PLOS ONE

RESEARCH ARTICLE

Correlations between parameters of glycaemic variability and foetal growth, neonatal hypoglycaemia and hyperbilirubinemia in women with gestational diabetes

Immacolata Blasi[1], Jessica Daolio[1]*, Valeria Pugni[2], Giuseppina Comitini[1], Marcello Morciano[3,4], Giorgio Grassi[5], Tullia Todros[6], Giancarlo Gargano[7], Lorenzo Aguzzoli[1]

1 Department of Obstetrics and Gynecology, Azienda USL-IRCCS di Reggio Emilia, Reggio Emilia, Italy, 2 Endocrinology and Metabolism Unit, Azienda USL-IRCCS di Reggio Emilia, Reggio Emilia, Italy, 3 Health Organisation, Policy and Economics (HOPE) Research Group, University of Manchester, Manchester, United Kingdom, 4 Research Centre for the Analysis of Public Policies (CAPP), University of Modena and Reggio Emilia, Modena, Italy, 5 Department of Endocrinology, Diabetology and Metabolism, Azienda ospedaliera Città della Salute e della Scienza, Turin, Italy, 6 Department of Surgical Sciences, University of Turin, Turin, Italy, 7 Department of Neonatology, Azienda USL-IRCCS di Reggio Emilia, Reggio Emilia, Italy

* jessica.daolio@ausl.re.it

🔓 OPEN ACCESS

Abstract

The diagnosis of gestational diabetes mellitus (GDM) is important to prevent maternal and neonatal complications. This study aimed to investigate the feasibility of parameters of glycaemic variability to predict neonatal complications in women with GDM. A retrospective study was conducted on pregnant women tested positive at the oral glucose tolerance test (OGTT) during 16–18 or 24–28 weeks of gestation. Glycaemic measures were extracted from patients' glucometers and expanded to obtain parameters of glycaemic variability. Data on pregnancy outcomes were obtained from clinical folders. Descriptive group-level analysis was used to assess trends in glycaemic measures and foetal outcomes. Twelve patients were included and analysed, accounting for 111 weeks of observations. The analysis of trends in parameters of glycaemic variability showed spikes of glycaemic mean, high blood glucose index and J-index at 30–31 weeks of gestation for cases with foetal macrosomia, defined as foetal growth >90˚ percentile, neonatal hypoglycaemia and hyperbilirubinemia. Specific trends in parameters of glycaemic variability observed at third trimester correlate with foetal outcomes. Further research is awaited to provide evidence that monitoring of glycaemic variability trends could be more clinically informative and useful than standard glycaemic checks to manage women with GDM at delivery.

Introduction

Gestational diabetes mellitus (GDM) affects 3–7% of pregnancies [1]. The joint presence of insulin resistance and the subsequent increase in levels of postprandial glucose during

pregnancy allow us to characterize the pregnancy as "diabetogenic". In pregnancies affected by with GDM, the inability of pancreatic β-cells to compensate for insulin resistance leads to inadequate insulin activity and reduced insulin sensitivity, both at central and peripheral levels, which alter the physiological glycaemic balance.

GDM exposes pregnant women to higher risks of preeclampsia (PE), caesarean delivery, macrosomia, neonatal hypoglycaemia and hyperbilirubinemia, respiratory distress syndrome, and gestational type 2 diabetes mellitus in the following years [2]. In 2008, the Hyperglycaemia Adverse Pregnancy Outcome study (HAPO study) showed that increased levels of maternal glycaemia are associated with adverse neonatal outcomes [3]. Careful monitoring of glycaemic levels is therefore crucial to maintain glycaemia within the physiological range. As such, the glycaemic variability resulting from glycaemic fluctuations are considered a risk factor for the development of diabetes-related complications [4].

In women affected by GDM, foetal macrosomia is a recurrent complication associated with hyperglycaemia and high levels of glycaemic variability. Foetal macrosomia in women with GDM is associated with an increased risk of the following: (i) maternal complications, such as the increased possibility of caesarean delivery, operative vaginal delivery and perineal lacerations; (ii) short-term neonatal complications, such as still birth, shoulder dystocia, Erb's palsy, hypoxia and acidosis; and (iii) long-term neonatal complications, such as metabolic syndrome, type 2 diabetes mellitus, obesity and insulin resistance [5,6].

