

Pregnancy, gestational diabetes and obesity: what is the relationship?

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ABSTRACT

Background: During pregnancy, body composition and carbohydrate metabolism undergo changes, mainly to meet the needs of the foetus. Unfortunately, pathological disturbances may occur.

Aim: To study the changes in body composition during pregnancy and to assess the relationship between body composition and gestational diabetes mellitus (GDM).

Methods: This study included 200 female volunteers aged 18 to 45 years (150 pregnant women and 50 control women). Pregnant women were referred for a 75-g oral glucose tolerance test (OGTT) as part of the screening for GDM. Anthropometric and clinical data were collected through a questionnaire. OGTT and bioelectrical impedancemetry were performed to each woman.

Results: GDM was found in 44% of the pregnant women of whom 31.5% had overweight and 17.6% were obese before pregnancy. Upon studying of body composition of pregnant, the percentage of fat mass was significantly higher than the percentage of free-fat mass. Concerning the OGTT results, we found a positive correlation between T0 min, T60 min, and T120 min blood glucose levels and body mass index before pregnancy and upon study inclusion. Fasting blood glucose values were also significantly correlated to fat mass.

Conclusion: This study determined that obesity is a risk factor for GDM, and confirmed that there is a relationship between glucose values and body composition parameters in pregnant women with GDM.

KEYWORDS

Gestational diabetes mellitus, pregnancy, obesity, body composition, impedancemetry, oral glucose tolerance test.

Introduction

Gestational diabetes mellitus (GDM) is a worldwide health-care problem. It is defined as any level of dysglycemia with onset or first acknowledgment during gestation^[1-3]. The incidence of diabetes in pregnant women increased by 9% annually between 2011 and 2017^[4]. This prevalence is affected by several factors, namely obesity^[5]. During pregnancy, there is also an adaptive physiological change in maternal body composition that helps support the fetus and prepare the mother for breastfeeding^[6]. Indeed, fat mass (FM), lean mass or fat-free mass (FFM), and total body water mass (TBWM) increase in different ways, and their effects on newborns represent a major area of interest in perinatal medicine^[7]. The present study aimed at studying changes in body composition during pregnancy and to investigate the relationship between the results of impedancemetry and the 75-g oral glucose tolerance test (OGTT) in pregnant women.

Methods

Subjects

We included 200 female volunteers aged between 18 and 45 years (150 pregnant women and 50 control women). Pregnant women were referred to our department for an OGTT as part

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of a screening for GDM. We excluded women with diseases or treatments known to have metabolic effect. Controls were healthy women part of our medical and paramedical staff. Those on oestro-progestogenic pills, and athletic women were excluded. For both groups, women with unknown pre-pregnancy weight and imprecise dates of the last menstrual period were also excluded.

Ethical approval

All women participating in this study had given consent for all questionnaires and exams conducted to be used for the study. No extra ethical approval was necessary since all exams were part of the systematical care services offered to women during pregnancy.

Questionnaire

Women were asked about their age, geographic location, and occupation. Personal medical histories were detailed. Risk fac-

tors for GDM were assessed according to the recommendations of the French College of Gynecologists and Obstetricians [8]: history of diabetes mellitus in first-degree relatives, maternal age > 35 years, body mass index (BMI) before pregnancy > 25 Kg/m², personal history of GDM, history of fetal macrosomia. Suggestive clinical signs of diabetes were also investigated.

Anthropometric measurements

Height, weight, hip circumference (HC), and uterine height (UH) were measured. BMI was calculated by dividing weight (Kg) by height (m) squared. Pre-pregnancy weight was provided by the interview or noted from the pregnancy follow-up notebook.

Obesity definition

According to WHO, obesity in pregnant women is defined by a BMI ≥ 30 Kg/m² referring to the woman's height and weight before pregnancy or those measured during the first prenatal visit if not available before pregnancy [9].

