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Obesity is associated with increased level of kisspeptin in mothers' blood and umbilical cord blood – a pilot study

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Abstract. – OBJECTIVE: Kisspeptin (KP) is a major regulator of reproductive functions. It has also been shown to be involved in the metabolic changes associated with obesity. According to the well-established concept of prenatal programming, environmental factors can influence physiological and behavioral systems at the early stages of development. Thus, we hypothesized that in pregnant women, obesity can be associated with alterations in the levels of KP. We also assumed that the observed changes in obese mothers' blood (MB) would be reflected in the umbilical cord blood (CB).

MATERIALS AND METHODS: We collected MB and CB from obese and nonobese women and analyzed the differences in metabolic and hormonal profiles, including KP concentration, using commercially available assays.

RESULTS: We found that the level of KP was increased in the MB and CB of obese patients compared to nonobese subjects ($p<0.05$). A strong correlation was observed between the concentration of KP in MB and CB (r=0.8343; $p<0.01$). Moreover, we detected that the differences in the adipokine profile observed in the MB were not reflected in CB.

CONCLUSIONS: Our results indicate that blood KP concentration can serve as a valuable marker in pregnant women. However, further studies are needed to understand the alterations of this peptide in obese pregnant woman and their potential effects on offspring.

Key Words:
Kisspeptin, Obesity, Mothers' blood, Umbilical cord blood.

Introduction

According to the well-established concept of prenatal programming, environmental factors can influence physiological and behavioral systems during the early stages of development[1-3]. Studies performed by Phillips[4,5] on children showed a strong correlation between low birth weight, high cortisol levels, and later development of metabolic disorders such as obesity and type 2 diabetes (DM2).

A wide range of data indicates that the dietary environment of the mother plays an important role in critical stages in the prenatal development of the fetus. Poor maternal diet may also increase the risk of metabolic diseases in later life. The offspring of mothers who suffered from *diabetes mellitus* during pregnancy are at a higher risk of developing obesity and experiencing abnormal glucose metabolism in childhood, adolescence, or adulthood[6]. It was shown that high fetal mass is associated with increased glucose flow through the placenta, which contributes to raised insulin secretion by the fetal beta pancreas cells and consequently fetal macrosomy (abnormal increase in the body mass of a newborn)[6]. Moreover, it has been proved that poor nutrition during pregnancy can have a detrimental effect on the newborn's physiological systems (e.g., reproductive), besides metabolism[7,8].

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Reproduction is governed by the hypothalamic-pituitary-gonadal (HPG) axis. The first component of HPG is the hypothalamus, which secretes gonadotropin-releasing hormone (GnRH), stimulating the pituitary gland to release gonadotropins (luteinizing hormone and follicle-stimulating hormone). These gonadotropins act on the gonads—ovaries and testes—to promote their maturation, and subsequently gametogenesis and steroid hormone production[9]. The HPG axis is regulated by various factors, including neuropeptide kisspeptin (KP), a very potent stimulator, which acts via the G-protein-coupled receptor, GPR54 (also known as Kiss1r)[10].

Data on the direct impact of maternal obesity on the reproductive functions of children are limited. A study performed[11] on a Danish pregnancy cohort showed that a high maternal body mass index (BMI) diminishes semen quality, sperm concentration, and Sertoli cell function in the male offspring. These reproductive abnormalities were related to the increased conversion of androgens to estrogens[11]. It was also found that women born to mothers with a BMI of over 25 had menarche at an earlier age[12].

Obesity is currently a global health problem and becoming more common in pregnant women. It alters the quality of labor and leads to preterm labor, prolonged labor, or higher oxytocin requirements. Maternal obesity is also associated with a number of factors that control the metabolism of lipids and carbohydrates, and thus can affect the course of pregnancy[13-15]. In general, obesity is characterized by the production and release of adipokines and the development of chronic inflammation[16,17]. Research implicates that the dysregulation of leptin, adiponectin, and KP during pregnancy contributes to gestational diabetes mellitus and pre-eclampsia[18]. It is also well documented that obesity caused by an unhealthy diet could lead to the development of DM2, which accounts for about 90% of all diabetic cases. Moreover, according to the fetal programming concept, changes in mothers' metabolism during pregnancy could have a long-term effect on the neuroendocrine system of the offspring. This could result in both metabolic (e.g., obesity and DM2) and reproductive diseases in later life. Thus, it is important to study the role of adipokines in obese pregnant women and their possible impact on the metabolic and reproductive systems of children. Recent data indicate that KP is involved in the integration of metabolic and reproductive functions[19].

