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MATERNAL LIPIDS MAY PREDICT FETAL GROWTH IN TYPE 2 DIABETES MELLITUS AND GESTATIONAL DIABETES MELLITUS PREGNANCIES

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Abstract

Aim: During diabetic pregnancy, complex metabolic changes occur in the lipid profile. The aim of the study was to determine the predictive values of maternal serum lipid levels on large-for-gestational age newborns during the third trimester in pregnancies of women with type 2 diabetes mellitus (DM2) and gestational diabetes mellitus (GDM).

Material and methods: Data of forty three pregnancies of women with DM2 and two hundred women with GDM were analyzed. The analysis encompassed the following parameters: age, body mass index (BMI), lipid parameters, HbA1c in first, second and third trimester of pregnancy, preeclampsia and baby birth weight.

Results: DM2 and GDM groups showed statistically significant differences in the following variables: total lipids, triglycerides, total cholesterol, BMI, age, baby birth weight, incidence of SGA and preterm delivery $(9.4 \pm 2.3 \text{ vs. } 11.0 \pm 2.3 \text{ mmol/L}, 2.4 \pm 1.4 \text{ vs. } 3.4 \pm 1.6 \text{ mmol/L}, 5.5 \pm 1.2 \text{ vs. } 6.4 \pm 1.4 \text{ mmol/L}, 30.6 \pm 5.4 \text{ vs. } 26.9 \pm 5.2 \text{ kg/m}^2, 34 \pm 7.8 \text{ vs. } 31.5 \pm 5.6 \text{ years, } 3183 \pm 972 \text{ vs. } 3533 \pm 699 \text{ g., } 20\% \text{ vs. } 7.5\%, 27.9 \text{ vs. } 14\%, \text{ respectively, p < 0.05}). Linear multiple regression analysis demonstrated that triglycerides, LDL-C and total cholesterol were independent predictors of LGA (p < 0.05).$

Conclusion: Triglycerides and LDL-C in the third trimester of pregnancy are independent predictors for fetal macrosomia in DM2 and GDM pregnancies. Thus, the maternal serum triglycerides and LDL-C levels determined in the maternal blood taken in the third trimester of pregnancy may indentify women who will give birth to LGA newborns.

Key words: lipid parameter, triglycerides, type 2 diabetes mellitus, gestational diabetes mellitus, fetal macrosomia

1. Introduction

Macrosomia still complicates a signifycant proportion of diabetic pregnancies. Studies show that variations in birth weight which occur more often in diabetic pregnancies are not determined only by the maternal glycemic state, but other metabolic factors as well, such as lipids, may have important influence [1–4].

Maternal lipid metabolism is altered during normal and diabetic pregnancy with increased insulin resistance combined with increased peripheral adipose tissue lipolysis. It re-

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sults in increased maternal lipoprotein concentrations [5, 6]. The abnormal lipid metabolism has been associated with preterm delivery, preeclampsia and macrosomia. Furthermore, Freinkel et al. [7] proposed that "mixture" of maternal nutrients (glucose, amino acids and lipids) changes the metabolic environment of the fetuses and these changes not only that affect the fetal growth and development but also influence future obesity, diabetes and neurocognitive development in the offspring ("fuel mediated-teratogenesis"). Lipids treatment is the key therapeutic target in non-pregnant diabetic settings. There are no recommendations for clinic management of lipids in pregnancy complicated by diabetes. Studies show lipid changes in the first, second and third trimester both in normal [8] as well as in diabetic pregnancies [9]. There is an association between insulin resistance and serum lipid levels in pregnancy, as adaptive mechanism allowing fetal grown. Serum lipids are higher in obesity, gestational diabetes mellitus (GDM), type 1 diabetes mellitus (DM1), and type 2 diabetes mellitus (DM2). More, but not all studies have reported the same lipid profiles in wellcontrolled DM1 as in normal pregnancies, high triglyceride and low HDL-C are found in DM1 women with metabolic syndrome in pregnancy, and those diabetic women who develop preeclampsia have high LDL-C and cholesterol levels. In pregnant GDM women frequent founding are the elevated triglycerides while in DM2 pregnancies the lipid profiles are between those observed in DM1 and GDM [9, 10]. Hence, we decided to explore lipids in DM2 and GDM pregnancies because the latter is a form of type 2 diabetes and they share the same underlying pathophysiology (insulin resistance and beta cell dysfunction).

The aim of the present study was to determine the contribution of maternal lipids in predicting (large-for-gestational age) macrosomic newborns in pregnant women with DM2 and GDM, with emphasis on the intergroup differences and development of macrosomia.