Impedancemetry

Impedancemetry corresponds to the analysis of the human body compartments. Bioelectrical impedance analysis, based on the capacity of tissues to conduct electrical energy, allows the measurement of the resistance of biological tissues by sending a sinusoidal electrical signal of low intensity and high frequency (10-100 kHz) through electrodes [10]. The impedancemeter used in this study was the Tanita TBF-410. The results provided by impedancemetry were BMI, weight, FM expressed in Kg, FFM expressed in Kg, TBWM expressed in Kg, and basal metabolic rate. The percentages of FM, FFM and TBWM were calculated according to the following formulas:

$$\text{FM}\% = \text{FM (Kg)} / \text{total body weight} \times 100;$$

$$\text{FFM}\% = 100\% - \text{FM}\%$$

$$\text{TBWM}\% = (\text{TBWM (Kg)} / \text{FFM (Kg)}) \times 100$$

OGTT for pregnant women

Pregnant women should have fasted 8 to 14 hours the day before the test without following a special diet. A first blood sample was taken before ingesting the glucose solution (T0 min). Then, women were asked to drink 250 ml of 30% concentrated glucose serum (equivalent to 75 g of glucose) in less than 5 minutes. Two more blood glucose tests were taken at T60 min and T120 min. The samples are mixed with Sodium Fluoride/Potassium Oxalate for prolonged storage (> 2 h) and glycolysis *in vitro* prevention.

The test was stopped in case of intolerance, fasting blood glucose ≥ 1.26 g/L (type 2 diabetes), blood glucose ≤ 0.70 g/L with clinical signs of hypoglycemia, or blood glucose < 0.54 g/L.

Diagnosis of GDM was confirmed if at least one value was greater than or equal to the standards: T0 min ≥ 0.92 g/L and/or T60 min ≥ 1.8 g/L and/or T120 min ≥ 1.53 g/L [7].

Statistical analysis

Statistical analysis was performed with the Statistical Package

for the Social Sciences software (SPSS13). A descriptive study for qualitative and quantitative variables was performed. Normality of data distribution was checked with the Shapiro-Wilk test. For the analytical study, Chi 2 test, Student's T test and ANOVA were used for normally distributed parameters. Fisher's exact test, the Mann-Whitney U test and the Kruskal and Wallis tests were used for non-normally distributed parameters. Differences were considered statistically significant when the value of p was < 0.05.

Results

We divided our population into 3 groups: NGD: pregnant women without GDM ($n=84$), GD: pregnant women with GDM ($n=66$), and CG: control group ($n=50$). Data collection, OGTT and impedancemetry were performed at a mean term of 26 ± 3.7 weeks of gestation.

General data

The 3 groups were comparable in terms of geographical origin and age. Most women of our population were housewives with no significant difference between the 3 groups.

Medical history

First degree familial diabetes was found higher in the GD group as compared to the NGD and CG groups (28% vs. 8.3% and 12.1%, respectively, $p < 0.01$). The most common medical history was high blood pressure in the control population ($n=6$) and anemia in both groups of pregnant women ($n=4$). The 3 groups were comparable in the number of gestations and parity.

Anthropometric data

The 3 groups were matched for height. We did not find a significant difference between the pre-pregnancy weight in the first two groups compared to CG (NGD: 64.9 ± 12.6 Kg, GD: 67.0 ± 13.2 Kg, and CG: 65.9 ± 14.0 Kg; $p > 0.05$). However, a significant difference was found for baseline weight and baseline BMI (Table 1).

For all pregnant women pregnant women, 31.5% were overweight and 17.6% obese before pregnancy. The mean weight of the NGD group was lower than that of the GD group yet not significant (72.4 ± 12.6 Kg vs. 76.1 ± 14.3 Kg, respectively, $p > 0.05$). There was no significant difference in gestational weight gain between both studied pregnant groups (NGD: 7.46 ± 5.39 Kg and GD: 8.35 ± 4.64 Kg, $p > 0.05$).

Also, uterine height was significantly greater in GD women compared to NGD ones (32.7 ± 4.1 cm vs. 34.1 ± 3.6 cm, respectively, $p < 0.05$). Table 1 summarizes the anthropometric data for the 3 groups.

Risk factors for GDM

Rate of risk factors for GDM were present among 60% of NGD and 74.2% of GD and were significantly higher, in number, among the GD group (1.34 ± 1.08 vs. 0.98 ± 1.02 , respectively, $p < 0.05$). Personal history of GDM was reported in 7.1% of NGD women and 16.7% of GD women. No significant difference was noted between the 2 groups of pregnant women for

Table 1 Anthropometric data of pregnant women and controls.