Therefore, we hypothesized that in pregnant women obesity is associated with changes in metabolic profiles (glucose, insulin, cholesterol, triglycerides [TG], nonesterified fatty acids [NEFA]) as well as with the blood levels of adipokines (leptin and adiponectin) and KP. In addition, we assumed that the observed changes in obese mothers' blood (MB) would be reflected in the umbilical cord blood (CB).

Materials and Methods

Ethics

The study was conducted according to the principles of the Declaration of Helsinki. All participants were informed about the study objectives and the methodology, and each of them provided a written consent to participate in the study. The study protocol was approved by the Clinical Research Ethics Committee of the Poznan University of Medical Sciences (Approval No. 997/18).

Material

The research material was maternal peripheral blood obtained on the day of childbirth (MB) and CB. Serum samples were collected from mothers using BD Vacutainer® SST™ II Advance tubes (BD Diagnostics, Franklin Lakes, NJ, USA). Then, the samples were allowed to clot at room temperature for 15 min. Next, the clotted samples were centrifuged at 3500 rpm for 15 min at 4°C. CB samples were collected immediately after birth using the abovementioned tubes, and a similar procedure was performed as with the MB samples. After processing, both MB and CB serum samples were transferred to new Eppendorf (EP) tubes and stored at −80 °C until analysis.

Anthropometric Data

Anthropometric measurements, such as body weight as well as head, abdominal, chest, thigh, and arm circumference, were taken in newborns immediately after birth by trained medical staff. Obesity was defined by a BMI of over 25 according to the World Health Organization (WHO) standards. The data on age, body weight, and BMI of study participants before pregnancy are shown in Table I. According to the criteria of the American Diabetes Association (2016), diabetic individuals were excluded from the study.

Table I. Characteristics of mothers.

Parameter	Non-obese (n = 16)	Obese (n = 16)
Before pregnancy		
Women's age (years)	30.44 ± 1.232	32.5 ± 1.155
Height (cm)	167.9 ± 1.326	168.3 ± 1.164
Body weight (kg)	63.53 ± 1.006	91.44 ± 3.045**
BMI (kg/m^2)	22.53 ± 0.162	32.25 ± 0.909**
On the day of birth		
Body weight (kg)	79.33 ± 1.565	99.91 ± 2.819**

Statistically significant differences between means for nonobese and obese subjects are marked where **$p < 0.01$. BMI—body mass index.

Metabolic Profile

Metabolic profile was determined using commercially available colorimetric tests. The levels of glucose, cholesterol (total—test range: 0–750 mg/dL, cat. no.: C7510, CV: 1.3%, wavelength: 520 nm; high-density lipoprotein [HDL]—test range: 2–200 mg/dL, cat. no.: H7545, CV: 1.18%, wavelength: 546/660 nm; low-density lipoprotein [LDL]—test range: 0–250 mg/dL, cat. no.: G7574, CV: 1.6%, wavelength: 600 nm), and TG (test range: 0–1000 mg/dL, cat. no.: G7521, CV: 1%, wavelength: 500 nm) were measured using kits according to the manufacturer's instructions (Pointe Scientific, Canton, MI, USA). NEFA levels were also measured in samples using a commercial kit (test range: 0.01–4.00 mmol/L, cat. no.: 434-91795 and 434-91995, CV: 1.18%, wavelength: 546/660 nm; Wako Chemicals, Richmond, VA, USA). However, the volume of the sample and reagent was adjusted to the micromethod (determinations were performed on 96-well microplates). Absorbances were read on a Synergy 2 apparatus (Biotek, Winooski, VT, USA).