2. Material and methods

2.1. Study subjects

The study was conducted at the University Clinic of Endocrinology, Diabetes and Me-

tabolic Disorders. Data of 43 pregnant women with DM2 and 200 women with GDM were analyzed. All were with singleton pregnancies, and neonates were delivered at the University Clinic of Gynecology and Obstetrics. The diagnosis of DM2 was made when patients have been treated with oral lowering medication before conception, were switched to insulin before or at early pregnancy, and without evidence of ketoacidosis. GDM was generally diagnosed by OGTT in the second part of pregnancy. The OGTT was performed in the morning after an overnight fasting from 8 to 12 h. The criteria for diagnosis of GDM were at least one abnormally high out of three plasma glucose value measurements during the 75 g OGTT (normal values: a fasting level < 5.1, 1-hour level < 10.0, 2-hour level < 8.5 mmol/l). The venous blood glucose levels were measured using the glucose oxydase method (Glucose Analyzer; Beckman, Brea, CA). The glucose tolerance was classified by the latest criteria of the International Association of Diabetes in Pregnancy Study Group (IADPSG) [11].

All patients gave informed consent to participate in the study after a careful explanation of the testing protocol. The study was made in accordance with the Declaration of Helsinki.

2.2. Measurements

The analyzed outcome measures were: age, BMI, lipid parameters in the third trimester, HbA1c in the first, second and third trimester of the pregnancy, preeclampsia, and baby birth weight. The maternal glycemic parameters included glycosylated hemoglobin (HbA1c) in all three trimesters. HbA1c was measured with an ionexchange HPLC instrument (DS5; Drew, USA) with a reference range of 4.2–6%. In the third trimester the fasting maternal total cholesterol, triglycerides, highdensity lipoprotein cholesterol (HDL-C), lowdensity lipoprotein cholesterol (LDL-C) were measured in both groups. The lipids assessment was taken after the overnight fasting. The blood samples for lipoproteins were analyzed using Cobas Integra 700, according to the standard methods. Total cholesterol and triglycerides were determined with the full enzymatic methods (TCH-CHOD-POD-PAP and triglycerides-GPO; Cobas Integra 700, Hoffmann-La Roche, Basel, Switzerland). The high density lipoprotein cholesterol (HDL-C) was measured using the polyanion precipitation method, while LDL-C was calculated using the Friedewald formula. The low density lipoprotein cholesterol (LDL-C) was fractioned using ultracentrifugation in the cases of triglycerides exceeding 4mmol/l. The body mass index (BMI) of women with type 2 diabetes and GDM was calculated by dividing the weight with the height squared (kilograms/metar²). The BMI of 26 kg/m² was defined as overweight and 30 kg/m² as obesity.

The baby birth weight was measured immediately after delivery. The macrosomia was defined as birth weight greater than 4000 g; the large for gestational age (LGA) was defined as weight greater than the 90th percentile for gestational age and sex. The small for gestational age (SGA) was defined as birth weight lower than 2700 g. or the 10th percentile for gestational age and sex. The gestational age was estimated from the date of the last menstrual period. The pre-term delivery was the birth before 37 weeks of pregnancy.

The patients with DM2 were treated only with insulin. The patients with GDM were

Table 1

treated with diet, metformin or insulin according to glycemic profiles.

2.3. Statistical analyses

The statistical analysis was performed using Statistics 7.0 version for Windows. The normal distribution of all variables was verified using Kolmogorov-Smirnov test. The data were presented as mean values, median and percentage. The comparisons between the two groups were determined with the t-test for independent samples. The differences in proportions were compared with the Chi-square test. The correlations were determined by the Pearson correlation test. To estimate the independent effect of analyzed variables on LGA, linear multiple regression analysis was done. P < 0.05 was considered to be statistically significant.

3. Results

The study group consisted of 43 women with DM2 and 200 with GDM. The maternal characteristics of the diabetic pregnancies are listed in Table 1. The investigated groups were statistically different in the following variables: total lipids, triglycerides, total cholesterol, BMI, age, baby birth weight, and incidence of SGA (Table 1).