In 2007, Herranz et al. [7] compared the mean overall, pre- and postprandial glucose levels and the percentage of glucose readings above and below target and glycated haemoglobin (HbA1c) of 37 Large for Gestational Age (LGA) infants and 36 Appropriate for Gestational Age (AGA) infants from mothers with type 1 diabetes mellitus. The study found no statistically significant difference in the preconception glycaemic parameters between the two groups but showed a significant association between the parameters and LGA infants during the third trimester of pregnancy. As such, the authors concluded that glycaemic fluctuations are the best predictors of macrosomia [7]. On the one hand, high levels of basal glycaemia explain only a small percentage (12%) of glycaemic fluctuations and foetal macrosomia; on the other hand, high levels of postprandial glycaemia account for 40% of cases with elevated foetal weights [8]. Therefore, HbA1c levels used to assess the mean levels of glucose 4–6 weeks before delivery is not associated with foetal weight at birth [9]. In 2012, Mazze et al. [10] compared the glycaemic patterns from four groups of women experiencing physiological changes in pregnancy, pregnancy complicated by GDM, diagnosed with type 1 diabetes mellitus and nonpregnant women. The results showed that glycaemic patterns differed by 20% when pregnant and nonpregnant women were compared. As such, the authors concluded that further research is required to better define the role of glycaemic fluctuations, to improve the therapeutic approach and to reduce the incidence of maternal and foetal complications related to GDM.

The aim of this study is twofold. First, to establish the association, if any, between different measures of glycaemic variability in pregnancies affected by GDM. Second, it investigates the presence of differentiated trends in these measures for those which experienced birth weights above the 90th percentile, neonatal hyperglycaemia and/or hyperbilirubinemia, and those who did not.

Materials and methods

This was a retrospective observational study collecting data on pregnant women treated at the Department of Obstetrics and Gynaecology in collaboration with the Endocrinology and Metabolism Unit at the "Santa Maria Nuova" hospital, AUSL–IRCCS in Reggio Emilia, Italy.

The Ethics Committee AREA VASTA NORD of Azienda USL-IRCCS di Reggio Emilia, Italy, approved the study with approval n.2021/0003450. The methodology is in accord with

the Declaration of Helsinki. In accordance with study protocol approved, data were treated anonymously after collection.

Participants

In the present study, we included all pregnant women carrying a singleton pregnancy and receiving the diagnosis of GDM in the period between December 2015 and March 2016 at our hospital. The diagnosis of GDM was based on a 75-gram oral glucose tolerance test (OGTT) performed at 16–18 or 24–28 weeks of gestation [11]. We excluded twin pregnancies, women affected by pre-pregnancy type 1 or type 2 diabetes mellitus, and those with chronic hypertension or other pre-pregnancy diseases.

According to clinical practice, all the participants received a specific diet to follow and instructions on how to measure and report glycaemia data at home until delivery. Each patient was asked *(i)* to collect the glycaemic profile measuring glucose levels by fingertip blood tests at baseline, and one hour after each meal once a week, and at different times in the remaining days of the week, and *(ii)* to attend the Endocrinology and Metabolism Unit every two weeks for insulin therapy or four weeks for diet therapy. Insulin therapy was given because of a trend in glucose levels >95 mg/dL at baseline or >140 mg/dL one hour after meals, after at least a 2-week period of diet therapy.

Glycaemic data were collected in our database to calculate parameters of glycaemic variability as detailed below. Maternal characteristics and data on pregnancy outcome were obtained from clinical folders and were recorded in our database as well.

Obstetric and foetal outcomes collected

The following obstetric and foetal outcomes were recorded. With regard to obstetric outcomes, complications, such as PE and cholestasis, type of delivery, labor induction and delivery blood loss were considered. With regard to foetal outcomes, we recorded data on foetal weight and length, Apgar score and gestational age at birth. The eventual presence of neonatal hypoglycaemia and hyperbilirubinemia requiring phototherapy, distress, congenital malformations, shoulder dystocia, trauma during birth, stillbirth, and perinatal death were recorded as well. Of note, we used the most widely definition of neonatal hypoglycaemia to diagnose it, namely a glucose concentration of <40 mg/dl (2.6 mmol/l) in late preterm and term babies more than a few hours old [12–14], and the European Standards of Care for Newborn Health (EFCNI) to diagnose the condition of neonatal hyperbilirubinemia, namely it appears within 24 hours of birth following the detection of a bilirubin level >15 mg/dl (259 μmol/L), with an increase of >5 mg/die [15].

Given the absence of general agreement about the definition of macrosomia [6], for the purpose of this study, newborns weighted >4 kg were considered macrosomic. To identify LGA and SGA infants defined by the birth weight above the 90th percentile and below the 10th percentile, respectively, we referred to the Italian neonatal anthropometric values of reference [16].

