et al., 2004). We suggest that the relationship between Se and iron metabolism is complicated and needs further detailed research.

We found a strong inverse relationship between serum Cu and Se concentrations at the beginning of third trimester. Similarly, other authors have also reported a strong inverse correlation between serum Se and Cu in non-smoking pregnant women at term, although a positive correlation has been found in smokers (Kantola et al., 2004). The underlying mechanism in the relations between Cu and Se needs further investigation.

CONCLUSIONS

Investigation of the relationships between parameters of iron metabolism and serum concentrations of Cu and Se in this study reveals that the homeostasis of Fe, Cu and Se is tightly interlinked during pregnancy. The molecular basis of their interrelations is still to be elucidated. It is well-known that requirements for macro- and micronutrients are elevated during pregnancy, but the possible interactions between trace elements should be kept in mind when prescribing trace element supplementation. Serum Cu and Se levels may be useful in anticipating effects of iron supplementation in pregnant women. Given the fact that significant part of iron deficiency states during pregnancy is not responsive to iron supplementation, it is necessary to evaluate serum Cu and Se levels prior to prescribing iron supplements. Dynamical assessment of parameters of iron metabolism and serum concentrations of Cu and Se during pregnancy would allow appropriate trace element supplementation, would optimize its period and duration for the purpose of carrying and delivering healthy children. Special attention should be given to pregnant women, who live in regions with poor selenium soil content, susceptible to reduced antioxidant defense and complications over the course of pregnancy.

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