PARAMETERS	NGD	GD	CG	p VALUE
Weight (Kg)	72.4 ± 12.6	76.1 ± 14.3	65.9 ± 14.0	< 0.01* / 0.09**
Height (m)	1.6 ± 0.06	1.6 ± 0.07	1.6 ± 0.05	0.83* / 0.45*
BMI at baseline (Kg/m ²)	27.8 ± 5.2	29.4 ± 4.9	24.6 ± 6.0	< 0.01* / 0.06**
HC (cm)	109.1 ± 9.1	111.0 ± 9.7	105.8 ± 10.2	0.12* / 0.26**
Pre-pregnancy weight (Kg)	64.9 ± 12.6	67.0 ± 13.2	–	0.34**
Pre-pregnancy BMI (Kg/m ²)	24.8 ± 5.2	25.8 ± 4.5	–	0.24**
Weight gain during pregnancy (Kg)	7.5 ± 5.4	8.4 ± 4.6	–	0.50**
Uterine height (cm)	32.7 ± 4.1	34.1 ± 3.7	–	0.036**

*: comparison between 3 groups; **: comparison between NGD and GD
 BMI, Body Mass Index; HC, Hip circumference; NGD, pregnant women without gestational diabetes mellitus (GDM); GD, pregnant women with GDM; CG, healthy non pregnant control women; NS: non-significant.

Table 2 Body composition of pregnant women and controls.

	NGD	GD	CG	p value	r
FM (Kg)	26.3 ± 8.6	29.4 ± 10.3	21.8 ± 9.6	<0.001* / 0.09 **	0.22
FM (%)	36.0 ± 5.9	37.5 ± 6.8	31.3 ± 7.6	<0.001* / 0.13 **	0.20
FFM (Kg)	45.8 ± 4.8	46.8 ± 4.8	44.2 ± 4.9	0.010* / 0.81**	
FFM (%)	64.0 ± 5.9	62.5 ± 6.8	68.7 ± 7.6	<0.001* / 0.12 **	
TBW (Kg)	33.6 ± 3.5	34.3 ± 3.4	32.2 ± 4.0	0.003* / 0.13**	
TBW (%)	73.2 ± 0.2	73.2 ± 0.2	73.2 ± 0.1	0.89* / 0.37**	

*: comparison between 3 groups; **: comparison between NGD and GD
 FM, fat mass; FFM: fat-free mass; TBWM, total body water mass; NGD: pregnant women without gestational diabetes mellitus (GDM); GD: pregnant women with GDM; CG, healthy non pregnant control women; NS: non-significant

the following 2 risk factors: pre-pregnancy BMI > 25 Kg/m² and maternal age > 35 years. However, we noted a positive correlation between BMI of pregnant women at baseline and weight gain ($p < 0.001$; $r = 0.27$).

Impedancemetry: interpretation of body composition

FM and FFM in percent of body mass and in kilograms were higher in GD than other groups with positive correlations. However, TBW was significantly different among the three groups only in Kg but not in percentage (Table 2). Uterine height and FM (%) were also positively correlated ($p < 0.001$; $r = 0.77$).

In pregnant women with a pre-pregnancy BMI < 25 Kg/m², the percentage of FFM was higher compared to those who had overweight or were obese with, respectively, (67.4 ± 5.2% vs. 59.2 ± 4.1%, respectively, $p < 0.001$). We noted a negative correlation between FFM (%) and weight gain ($p < 0.05$; $r = -0.20$).

Diagnosis of GDM by OGTT: analysis of values

GDM was diagnosed in 44% of pregnant women based on high level of T0 blood glucose. The OGTT values according to the T0 min, T60 min, and T120 min schedule are shown in Table 3.

Fasting blood glucose values were positively correlated

to age ($p < 0.05$; $r = 0.25$) and to pre-pregnancy BMI ($p < 0.05$; $r = 0.18$) (Figure 1). However, no significance was found for a BMI being superior to 25 Kg/m².