Parameters of glycaemic variability collected

The parameters of glycaemic variability were calculated by EasyGV software [17]. The parameters considered were: the glycaemic mean (GM), defined as the arithmetic mean of all blood glucose values measured by women; the glycaemic mean value (GMV), defined as weighted average of the glycaemic means divided by the number of measurements and compared to an ideal average blood glucose value [18]; the mean amplitude of glucose excursions (MAGE), which quantifies the main glycaemic variations [19]; the classical standard deviation (SD); the

high blood glucose index (HBGI) and the low blood glucose index (LBGI), which indicate the frequency and the amplitudes of hyperglycaemic and hypoglycaemic events, respectively, and define the risks for patients to experience adverse glycaemic events [20]; the J-index, resulting from the means of glycaemic measures combined with the SDs of glycaemic values [21]; and the mean absolute glucose (MAG), as the sum of differences of consecutive glycaemic values divided by the total number of hours of observation [22]. All these parameters indicate data fluctuations, with higher values indicating higher variability.

Statistical analysis

First, Kendall's correlation analyses were used to examine the presence of correlations among the parameters of glycaemic variability. Second, we plotted parameter values over time with a linear interpolation function to graphically verify the presence of temporal trends. Finally, we graphically assessed the presence of differentiated trends for the parameters of glycaemic variability with foetal study outcome. For this, we defined two groups according to whether the foetal weight was above or below the 90th percentile, the presence and absence of neonatal hypoglycaemia and hyperbilirubinemia, as well. In Figs 1, 2 and 3, we displayed adjusted means for pre-pregnancy BMI and the administration or not of insulin therapy obtained after Anova regressions, with covariates set to the group-specific mean values. We also computed un-weighted group average (and 95% confidence intervals, CI) of GM and then performed a two-sample T-test on the equality of means (allowing for unequal variances) between those of women with adverse neonatal outcomes (LGA or Macrosomia) affected by obesity and those with physiological neonatal outcomes. All analyses were performed with the STATA software, version 17.
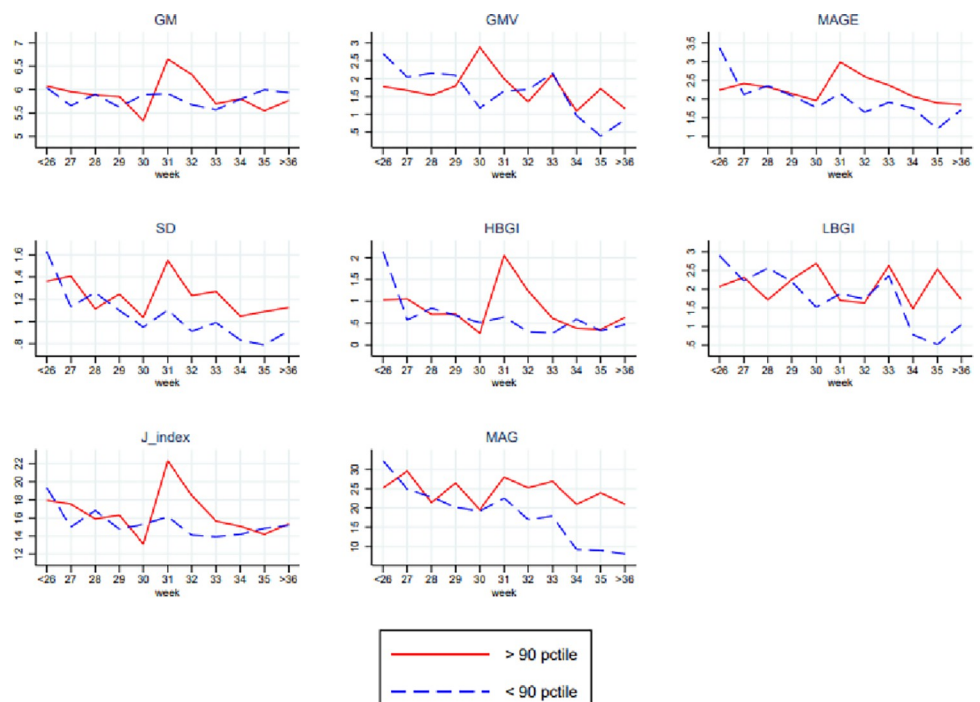

Fig 1. Trends in parameters of glycaemic variability by foetal growth. GM = Glycaemic Mean; GMV = Glycaemic Mean Value; HBGI = High Blood Glucose Index; LBGI = Low Blood Glucose Index; MAG = Mean Absolute Glucose; MAGE = Mean Amplitude of Glucose Excursions; SD = Standard Deviation. Adjusted values reported (see the Statistical analysis sub-section for details).