Hormonal Profile

Hormonal profile was determined using enzyme-linked immunosorbent assay (ELISA) or radioimmunoassay (RIA) kits. Serum KP levels were measured using Human Kisspeptin 1 (KISS1) ELISA Kit (test range: 50–800 pg/mL, intra-assay: CV<10%, interassay: CV<12%, cat. no.: 201-12-4106; Sunred, Shanghai, China). Other hormones were measured using the following tests: insulin—Human Insulin-Specific RIA (test range: 2–200 µU/mL, intra-assay: CV<4.4%, interassay: CV<6%, cat. no.: HI-14K; Merck Millipore, Burlington, MA, USA), adiponectin—Adiponectin ELISA (test range: 0.6–31000 µg/L, CV<9%, cat. no.: E09; Mediagnost, Reutlingen, Baden-Württemberg, Germany), and leptin—Multi-Species Leptin RIA (test range: 1–50 ng/mL, intra-assay: CV<5%, interassay: CV<9%, cat. no.: XL-85K; Merck Millipore, Burlington, MA, USA). For the RIA tests, gamma radiation was quantified by Wallac Wizard 1470 Gamma Counter (Perkin Elmer, Waltham, MA, USA). ELISA measurements were performed on a Synergy 2 microplate reader (Biotek, Winooski, VT, USA).

Statistical Analysis

Statistical analysis was performed using unpaired Student's *t*-test (two-tailed distribution). If the data did not meet the assumptions of *t*-test (Gaussian/normal distribution), Mann-Whitney *U* test was used. Statistical significance was accepted at $p<0.05$ (*), $p<0.01$ (**), and $p<0.001$ (***). All analyses were carried out in GraphPad Prism 6.0 software (GraphPad Software, San Diego, CA, USA). Correlations between the concentrations of KP in MB and CB were analyzed by Pearson's correlation model and linear regression.

Results

This study is the first to analyze the changes in KP concentration in MB and CB. However, we excluded as many variables as possible in the study groups.

Characteristics of the Participants

We selected a representative group of both metabolic conditions (obese and nonobese) and excluded other factors that may affect the study. Therefore, the only anthropometric indexes that differed in the two groups before pregnancy were body weight and thus BMI (Table I). We found

Table II. Anthropometric parameters of newborns.

Parameter	Non-obese	Obese
Child gender (F/M)	8/8	8/8
Body weight (kg)	3.60 ± 0.17	3.41 ± 0.23
F/M weight (kg)	3.61 ± 0.31, 3.60±0.11	3.36 ± 0.23, 3.47 ± 0.33
Head circumference (cm)	36.25 ± 0.62	35.88 ± 0.60
Chest circumference (cm)	34.25 ± 0.64	33.75 ± 0.61
Abdominal circumference (cm)	34.38 ± 0.93	34.81 ± 0.61
Thigh circumference (cm)	12.44 ± 0.29	12.25 ± 0.28
Arm circumference (cm)	10.19 ± 0.24	10.06 ± 0.23

Statistically significant differences between means for children from nonobese and obese mothers. F—female; M—male.

that body weight was higher in obese women compared to nonobese women (91.44±3.045 *vs.* 63.53±1.006; $p<0.01$). Similarly, BMI was higher in obese women compared to nonobese women (32.25±0.909 *vs.* 22.53±0.162; $p<0.01$). The body weight on the day of birth was also higher in obese women in comparison to nonobese women (99.91±2.819 *vs.* 79.33±1.565; $p<0.01$).

Anthropometric Parameters of Newborns

For both metabolic conditions investigated, the gender distribution was equal in newborns (8 girls and 8 boys), and so, the effect of the gender of newborns on the studied parameters was excluded. Additionally, no statistically significant differences in body weight were found between newborn girls and boys ($p>0.79$ in the obese group and $p>0.98$ in the nonobese group). Furthermore, a comparison of the circumference measurements showed no statistically significant differences in those parameters between the groups (Table II).

Metabolic and Hormonal Profiles in MB and CB

We found a statistically significant increase in the levels of NEFA (obese *vs.* nonobese: 0.895±0.09 *vs.* 0.670±0.054; $p<0.05$), total cholesterol (obese *vs.* nonobese: 219.1±9.42 *vs.* 187.0±7.67; $p<0.05$), LDL cholesterol (obese *vs.* nonobese: 154.3±9.16 *vs.* 119.6±9.658; $p<0.05$), TG (obese *vs.* nonobese: 327.4±21.76 *vs.* 226.7±21.07; $p<0.01$), and leptin (obese *vs.* nonobese: 9.777±0.261 *vs.* 8.447±0.406; $p<0.05$) in MB. On the other hand, we observed a decrease in the concentrations of HDL cholesterol (obese *vs.* nonobese: 44.91±2.34 *vs.* 54.84±2.89; $p<0.05$) and adiponectin (obese *vs.* nonobese: 7.332±0.59 *vs.* 9.587±0.50; $p<0.01$) in MB. We also noted a small increase in insulin and glucose levels in MB in obese women; however, the changes were not statistically significant ($p>0.33$ and $p>0.15$, respectively). There were also no statistically significant changes in these parameters in CB (Table III).