The comparison of analyzed variables between the groups (DM2 and GDM)

	DM2	GDM	p value
Total lipids (mmol/L)	9.4 ± 2.3	11.0 ± 2.3	< 0.01
Triglycerides (mmol/L)	2.4 ± 1.4	3.4 ± 1.6	< 0.01
Total cholesterol (mmol/L)	5.5 ± 1.2	6.4 ± 1.4	< 0.01
HDL-C (mmol/L)	1.4 ± 0.3	1.5 ± 0.4	NS
LDL-C (mmol/L)	3.1 ± 1.0	3.5 ± 1.2	NS
HbA1c I trimester (%)	6.6 ± 1.5	7.1 ± 2.2	NS
HbA1c II trimester (%)	5.9 ± 1.1	5.8 ± 0.9	NS
HbA1c III trimester (%)	5.8 ± 1.0	5.8 ± 1.0	NS
BMI kg/m ²	30.6 ± 5.4	26.9 ± 5.2	< 0.05
Age (years)	34 ± 7.8	31.5 ± 5.6	< 0.05
Baby weight (g.)	3183 ± 972	3533 ± 699	< 0.05
Preeclampsia	5/42 (11.9%)	8/155 (5.2%)	NS
LGA	9/35 (25.7%)	44/187 (23.5%)	NS
SGA	7/35(20%)	14/187 (7.5%)	< 0.05
g.w. of delivery (median)	37.5	39	NS
preterm delivery	12/43 (27.9%)	28/200 (14%)	< 0.05

All data are presented as mean±SD, median and percentages; DM2: type 2 diabetes mellitus; GDM: gestational diabetes mellitus; HDL-C: high density lipoprotein cholesterol; LDL-C: low density lipoprotein cholesterol; HbA1c: glycosylated hemoglobin; BMI: body mass index; LGA: large for gestational age; SGA: small for gestational age; g.w.: gestational week; NS: not significant.

The mean age of patients with DM2 and GDM were 34 ± 7.8 and 31 ± 5 years, respectively, showing statistically significant difference (p<0.05). The patients with DM2 were heavier (mean BMI 30.6 ± 5.6 kg/m²) in comparison to the GDM patients (mean BMI 26.9 ± 5.2 kg/m²) and the difference was statistically significant (p < 0.05).

3.1. Glycemic control

The maternal HbA1c values were similar in the DM2 patients and the GDM patients; there was no difference between the groups during the first, second and third trimester (Table 1), yet all values indicated generally acceptable glycemic control.

3.2. Lipids

The plasma total lipids were lower in the DM2 (mean 9.4 ± 2.3 mmol/L) in comparison to the GDM patients (mean 11.0 ± 2.3 mmol/L), this difference was significant (p < 0.01). Statistically significant differences were found between the GDM patients and the DM2 patients in the mean plasma triglycerides concentrations (3.4 ± 1.6 and 2.4 ± 1.4 mmol/L, respectively), and the mean total cholesterol concentrations (6.4 ± 1.4 and 5.5 ± 1.2 mmol/L), (p < 0.01), whereas HDL-C and LDL-C concentrations were similar in both groups, using the National Education Program criteria in the non-pregnant state.

3.3. Neonate size

Table 2

The pregnancy outcomes are shown in Table 1. The babies who were born by mothers

with GDM had a mean birth weight of 3533 ± 699 g, they were heavier than those of mothers with DM2 (3183 ± 972 g) and the statistical analyses showed significant differences between the two groups (p < 0.05) (Table 1.) The rate of macrosomia was comparable between two groups: 25.7% and 23.5% in DM2 and GDM groups, respectively.

3.4. Interrelationships between metabolic control, lipids and neonatal size (macrososmia /large-for-gestation age)

Triglycerides concentrations were directly related to the HbA1c levels (r = 0.18, p < 0.05) and the HDL-C concentrations were associated with the values of HbA1c (r = 0.19, p < 0.05). The HDL-C concentrations were related to the newborn size, i.e. large-for-gestation age newborns (r = 0.17, p < 0.05). The HbA1c levels were associated with the small-for-gestation age newborns (r = 0.29, p < 0.05). The BMI was related with the LGA (r = 0.173, p < 0.01), preeclampsia (r = 0.228, p < 0.01), triglycerides (r = 0.137, p < 0.05), and age (r = 0.202, p < 0.01).

The linear multiple regression analysis demonstrated that, triglycerides, LDL-C and total cholesterol concentrations were independent predictors of LGA (p < 0.05) (Table 2).

There were no independent relationships with the HDL-cholesterol concentrations and the HbA1c levels with the LGA. Surprisingly, the BMI was not independent predictor for LGA, as well as preeclampsia.