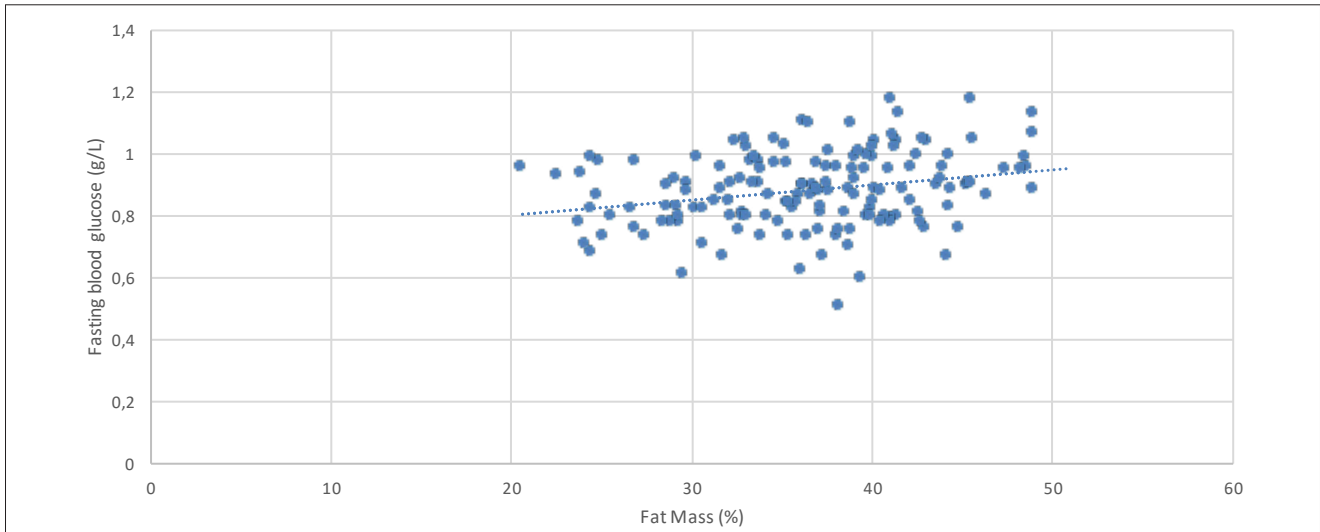
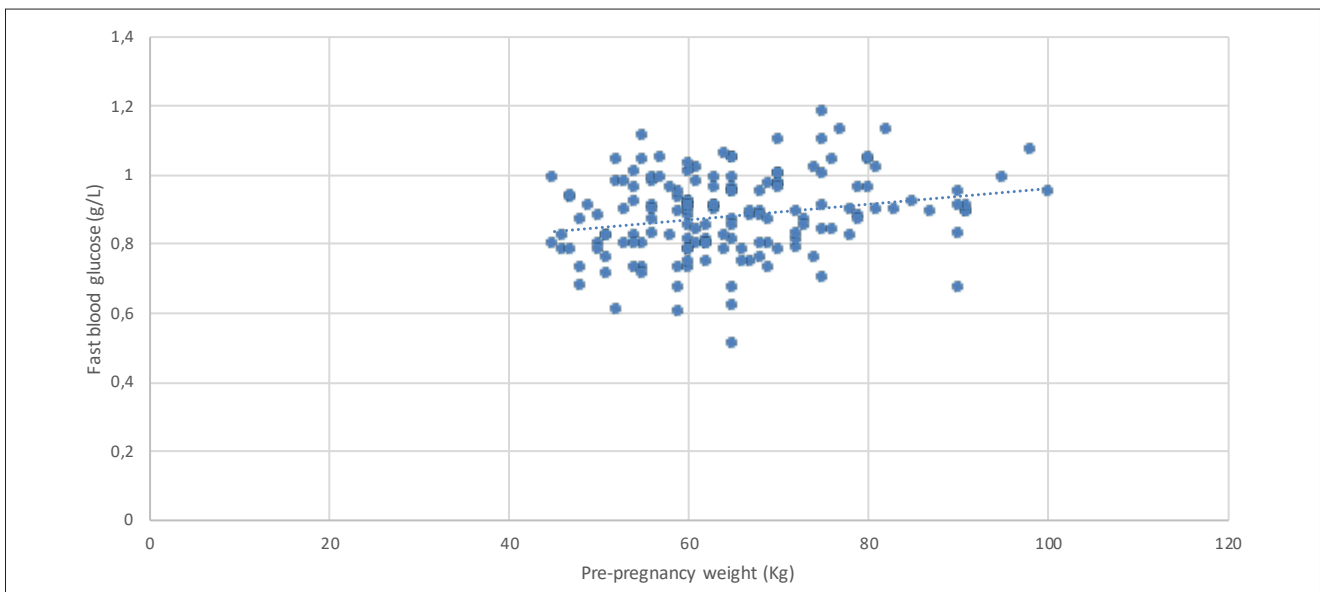
Relationship between OGTT results and impedancemetry results

Blood glucose levels at T0 min were lower in pregnant women with a higher FFM (%) ($p < 0.05$; $r = -0.23$). Pregnant women with higher FM (%) values had higher fasting blood glucose levels ($p < 0.05$; $r = 0.23$) (Figure 2).