Table III. Metabolic and hormonal profiles of mother's blood and cord blood in nonobese and obese subjects.

Parameter	Non-obese		Obese	
	MB	CB	MB	CB
Glucose (mg/dL)	95.54 ± 5.96	80.21 ± 5.03	107.7 ± 5.87	88.38 ± 6.57
NEFA (mmol/L)	0.670 ± 0.054	0.409 ± 0.045	0.895 ± 0.09*	0.425 ± 0.051
Cholesterol (mg/dL)	187.0 ± 7.67	71.35 ± 5.81	219.1 ± 9.42*	74.23 ± 6.07
HDL cholesterol (mg/dL)	54.84 ± 2.89	18.13 ± 0.90	44.91 ± 2.34*	18.44 ± 0.66
LDL cholesterol (mg/dL)	119.6 ± 9.658	43.27 ± 5.33	154.3 ± 9.16**	45.69 ± 5.36
TG (mg/dL)	226.7 ± 21.07	146.9 ± 19.46	327.4 ± 21.76**	130.4 ± 24.25
Adiponectin (μg/mL)	9.587 ± 0.50	37.74 ± 2.546	7.332 ± 0.59**	36.92 ± 2.617
Leptin (ng/mL)	8.447 ± 0.406	7.553 ± 0.584	9.777 ± 0.261*	6.255 ± 0.783
Insulin (ng/mL)	11.69 ± 1.095	3.710 ± 0.617	13.77 ± 1.822	3.875 ± 0.397

Statistically significant differences between means for maternal blood from nonobese and obese subjects are marked where *$p<0.05$ and **$p<0.01$. MB—mother's blood; CB—cord blood; NEFA—nonesterified fatty acids; HDL—high-density lipoprotein; LDL—low-density lipoprotein; TG—triglycerides.

Changes in Kisspeptin (KP) Level

An analysis of changes in KP level in MB from obese and nonobese volunteers revealed an increase in this peptide in the obese group (Figure 1; $p<0.05$). Similarly, a statistically significantly higher concentration of KP was found in the CB of obese mothers compared to nonobese mothers (Figure 1; $p<0.05$). We also found a strong positive correlation between the concentrations of KP in MB and CB (Figure 2; $r=0.8347$; $p<0.01$).

Discussion

In this study, we have demonstrated for the first time that KP levels were higher in both MB and CB in obese women in comparison to nonobese women. Our results confirm the previous findings that obese pregnant women have increased blood levels of cholesterol, TG, and leptin as well as decreased adiponectin. However, similar changes in metabolic parameters were not observed in the CB of our studied obese group.

Although we excluded as many variables as possible in the study groups, the sample size was relatively small. Nevertheless, the data set was sufficient to perform statistical analyses. Therefore, we have planned to expand sampling as well as include additional markers of the reproductive system (e.g., sex steroids such as estrogen [E_2] and progesterone [P_4]). Moreover, our further studies will focus on the correlations between the prepregnancy BMI and metabolic and hormonal profiles at delivery.

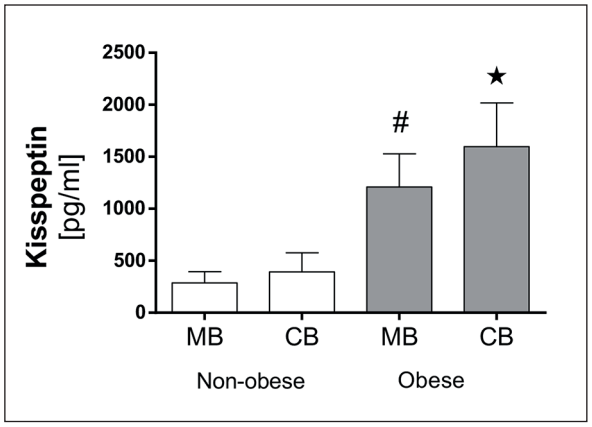

Figure 1. Changes in kisspeptin concentration in maternal and cord blood in nonobese and obese mothers. Values are presented as mean±standard error of the mean (n=32). Statistically significant differences between the means for maternal blood from nonobese and obese subjects are marked where $p<0.05$ (*), and for umbilical cord blood from nonobese and obese subjects are marked where $p<0.05$ (#). MB—mother's blood; CB—cord blood.