Fig 2. Trends in parameters of glycaemic variability by neonatal hypoglycaemia. GM = Glycaemic Mean;
GMV = Glycaemic Mean Value; HBGI = High Blood Glucose Index; LBGI = Low Blood Glucose Index; MAG = Mean
Absolute Glucose; MAGE = Mean Amplitude of Glucose Excursions; SD = Standard Deviation. Adjusted values
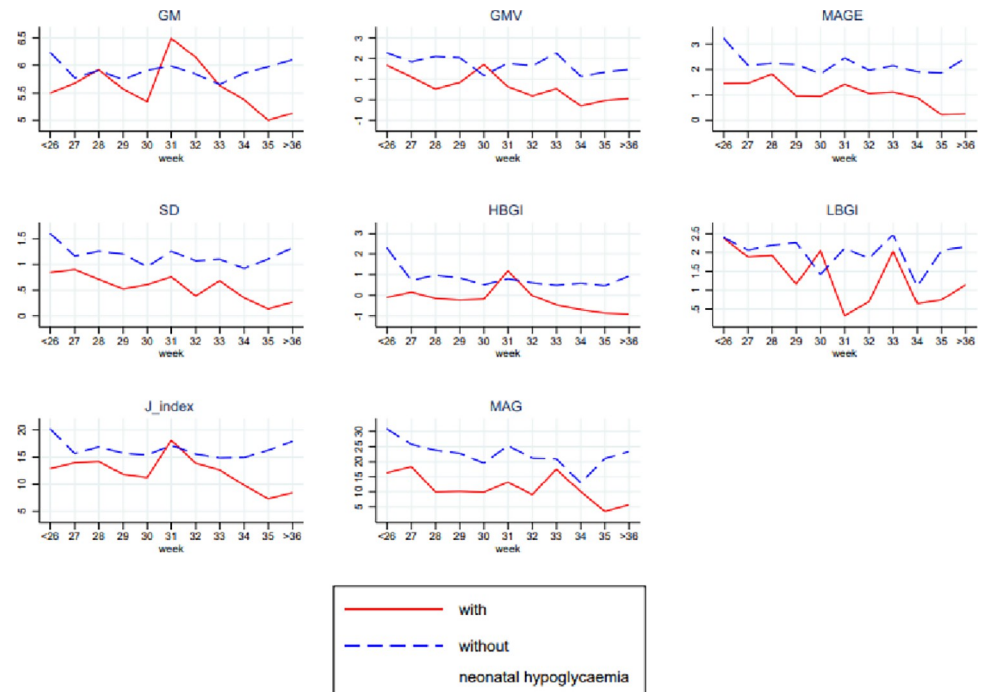reported (see the Statistical analysis sub-section for details).

https://doi.org/10.1371/journal.pone.0282895.g002

Results and discussion

In the study period, we recruited 12 pregnant women affected by GDM. The mean age of participants was 36 years old, and the mean BMI indicated overweight. Participants gained a physiological weight, in average. All women received the diagnosis of GDM at 24–28 weeks, except for one woman who received it at 16–18 weeks. All women underwent diet therapy, with three women requiring also insulin therapy. According to standard clinical practice, all women treated with diet- and/or insulin- therapy achieved acceptable blood glucose levels.

All new-borns had Apgar scores >7 at 5'. Macrosomia and LGA condition were identified in three babies, whose mothers presented a gestational weight gain greater than (case 2 and case 5) and less than (case 11) the Institute of Medicine (IOM) recommendations [23]. Neonatal hypoglycaemia was found in three out of twelve babies. Neonatal hyperbilirubinemia was found in four babies. Two out of three pre-term babies were admitted to neonatal intensive care unit (NICU). Specifically, one baby had respiratory distress, hypocalcaemia and sepsis. Detailed characteristics and pregnancy outcomes were reported in Table 1 for each participant.

Parameters of glycaemic variability

The statistical analysis was conducted on a sample consisting of 111 weeks of observations, accounting for a collection of glycaemic measures over a period of 9.25 weeks in average per woman. The mean values of glycaemic variability parameters are reported in S1 Table.