Multivariate analyses of maternal LDL-C, triglycerides, and total cholesterol in the third trimester as independent predictors of neonate size (LGA)

	Unstandardized Coefficients		Standardized Coefficients	t value	p value
	В	Std. Error	Beta		
(Constant)	-0.072	1.019		-0.071	0.944
Triglycerides	0.253	0.119	0.608	2.133	0.043
Total cholesterol	-0.428	0.186	-1.161	-2.298	0.031
HDL-C	0.047	0.299	0.033	0.156	0.877
LDL-C	0.538	0.215	1.184	2.509	0.019
BMI	0.007	0.019	0.068	0.348	0.731
Hba1c I trimester	-0.073	0.059	-0.278	-1.238	0.228
Hba1c II trimester	0.118	0.156	0.200	0.754	0.458
Hba1c III trimester	0.007	0.137	0.013	0.053	0.958
Preeclampsia	-0.260	0.253	-0.197	-1.027	0.315

Dependent Variable: LGA; HDL-C: high density lipoprotein cholesterol; LDL-C: low density lipoprotein cholesterol; BMI: body mass index; HbA1c: glycosylated hemoglobin.

4. Discussion

In this retrospective study on pregnant women with DM2 and GDM, we found maternal triglyceride and cholesterol values in the third trimester of gestation to be important predictors for macrosomia, independent of the maternal pregnancy BMI and HbA1c values.

Macrosomia occurs in 30-56% of the pregnancies complicated by DM2 and 10-20% of the pregnancies complicated by GDM, according to the data from the literature. In this study, the frequency of macrosomia was similar in the DM2 and the GDM patients partly due to "good" glycemic control. HbA1c values were better than those found in the literature which emphasizes that in the glycemic control "almost good is not good enough". Further, in a recent study we found no correlation between HbA1c and fetal macrosomia. It means that HbA1c is not a sensitive marker for prediction of macrosomia, or that the strict glycemic control can fail to prevent macrosomia. The findings in our study showed that fetal growth, behind maternal glucose concentration is influenced also, by lipids. Many studies [12, 13] observed that maternal serum lipids are associated with baby birth weight, independent of maternal weight gain and glucose.

The normal pregnancy is a hyperlipidemic state mainly influenced by increments of estrogen [8]. The rising levels of estrogen with the progression of pregnancy reflects the changes in lipoproteins [9]. On the other hand, dyslipidemia increases with the insulin resistance found in obesity, GDM and preexisting diabetes, as it has been extensively researched in the studies of Kitajima et al. [10], Knopp et al. [14] and Gobi et al. [15]. Hyperlipidemia in diabetes might also affect the fetal growth. In this paper we focused on lipids in the third trimester in pregnant women with DM2 and GDM. Key points were macrosomia and preeclampsia. We found higher levels of total cholesterol (6.4 \pm 1.4 mmol/l) in the GDM patients in comparison to the levels $(5.5 \pm 1.2 \text{ mmol/L})$ of the DM2 patients. Further, the multivariate analysis showed the positive correlation between the maternal cholesterol levels and the neonatal birth weight. Actually, the maternal cholesterol levels were independent predictor for macrosomia. This is in line with Schaefer et al. [16] who observed a positive correlation between the maternal cholesterol levels in the third trimester and the neonatal birth weight. Some studies have reported that lipids play an important role in baby weigh as compared to glucose [12, 13].

The most consistent lipid change in all studies is the level of triglycerides which shows progressive rise from the first, through the second, and to the third trimester [15]. The increase in the lipid metabolism is an adaptation of the increased fuel delivery to the fetus. When they are too high, macrosomic babies are more likely to be born. Mothers with GDM had higher triglycerides levels $(3.4 \pm 1.6 \text{ mmol/L})$ in comparison to the triglyceride levels in the DM2 patients $(2.4 \pm 1.4 \text{ mmol/L})$ (p < 0.01). In this study, the triglycerides in the third trimester were the strongest predictor of macrosomia. The increased triglycerides during the late pregnancy are due to the increased hepatic production of VLDL under the stimulation of estrogens and increased adipose tissue lipolysis.

The low density lipoprotein cholesterol and VLDL increase in parallel to the estrogen. HDL-C levels did not change dramatically during pregnancy, i.e. HDL-C increases slightly and remains high or in some patient goes down, but basically normal to delivery [8]. In the study that was conducted by Yang et al. [17], a negative correlation was found between the HDL-C levels and the neonatal birth weight. In the present study we stated that the negative correlation was found between HDL-C and LGA newborns (r = -0.17, p < 0.05), but without independent influence on LGA, when other lipid parameters were added. HDL-C values were in the normal range according to NCEP [18] in both groups. LDL-C levels were above the normal range in both diabetic pregnant women, and the linear multiple regression analysis revealed their predictive value for LGA newborns.