Table 3 Oral glucose tolerance test values of NGD and GD groups.

	NGD	GD	p value
T0 min	0.8 ± 0.1	1.0 ± 0.1	<0.001
T60 min	1.2 ± 0.3	1.5 ± 0.4	<0.001
T120 min	1.0 ± 0.2	1.2 ± 0.3	<0.001

T0 min, fasting value; T60 min: 1-hour value; and T120 min: 2-hour value; NGD, pregnant women without gestational diabetes mellitus (GDM); GD: pregnant women with GDM.

Figure 1**Figure 2**

Discussion

Body composition and pregnancy

Prevalence of obesity

A study published in 2018 showed that obesity rate has increased worldwide to 100 million among women of reproductive age, with 250 million classified as being overweight^[9]. According to a Tunisian study published in 2012 and 2013, obesity among women was 37%^[11,12]. In our study, 12.2% of women of CG were obese and 26.53% were overweight. For pregnant women, 17.6% were obese and 31.5% were overweight before pregnancy. This little decrease compared to general population, could be explained by the relatively young age of our population.

Changes in body composition during pregnancy

Among the pregnant women in our population, FM (%) was

significantly higher than FFM (%). According to literature, all pregnant women display a gain in total weight, FM, and TBWM during pregnancy in similar percentages^[6]. Pregnancy products account for approximately 35% of weight gain^[13]. Catalano *et al.*^[14] have shown that the increase of FM is mostly seen among thin women, situation that was also found in our study.

According to the literature, white adipose tissue represents up to 20-30% of body weight in non-obese and non-pregnant women^[15]. In our study, the percentage of fat in control women was 31.3% and 36.8% in pregnant women. FFM increases during pregnancy with highly variable mean values^[16]. In our study, we found that in pregnant women, FFM (Kg) increased compared to CG. However, we did not find a significant difference in FFM between NGD and GD women. We also found that TBWM (%) was comparable between the 3 studied groups. However, TBWM (Kg) was higher in pregnant wom-

en. According to Mbungu *et al.* [16], the evolution of pregnancy is characterized by a very significant increase in the TBWM during the last trimester of pregnancy, which is explained by the physiological retention of water that allows maternal body improve the utero-placental fetal flow.

GDM and pregnancy

Prevalence of GDM

According to the literature, the prevalence of GDM is difficult to estimate. It depends on the screening method and diagnostic criteria used in each country. Published global prevalence figures may range from 1.7% to 11.7% [17]. According to the latest estimation from the International Diabetes Federation in 2017, GDM affects about 14% of pregnancies worldwide, or about 18 million births per year, using IADPSG diagnostic criteria [18]. In the present study, the prevalence of GDM was 44%. In fact, pregnant women who were referred to our department for OGTT generally had risk factors for GDM. Our study is not based on the entire population of pregnant women, which may explain in part the relatively high prevalence of GDM.

Risk factors for GDM

Age was not correlated to GDM in our study when comparing 3 groups. However, it was positively correlated to glycemic values without a threshold. This can be related to variability of risk factors among societies. Actually, Farrar *et al.* [19] have shown that an age > 25 years is a risk factor for the prediction of GDM. Whereas, in a Malaysian study, women aged 35 or more were 2.5 times more likely to have GDM compared to those aged 25-34 years (26.3% vs. 9.7%) [20].

Women with a personal history of GDM face a higher risk of type 2 diabetes mellitus and metabolic syndrome later in life [15]. Women with GDM have a 7 times higher risk of developing type 2 diabetes after pregnancy. This relation could not be verified in our study since we did not follow-up on patients. Pre-pregnancy weight and BMI will be discussed in the next section.