As shown in Table 2, the analysis of glycaemic variability revealed positive and statistically significant (p-value <0.01) correlations of GM with all other measures but not with GMV and
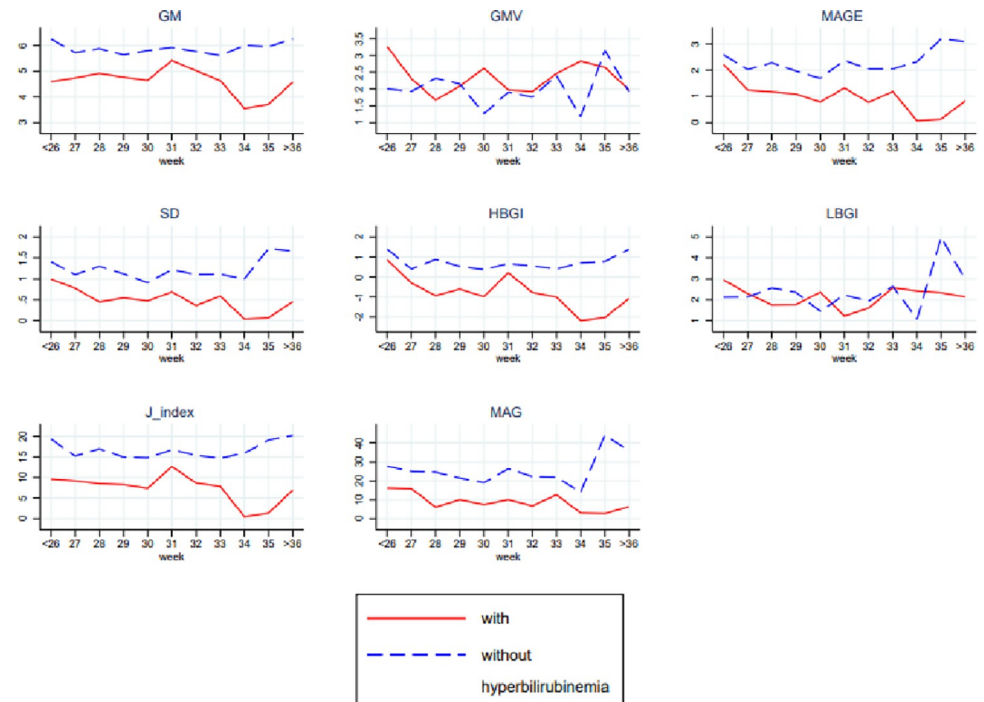
Fig 3. Trends in parameters of glycaemic variability by neonatal hyperbilirubinemia. GM = Glycaemic Mean; GMV = Glycaemic Mean Value; HBGI = High Blood Glucose Index; LBGI = Low Blood Glucose Index; MAG = Mean Absolute Glucose; MAGE = Mean Amplitude of Glucose Excursions; SD = Standard Deviation. Adjusted values reported (see the Statistical analysis sub-section for details).

LBGI (negative correlation, p-value <0.01). On the other hand, no significant correlations were found for GMV and HBGI, HBGI and LBGI, LBGI and J-index.

A significant decrease over time occurred mainly for the HBGI measure, in particular at the beginning of the observational window [S1 Fig]. The adjusted analysis in Figs 1, 2 and 3 revealed differentiated trends of glycaemic measures by whether the birth weight was above the 90th percentile, and with cases of neonatal hypoglycaemia and of hyperbilirubinemia. Specifically, for the three observed macrosomic cases (Fig 1), we found spikes in the GM, GMV, MAGE, SD, HBGI and J-index values around weeks 30–31. A no clear pattern was found for LBGI whereas the MAG values associated to macrosomic cases were generally higher than what observed for non-macrosomic cases.

For the three observed cases with neonatal hypoglycaemia (Fig 2), we found spikes in the GM, HBGI and J-index values also around weeks 30–31 of gestation. A no clear pattern was found for GMV whereas MAGE, SD and MAG values associated to hypoglycaemic cases were generally lower than what observed for euglycaemic cases. The LBGI values associated to hypoglycaemic cases showed an opposite trend, revealing a drop around weeks 30–31 of gestation. For the four observed cases with neonatal hyperbilirubinemia (Fig 3), we also found spikes in the GM, HBGI and J-index values around weeks 30–31 of gestation. A no clear pattern was found for GMV whereas MAGE, SD, MAG and LBGI values associated to cases with neonatal hyperbilirubinemia were generally lower than what observed for other cases. We also performed a test to assess whether the mean of GM differs significantly between the group of women with LGA/macrosomia fetuses (cases 2, 5 and 11) and those without (S1 Table). The corresponding two-tailed p-value was 0.2372, which is greater than 0.05. We concluded that the difference of means in GM between the group of women with macrosomia (mean 6.06,

Table 1. Maternal data and pregnancy outcomes of GDM participants.