Lipids exhibit some adverse effects on the mother. Women with preeclampsia have high serum lipids in pregnancy. Hyperlipidemia might favor production of lipid peroxides and alter the balance of the vasoactive compounds leading to endothelial dysfunction i.e. vasoconstrictions. GDM patients are at increased risk for preeclampsia and it is equal to that seen in the insulin resistant pregnant women with DM2. Our findings are very supportive to this notation. The rate of preeclampsia was 11.5% and 5.2% in DM2 and GDM patients, respectively. Patients with DM2 were older and obese. Patients with GDM were overweight and had worst lipid profiles. The babies born by mothers with GDM were heavier in comparison to the babies of the DM2 patients. On the other side, not all GDM mothers were treated with insulin. Lower birth weight in DM2 than in GDM pregnancies might result from better Brankica Krstevska, et al.

glycemic control throughout the pregnancy and insulin therapy starting before or in early pregnancy. In spite of that, mild hypertriglyceridemia and hypercholesterolemia were found, whereas the HDL-C levels were in the normal range according to NCEP [18]. The proportion of SGA in DM2 was higher than in GDM, due to the higher percentage of preterm delivery in DM2. Probably preeclampsia was the main reason for preterm delivery.

Obesity is a significant risk factor for both DM2 and GDM and may be related to the fetal size and adverse perinatal outcomes. In this study DM2 patients were obese and GDM patients were overweight. Almost all studies show the effects of obesity on newborn macrosomia [19, 20]. In this study the BMI was statistically significantly correlated with LGA, but without an independent effect on LGA. This finding can be explained by the lack of normal weight pregnant women in the observed cohort. Actually it is one limitation of this study.

5. Conclusion

This study pointed out the usefulness of measurement of the serum lipids in pregnancy. Triglycerides and LDL-C in the third trimester of pregnancy are independent predictors for fetal macrosomia in DM2 and GDM pregnancies. Maternal serum triglycerides and LDL-C levels determined in the maternal blood taken in the third trimester of the pregnancy may identify women who will give birth to LGA newborns. With proper regulation of the lipid profile we can avoid macrosomia in DM2 and GDM pregnancies.

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Резиме

МАЈЧИНИТЕ ЛИПИДИ МОЖЕ ДА ГО ПРЕДВИДАТ ФЕТАЛНИОТ РАСТ КАЈ БРЕМЕНИ ЖЕНИ СО ДИЈАБЕТЕС МЕЛИТУС ТИП 2 И ГЕСТАЦИСКИ ДИЈАБЕТЕС МЕЛИТУС

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Цел: Комплексни метаболни промени во липидниот статус настануваат во тек на бременост кај жени со дијабетес. Целта на истражувањето беше да се открие предиктивната вредност на серумските липиди од третиот триместар на бременост кај жени со дијабетес тип 2 (DM2) и гестациски дијабетес (GDM) за раѓање на новородено големо за гестациската возраст (LGA).

Машеријал и мешооји: Анализирани беа податоци за 43 бремени жени со DM2 и 200 жени со GDM. Анализите ги вклучуваа следниве параметри: возраст, индекс на телесна маса (BMI), липиден статус, HbA1c во првиот, вториот и третиот триместар од бременоста, прееклампспија и телесна тежина на новородено.

Резулшаши: DM2 и GDM-групите покажаа статистички значајни разлики во следниве параметри: вкупни липиди, триглицериди, вкупен холестерол, BMI, возраст, телесна тежина на новородено, инциденца на мало за гестациска возраст (SGA) и предвремено пораѓање: 9.4 ± 2.3 vs. 11.0 ± 2.3 mmol/L, 2.4 ± 1.4 vs. 3.4 ± 1.6 mmol/L, 5.5 ± 1.2 vs. 6.4 ± 1.4 mmol/L, 30.6 ± 5.4 vs. 26.9 ± 5.2 kg/m², 34 ± 7.8 vs. 31.5 ± 5.6 years, 3183 ± 972 vs. 3533 ± 699 g., 20% vs. 7.5%, 27.9 vs. 14%, соодветно, p < 0.05). Линеарната мултипна регресиона анализа покажа дека триглицеридите, LDL-С и вкупниот холестерол беа независни предиктори за LGA (p < 0.05).

Заклучок: Триглицеридите и LDL-С во третиот триместар од бременоста се независни предиктори за фетална макросомија во групите бремени жени со DM2 и GDM. Одовде, серумските триглицериди на мајката и нивото на LDL-С во крвта на мајката во третиот триместар од бременоста може да ги откријат жените кои раѓаат LGA-новородени.

Клучни зборови: липидни параметри, триглицериди, дијабетес тип 2, гестациски дијабетес, фетална макросомија