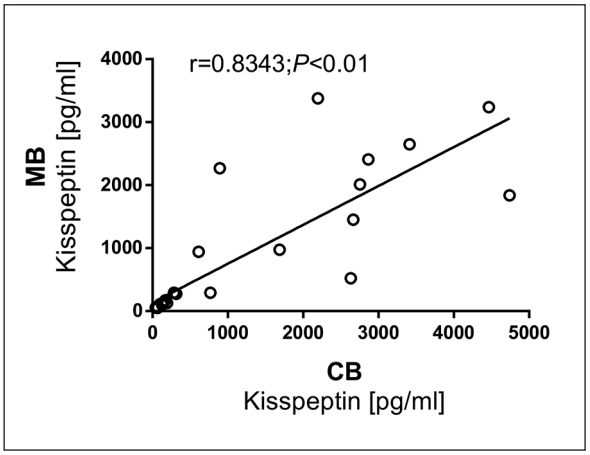

Figure 2. Correlations between kisspeptin level in mother's blood and umbilical cord blood. Values for r and p are indicated in the graph. Solid and dashed lines show the means and 95% confidence intervals, respectively, following linear regression analysis. The r-value indicates correlation, and the p-value indicates the significance of the correlation. MB—mother's blood; CB—cord blood.

Obesity has a significant impact on the regulation and functioning of not only the metabolic but also the reproductive system[20,21]. Impaired metabolic status observed in obesity may cause a decrease in the secretion of GnRH from the hypothalamus, contributing to lower plasma levels of sex steroid hormones—estrogens, progesterone, and testosterone. Moreover, maternal obesity may lead to metabolic abnormalities and, eventually, reproductive dysfunctions in the progeny. The offspring of obese mothers have a higher birth weight and tend to have a higher BMI in adulthood[22]. KP not only plays a crucial role in the regulation of the HPG axis, but also links the metabolic and reproductive functions[19,23]. Reproduction is a complex process, and its success is determined by energy and nutrient reserves. Thus, a strong relationship exists between metabolism and reproduction, and altered energy balance leads to the suppression of reproductive function[24,25]. A study on female mice[26] with diet-induced obesity showed that impaired fertility was associated not only with systemic inflammation[26] but also with the reduced hypothalamic expression of GnRH gene[27]. In addition, alterations in the negative feedback control of gonadotrophin secretion, increased

risk of infertility and miscarriage, and lower *in vitro* fertilization success rates were found in obese women[28].

It is evident that changes in KP levels could affect both reproduction and metabolism. In genetic and diet-induced animal models of obesity, KP gene expression has been observed to be altered[29,30]. A direct role for KP in the regulation of energy balance and metabolism was reported based on Kiss1r-knockout (Kiss1rKO) studies. It was found that female Kiss1rKO mice had increased body weights and adiposity and exhibited impaired glucose regulation as well as reduced energy expenditure[31]. A study by De Bond[32] confirmed the above finding and showed that female Kiss1rKO mice at maturity had abnormal body weights and showed increased white adipose tissue mass. Together, these suggest that Kiss1rKO mice with altered metabolism are sexually dimorphic, with females showing higher metabolic alterations than males[32].