	Gestational characteristics						Maternal characteristics at delivery							Foetal characteristics and outcomes					
	Age (years)	Parity	BMI (kg/m²)	GDM diagnosis (gestational week)	Diet therapy	Insulin therapy	BMI (kg/m²)	Weight gain (kg)	Gestational week	Labor induction	Type of delivery	Blood loss (ml)	Complications	Gender	Weight (kg)—Percentile	Length (cm)—Percentile	Apgar 1 minute	Apgar 5 minutes	Outcomes
Case 1	39	0	27.0	26	Yes	No	33.0	14	40	Yes	Vaginal	300	None	Male	3.715–75	52–80	9	10	Hyperbilirubinemia
Case 2	38	2002	37.9	26	Yes	No	42.4	14	39	No	Vaginal	250	None	Male	4.245–98	52–80	9	10	Macrosomia
Case 3	32	2101	28.8	26	Yes	No	33.3	10	35	Yes	Iterative C-section	300	Cholestasis; PE	Male	2.495–42	47–51	9	10	Pre-term birth, Hypoglycaemia, Hyperbilirubinemia, NICU admission
Case 4	40	2101	26.9	17	Yes	Yes, initiate at 25 weeks	26.2	6	38	No	C-section due to breech presentation	300	None	Female	2.940–26	48–25	9	10	/
Case 5	34	1001	29.0	26	Yes	No	33.1	12	39	No	Iterative C-section	400	None	Male	3.980–92	52–80	10	10	LGA
Case 6	32	3201	23.8	27	Yes	No	27.5	10	39	Yes	Vaginal	50	None	Female	3.370–53	51–75	10	10	/
Case 7	42	2101	20.1	27	Yes	No	24.9	14	34	No	Iterative C-section and breech presentation	800	PE; Abdominal bleeding after delivery treated requiring operative laparoscopy	Male	2.134–40	46–60	8	9	Pre-term birth, Respiratory distress, Hypocalcaemia, Sepsis, NICU admission
Case 8	34	0	31.0	27	Yes	No	33.0	6	40	Yes	C-section due to CTG abnormalities	200	None	Male	3.155–21	50–34	9	9	/
Case 9*	na	0	na	27	Yes	Yes, initiated at 35 weeks	na	na	36	No	Vaginal	50	None	Male	3.064–81	50–89	10	10	Pre-term birth
Case 10	41	1001	33.9	26	Yes	No	36.6	6	38	No	Vaginal	300	None	Male	3.350–55	51–73	9	10	Hypoglycaemia
Case 11	26	0	38.5	26	Yes	Yes, initiated at 26 weeks	40.0	4	37	Yes	Vaginal	450	Cholestasis	Male	3.715–96	50–71	9	10	LGA Hypoglycaemia, Hyperbilirubinemia
Case 12	38	1001	na	26	Yes	No	na	14	37	No	Vaginal (PROM)	100	None	Female	2.695–21	49–59	9	10	Hyperbilirubinemia

BMI = Body Mass Index; CTG = CardioTocoGraphic; GDM = Gestational Diabetes Mellitus; LGA = Large for Gestational Age; NA = not available; NICU = Neonatal Intensive Care Unit; PE = Pre-eclampsia; PROM = Premature Rupture of Membranes

* = Case 9 delivered in a different hospital.

https://doi.org/10.1371/journal.pone.0282895.t001

Table 2. Results of correlation analyses among parameters of glycaemic variability.

	GM	GMV	MAGE	SD	HBGI	LBGI	J-index	MAG
GM	1							
GMV	-0.4236*	1						
MAGE	0.2775*	0.2380*	1					
SD	0.2621*	0.3012*	0.7140*	1				
HBGI	0.5260*	-0.077	0.5571*	0.5520*	1			
LBGI	-0.2822*	0.7079*	0.2948*	0.3833*	-0.0362	1		
J-index	0.6896*	-0.1197	0.5443*	0.5725*	0.7106*	-0.0108	1	
MAG	0.1867*	0.3140*	0.5813*	0.7202*	0.3957*	0.4265*	0.4421*	1

GM = Glycaemic Mean; GMV = Glycaemic Mean Value; HBGI = High Blood Glucose Index; LBGI = Low Blood Glucose Index; MAG = Mean Absolute Glucose; MAGE = Mean Amplitude of Glucose Excursions; SD = Standard Deviation. Notes

* Statistically significant at p<0.01.

https://doi.org/10.1371/journal.pone.0282895.t002

95% CI: 5.50–6.63) and the group without (mean 5.84, 95% CI: 5.64; 6.05) was not statistically different from 0.