Body composition and GMD

Relation between pre-pregnancy BMI and GDM

A meta-analysis published in 2019 showed that a high BMI before pregnancy is associated with a higher risk of developing GDM and that 24% of all gestational complications are attributed to maternal overweight and obesity [21]. This was not confirmed in the present study when comparing pre-pregnancy BMI between three groups but it was confirmed in terms of a positive correlation between pre-pregnancy BMI and blood glucose levels at T0 min, T60 min, and T120 min. However, we could not determine a threshold for BMI to be correlated to GDM as Farrar *et al.* [19] determined a BMI of 25 Kg/m² to be a predictive factor of GDM. Pre-pregnancy BMI was the most important modifiable risk factor for GDM [22]. The meta-analysis by Torloni *et al.* [23] showed that an increase in BMI of 1 Kg/m² increases the prevalence of GDM by 0.92% [23].

Relation between weight gain during pregnancy and GDM

Average weight gain of the pregnant women of our study at 26 weeks of gestation was 7.84 Kg, which corresponds to the same

margins of weight gain set by the American Institute of Medicine [24]. However, this may be imprecise as the pre-pregnancy weight was self-reported by some pregnant women who did not have a pregnancy follow-up record. The weight gain in our study was lower in pregnant women with GDM as that reported by other studies [25].

Although we found no correlations between weight gain and GDM, a study conducted in 2016 showed that a weight gain of more than 7 Kg in the second trimester increases the risk of GDM by 2.6 times in women with a negative first trimester GDM screening result [26]. Hedderson *et al.* [27] also showed that excessive weight gain during pregnancy, particularly in the first trimester, is associated with an increased risk of GDM. According to this study, early gestational weight gain can lead to an early increase in insulin resistance, which leads to the depletion of beta cells of the pancreas that become unable to secrete adequate levels of insulin [27]. This discordance could be explained by the high rate of obesity before pregnancy in our population.

Relation between FM, FFM and GDM

A recent review conducted by Najafi *et al.* [28] revealed that the risk of GDM was higher among overweight/obese pregnant women, which seems to be mostly related to FM. According to Wang *et al.* [29] BMI and body fat are associated with an increased risk of type 2 diabetes in obese Chinese women with GD [20]. This was confirmed in our study as FM was positively correlated to glucose levels in all pregnant women, whereas women with high FFM had lower glucose levels. This could be explained by the changes in maternal insulin sensitivity during pregnancy that effect maternal accretion of adipose tissue and energy expenditure [30].

Conclusion

The present study found that obesity is a risk factor for GDM and confirmed that there is a relationship between glycemic values and body composition parameters in all pregnant women. Moreover, during pregnancy, several clinical and biological parameters, for example, the lipid balance, also undergo profound gestational changes and deserve to be carefully studied to ensure better management of pregnant women and their newborns.

References

1. Baz B, Riveline JP, Gautier JF. ENDOCRINOLOGY OF PREGNANCY: Gestational diabetes mellitus: definition, aetiological and clinical aspects. *Eur J Endocrinol.* 2016;174:R43-51.
2. Buchanan TA, Xiang A, Kjos SL, Watanabe R. What is gestational diabetes? *Diabetes Care.* 2007;30 Suppl 2:S105-11.
3. Sandu C, Bica C, Salmen T, et al. Gestational diabetes - modern management and therapeutic approach (Review). *Exp Ther Med.* 2021;21:81.
4. Khajehi M, Assareh H. Temporal trend of diabetes in pregnant women and its association with birth outcomes, 2011 to 2017. *J Diabetes Complications.* 2020;34:107550.
5. Alfadhli EM. Gestational diabetes mellitus. *Saudi Med J.* 2015; 36:399-406.