However, clinical data on KP in obese patients are limited. A study reported that obese postmenopausal women of Asian origin had higher serum KP levels. Moreover, these women (n=84) were characterized by a higher serum leptin concentration and free leptin index (FLI), as well as lower serum soluble leptin receptor (sOBR) concentration, compared to nonobese controls (n=87)[33]. The study also showed that leptin and FLI were positively correlated with BMI, while sOBR and KP were negatively correlated[33]. However, it should be mentioned that the patients in the cohort had a wide age range (40–75 years). The mean age of subjects in the obese group was 58.79±6.99 years and in the nonobese group was 59.22±6.18 years. In another research performed on obese (n=15, BMI 40.23±1.31) and healthy (n=15, BMI 22.50±0.58) younger (mean age 42 years) Polish females, the blood levels of KP were lower in obese subjects. Additionally, a negative correlation was found between serum KP and BMI, HOMA-IR, as well as the serum levels of insulin, glucagon, active ghrelin, and leptin, On the other hand, a positive correlation was found between KP and QUICKI index, McAuley index, and adiponectin levels[34]. The differences between the results of these studies may be related to the hormonal status of participants. The KP neurons located in the brain, and in the arcuate nucleus of the hypothalamus, are sensitive to sex steroids milieu and estrogen, which is due to the negative feedback influence of the neurons. In postmenopausal women, where the level of this hormone declines, this feedback will not be exerted properly. Unfortunately, although the above authors measured serum E_2 levels, they presented only the concentration range of this hormone, without dividing the study group into obese and nonobese participants. Additionally, in obese subjects, estrogen can be produced by fat. In a study on rats with obesity induced by a high-fat diet, increased E_2 concentration was found in blood[35]. This could be the result of enhanced aromatase activity because this enzyme is responsible for synthesizing estrogen from testosterone and androstenedione in the adipose tissue. Thus, increased adiposity related to obesity may lead to an increase in the activity of aromatase, and consequently higher E_2 concentrations[36]. Indeed, elevated expression and activity of aromatase were observed in the mammary gland of obese mice[37] and also in the inflamed breast tissue of overweight and obese women[38].

In humans, KP has been found in numerous organs such as the brain, placenta, testes, ovaries, pancreas, liver, adipose tissue, and small intestine[39-41], which suggests the pluripotent role of this peptide. However, clinical data on the associations between obesity, pregnancy, and KP levels are limited. It is well known that KP is important for a healthy pregnancy, especially for implantation and placentation[18,42]. Horikoshi et al[43] reported a dramatic (900-fold) increase in the plasma concentration of KP during the first trimester of pregnancy, with a further increase to over 7000-fold in the third trimester, in pregnant women compared to nonpregnant women. It was also shown that KP is produced in the human placenta and circulates in MB throughout pregnancy. Moreover, due to increased KP levels, corresponding increases in plasma E_2 and progesterone concentrations were found during pregnancy. Additionally, significant positive correlations were noted between the concentrations of KP and E_2 as well as P_4. It was also reported that KP concentrations in the plasma of umbilical cord arterial and venous blood were not statistically different[43]. In agreement with the above data, in the present study, we found that KP levels were higher in both MB and CB samples of obese women compared to nonobese women. However, in the case of both controls and obese groups, no statistically significant differences were noted between MB and CB.

Besides studying the impact of obesity on KP levels, we determined the biochemical profile of MB and changes in CB at delivery. As previously

shown, we found differences between cholesterol and TG concentrations in MB, whereas no differences were found in CB. This finding led us to analyze the differences in the biochemical profile of obese and nonobese mothers. Moreover, it was previously shown that the level of KP could be correlated with that of TG. A study conducted on healthy obese women and women with the polycystic ovarian syndrome showed that the level of KP negatively correlated with TG[34,44]. Interestingly our study indicated a reverse (positive) correlation between these parameters in MB. However, due to the limited sample size, we decided not to correlate the investigated factors with KP. The differences may result from the fact that our study group had a different physiological status—pregnancy.

On the other hand, research performed on mice indicated that KP injection did not cause changes in TG concentration[45]. Unfortunately, there is no direct scientific evidence showing the relationship between KP and TG levels. Therefore, it is not possible to determine whether changes in the level of KP are caused by the increase in TG or vice versa.

Lipids and steroid hormones are closely linked. The level of steroid hormones (progesterone and estrogen) as well as cholesterol content in women changes both during pregnancy and in the later stage (decreases after menopause)[46]. Cholesterol is one of the most important lipids in the context of reproductive functions due to, inter alia, the fact that it acts as a precursor of steroid hormones, and during pregnancy, as a substrate for (placental) steroid hormone synthesis. Previous studies[47,48] showed that KP stimulated progesterone secretion by rat luteal cells and by chicken and porcine granulosa cells. A study performed on obese women[49] showed that progesterone concentration was lower in obese pregnant women compared to nonobese pregnant women. Perhaps, the increase in KP level in obese women is a compensatory response to the reduced level of progesterone, in order to simulate its synthesis from the placenta. However, these are only assumptions and require further research.