Discussion

In this study, we analysed parameters of glycaemic variability from a small cohort of patients adopting a pilot study-like approach. Our findings show that, in women with GDM, parameters of glycaemic variability reveal different trends at 30–31 weeks of gestation, according to the occurrence of foetal macrosomia, neonatal hypoglycaemia and hyperbilirubinemia. Specifically, we found that spikes of GM, HBGI and J-index values are similar among all the three conditions, and did not vary after adjustment for pre-pregnancy BMI and the administration or not of insulin therapy as known confounding factors for adverse neonatal outcomes. From a different perspective, we firstly provided a footprint of three foetal outcomes related to GDM at the beginning of third trimester, clinically challenging to manage women with GDM until delivery and newborns immediately after birth. One could hypothesize that the observed spikes could be related to low glycaemic control requiring insulin therapy rather than to a single parameter of estimated glycaemic variability. Due to the small sample size, we are not able to neither accept nor refuse such hypothesis, but future research is required to clarify this issue.

The risk of diabetes-related complications is illustrated as the diagonal arrow of a geometric cube whose three-dimensional coordinates on the three axes are basal, postprandial glycaemia, and glycaemic fluctuations. The therapeutic approach for diabetes should aim to reduce the values of such coordinates to reduce the diagonal arrow measure of the cube represented by diabetes-related complications [4]. With regard to GDM, it is associated with adverse pregnancy outcomes due to which proper glycaemic monitoring and treatments are widely acknowledged. In fact, the suboptimal glycaemic control could lead to a higher incidence of foetal macrosomia and other composite outcomes [24] in the same way by which GDM treatments limited to standard diet therapy [25] and routine care [26] determine when compared to GDM insulin treatment. In this context, the use of parameters of glycaemic variability to monitor GDM are still experimental and supported by limited literature. The relationship between glycaemic variability and GDM has been firstly reviewed in 2020 by Yu W. et al (2020) [27]. Authors found that glycaemic variability is significantly higher in women with GDM compared to pregnant women without GDM, but failed to find consistent conclusions

with regard to the relationship between glycaemic fluctuations in women with GDM and the occurrence of adverse neonatal events [27]. In light of this, results from this study are consistent with those from studies reporting that greater glycaemic fluctuations are more likely to cause adverse neonatal outcomes [28–30]. Of note, unlike these studies, we obtained our results via self-monitoring of blood glucose (SMBG) rather than CGM strategy.

The risk of foetal macrosomia in pregnant women with type 1 and type 2 diabetes mellitus ranges between 48% and 62%, and it is 2–3 times higher in women with GDM than in normal pregnant women [31]. Hence, it is important for women with GDM to maintain glycaemic levels within the normal range. Our body devotes much effort to maintaining glycaemia within the normal range, and vice versa, blood glucose could damage our body. Pregnancy in women with normal glucose metabolism is characterized by fasting levels of blood glucose that are lower compared to the non-pregnant status, due to insulin-independent glucose uptake by the fetus and placenta, and by mild postprandial hyperglycemia and carbohydrate intolerance as a result of diabetogenic placental hormones. "Flat blood glucose profiles" is however a condition characterized by glycemic excursions on average between 60 and 140mg /dl. "Flat blood glucose profiles" even in women with normal glucose tolerance it is characterized by a certain degree of glycemic variability favored by the excursion of postprandial glycaemia [32]. With this in mind, GDM monitoring has been ameliorated, but there is insufficient evidence to discriminate which parameters are able to predict foetal growth, as they are not properly informative about continuous glycaemic levels and variability. In addition, the relationship between glycaemic variability and foetal complications is poorly understood and investigated in women with GDM. In these women, small periods of transient hyperglycaemia seem enough to induce foetal growth acceleration responsible for macrosomia at birth. It is therefore of huge importance to monitor glycaemic variability in pregnant women affected by GDM. In 2011, Dalfrà et al. compared novel parameters of glycaemic variability, such as MAGE, glycaemic mean, SD, interquartile range (IQR), continuous overlapping net glycaemic action (CONGA), LGBI and HBGI, in two groups of pregnant women, the first affected by type 1 diabetes mellitus and the second with GDM, to healthy pregnant controls [33]. The study demonstrated that parameters of glycaemic variability reach higher levels in pregnant women with type 1 diabetes mellitus than in those with GDM and healthy controls. During the third trimester, parameters of glycaemic variability tend to decrease in women with type 1 diabetes mellitus and, on the contrary, increase in women with GDM, especially when insulin therapy is needed. Continuous glucose monitoring revealed alterations of the glycaemic profile in 61.3% of women with GDM for which insulin administration was required. The findings of the aforementioned study highlight that MAGE, GMV and SD reach higher levels in women with insulin-treated GDM than in women with diet therapy-treated GDM during the second trimester, and it is in this period that GMV and HBGI values predict and are correlated with asymmetric macrosomia. This measure was defined by the ponderal index value (PI). In women with type 1 diabetes mellitus, PI correlates with HBGI in the first trimester, CONGA and IQR in the second trimester, and GMV and SD in the third trimester. The CONGA parameter has been recently introduced to express glycaemic variability in a defined period, such as every day, throughout a continuous glucose analysis [34]. In none of the three groups or trimesters did glycated haemoglobin correlate with parameters of foetal growth. In accordance with the literature, we found that glycaemic variability shows clinically-relevant trends with foetal outcomes regarding foetal weight above >90˚ percentile, neonatal hypoglycaemia and hyperbilirubinemia.