6. Widen EM, Gallagher D. Body composition changes in pregnancy: measurement, predictors and outcomes. *Eur J Clin Nutr.* 2014; 68:643-52.
7. Most J, Marlatt KL, Altazan AD, Redman LM. Advances in assessing body composition during pregnancy. *Eur J Clin Nutr.* 2018;72:645-56.
8. Vambergue A. [Gestational diabetes: diagnosis, short and long term management]. *Presse Med.* 2013;42:893-9.
9. Vitner D, Harris K, Maxwell C, Farine D. Obesity in pregnancy: a comparison of four national guidelines. *J Matern Fetal Neonatal Med.* 2019;32:2580-90.
10. Boulrier A, Fricker J, Ferry M, Apfelbaum M. Body composition assessment by bioelectrical impedance analysis. *Nutr Clin Métabolisme.* 1991;5:165-74.
11. El Ati J, Traissac P, Delpeuch F, et al. Gender obesity inequities are huge but differ greatly according to environment and socio-economics in a North African setting: a national cross-sectional study in Tunisia. *PLoS One.* 2012;7:e48153.
12. Atek M, Traissac P, El Ati J, et al. Obesity and association with area of residence, gender and socio-economic factors in Algerian and Tunisian adults. *PLoS One.* 2013;8:e75640.
13. Gupta A, Faber P. Poids maternel et grossesse. *Prat En Anesth Réanimation.* 2012;16:218-23.
14. Catalano PM, Shankar K. Obesity and pregnancy: mechanisms of short term and long term adverse consequences for mother and child. *BMJ.* 2017;356:j1.
15. Vejrazkova D, Vankova M, Lukasova P, et al. Specific metabolic characteristics of women with former gestational diabetes: the importance of adipose tissue. *Physiol Res.* 2017;66:S349-S356.
16. Mbungu Mwimba R, Tandou-Umba N F B, Muls E. [A Longitudinal study of body composition and basal metabolic rate during pregnancy in a black population of Kinshasa, Democratic Republic of Congo (RDC)]. *J Gynecol Obstet Biol Reprod (Paris).* 2007;36:699-704.
17. Coustan DR. Gestational diabetes mellitus. *Clin Chem.* 2013;59: 1310-21.
18. Cho NH, Shaw JE, Karuranga S, et al. IDF Diabetes Atlas: Global estimates of diabetes prevalence for 2017 and projections for 2045. *Diabetes Res Clin Pract.* 2018;138:271-81.
19. Farrar D, Simmonds M, Bryant M, et al. Risk factor screening to identify women requiring oral glucose tolerance testing to diagnose gestational diabetes: a systematic review and meta-analysis and analysis of two pregnancy cohorts. *PLoS One.* 2017;12:e0175288.
20. Kalok A, Peraba P, Shah SA, et al. Screening for gestational diabetes in low-risk women: effect of maternal age. *Horm Mol Biol Clin Investig.* 2018;34(1).
21. Santos S, Voerman E, Amiano P, et al. Impact of maternal body mass index and gestational weight gain on pregnancy complications: an individual participant data meta-analysis of European, North American, and Australian cohorts. *BJOG.* 2019;126:984-95.
22. Vidanalage CJK, Senarath U, Silva KD, Lekamge U, Liyanage IJ. Effects of initial body mass index on development of gestational diabetes in a rural Sri Lankan population: a case-control study. *Diabetes Metab Syndr.* 2016;10:S110-3.
23. Torloni MR, Betrán AP, Horta BL, et al. Prepregnancy BMI and the risk of gestational diabetes: a systematic review of the literature with meta-analysis. *Obes Rev.* 2009;10:194-203.
24. Catalano P, deMouzon SH. Maternal obesity and metabolic risk to the offspring: why lifestyle interventions may have not achieved the desired outcomes. *Int J Obes (Lond).* 2015;39:6429.
25. Morisset AS, Tchernof A, Dubé MC, Veillette J, Weisnagel SJ, Robitaille J. Weight gain measures in women with gestational diabetes mellitus. *J Womens Health (Larchmt).* 2011;20:375-80.
26. Boriboonthirunsarn D. Second trimester weight gain > 7 kg increases the risk of gestational diabetes after normal first trimester screening. *J Obstet Gynaecol Res.* 2017;43:462-7.
27. Hedderson MM, Gunderson EP, Ferrara A. Gestational weight gain and risk of gestational diabetes mellitus. *Obstet Gynecol.* 2010;115:597-604.
28. Najafi F, Hasani J, Izadi N, et al. Risk of gestational diabetes mellitus by pre-pregnancy body mass index: A systematic review and meta-analysis. *Diabetes Metab Syndr.* 2021;15:102181.
29. Wang L, Liu H, Zhang S, et al. Obesity index and the risk of diabetes among Chinese women with prior gestational diabetes. *Diabet Med.* 2014;31:1368-77.
30. Catalano PM, Kirwan JP, Haugel-de Mouzon S, King J. Gestational diabetes and insulin resistance: role in short- and long-term implications for mother and fetus. *J Nutr.* 2003;133:1674S-1683S.