Similar to the present study, a study on Polish pregnant women (n=194) found much lower (about 60%) levels of leptin in cord in obese women compared to nonobese women[13]. In another study performed in Germany with 766 nonobese mothers, the median leptin concentrations in CB were lower compared to maternal serum[50].

In our study, elevated leptin concentration found in obese mothers can be attributed to substantial increases in the amount of adipose tissue, which is a source of its synthesis[51-53]. Leptin is released into circulation in proportion to the amount of lipid stores and acts at hypothalamic receptors, decreasing food intake and increasing energy expenditure. Additionally, it is produced in the human placenta and secreted into both maternal and fetal circulation. However, the placental production of this adipokine makes a substantial contribution only to the maternal concentration of circulating leptin[54,55]. It has been reported that umbilical leptin concentration is independent of placental leptin production. Furthermore, it is well established that, as in obesity, leptin resistance in pregnancy may result from its inhibited transport across the blood–brain barrier or sequestration of bioactive leptin in the circulation by a soluble receptor[56]. A Swedish study (n=740) identified that maternal BMI is the best positive explanatory factor for maternal leptin levels. Similar results were obtained in a smaller study performed by Brazilian researchers[57]. Moreover, leptin was recognized as a strong positive explanatory factor for gestational weight gain[58].

Previous data as well as our results indicate that adiponectin level is higher in CB compared to MB. The same results were obtained by Weyermann et al[50] in their study on nonobese pregnant women. This suggests that fetal adipose tissue also produces adipokinin. In another study[59] performed on a Polish population of 38 pregnant women who were overweight/obese (BMI>25 kg/m^2) and 42 pregnant women of normal weight, between 24th and 34th weeks of gestation, an increased concentration of leptin but not adiponectin was observed in the former group[59]. This is in line with the present study as we also found a decrease in adiponectin concentrations in obese mothers. Importantly, similar to our study, Thagaard et al[60] indicated that low adiponectin concentration during the first trimester was associated with the development of gestational diabetes mellitus.

We also measured insulin in the studied groups and found a small but insignificant increase in its level in obese women. However, it should be emphasized that our samples size was relatively small. Similarly, in overweight and obese pregnant women, insulin levels were shown to be higher, but the differences were not statistically significant[57]. It is well known that hormones produced by the placenta, particularly in the second

and third trimesters of pregnancy, generally antagonize the action of insulin, thereby establishing insulin resistance along with increasing the production of this hormone[61,62].

In summary, our study showed a novel finding that obesity during pregnancy leads to an increase in KP levels in both MB and CB. The effects of the elevated KP levels in both mothers and offspring need to be further investigated. Such an investigation is especially important because it has been shown that human liver samples and serum collected from DM2 patients, who also often suffer from obesity, had increased levels of KP[63].

According to the concept of fetal programming, alterations in the hormonal profiles of mothers during pregnancy could have long-term health consequences in children. In particular, as KP regulates reproductive functions, it could affect puberty and other reproductive characteristics of offspring in later life. Moreover, besides the brain, KP is expressed in tissues responsible for metabolism (pancreas, liver, fat) and is considered to link metabolism and reproduction[19]. Thus, increased levels of KP in both MB and CB may lead to long-term metabolic changes manifested as obesity, as well as reproductive dysfunctions, in offspring. The hormonal outcomes of obese pregnant women also emphasized the importance of weight control during pregnancy to avoid adverse outcomes to mother and their newborns. Surprisingly, the results of the present study seem to be contradictory to our previous work in which we proved that KP levels were lower in obese women[34].

Conclusions

Taken together, our results indicate that, besides metabolic status, KP concentrations may be a valuable marker (or diagnostic tool) for predicting pregnancy complications. Based on the literature data and the present study, we assume that a high concentration of KP in MB and CB during pregnancy, as well as its increased concentration in obese mothers, may be a defense mechanism. This mechanism, to some extent, may prevent or reduce pathological changes caused by maternal imprinting in children in the future. However, this hypothesis requires further research.

Conflict of Interest

The Authors declare that they have no conflict of interests.

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Authors' Contribution

EPO, PAK, MW designed the study, obtained the data, and wrote the manuscript. JHS contributed to manuscript writing and critically revised the manuscript. MW, NL, DS and MS obtained, collected, and analyzed the data. HK, JP, LN, and KWN critically revised the manuscript.

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