The development of GDM during pregnancy may constitutes a transient condition for which, in our reality, CGM together with the application of subcutaneous abdominal probes are not applied conversely to what occurs in case of type 1 or type 2 diabetes. Our study

investigated glycaemic variability by SMBG rather than CGM strategies for 48–72 hours as reported in previous studies. Although this approach could be inaccurate due to the lack of CGM data, on the other side, it allows to speculate that, in case of GDM, parameters of glycaemic variability could switch from being experimental to clinically informative, as we have firstly provided initial evidence to suggest their ability to early detect, and therefore prevent, potential adverse foetal outcomes. Considering that parameters were obtained from women's glycaemic glucometers without the application of subcutaneous probes (of note, this approach is carried out in experimental conditions), it could be also speculated that some glycaemic variability parameters are better suited to a survey consisting of fewer observations during the day and allow the identification of pregnant women with GDM that are at higher risk of developing complications than others.

Interestingly, a semiparametric statistical approach was proposed by Gupta R et al. [35] to identify the rate of progression in maternal glucose concentrations in specific gestational periods of LGA and AGA babies of mothers with type 1 diabetes mellitus. The study showed that time-specific fluctuations in glucose level velocity and changes in glucose velocity differ across gestational age in the same woman and between women delivering LGA and AGA infants. As such, in the first trimester, mothers delivering LGA infants show higher accelerations of glucose levels than those delivering AGA infants, suggesting the risk of neonatal hypoglycaemia. In the third trimester, after a steady state of glucose concentrations for both groups, there is a sharp decline for LGA foetuses, which indicates a higher risk of neonatal hypoglycaemia compared to AGA foetuses [35].

In our opinion, the strength of our study involves the following issue. It provides the basis for the clinical contextualisation of experimental parameters of glycaemic variability, in terms of sensibility and specificity for women with GDM, and potentially for women affected by other types of diabetes. This is important in case of transient forms of diabetes, such as GDM. To this regard, the study by Bapajeva G et al., (2022), showed that pre-existing insulin-dependent GDM increased the risk for pregnancy complications compared to other GDM types, such as the insulin independent form [36]. Of note, we found of interest the role of myo-inositol and D-chiro-inositol for the prevention and treatment of metabolic disorders, such as GDM [37,38], but we await future studies to fully examine the beneficial effects. Furthermore, the prevention of adverse neonatal events is fundamental to avoid short- and long-term complications on newborns' development, and requires many clinical efforts, ranging from early detection of trigger factors, such as GDM, to administration of care factors, such as diet and/or insulin therapy. Insights from this study show that the strategy based on parameters of glycaemic variability may be useful to tailor glycaemic control and care in pregnant women until delivery and, therefore, prevent adverse neonatal outcomes associated with GDM. Moreover, glycaemic variability is considered a new concept of glycaemic control, and it is higher in women with GDM compared to women without GDM [27]. As the concept of hyperglycaemia is profoundly changed so far [39], this study helps to broaden the landscape of glycaemic management, placing a special emphasis on women with GDM, for which there are limited data.

The limits of the study involve the small, but well-characterized sample size and the presence of pre-gestational overweight/obesity among women included in the study. Although obtained with a small simple size, our results should be considered preliminary and interpreted through the lens of a pilot study conducted with an exploratory intend, that aimed at contributing to the limited scientific knowledge on glycaemic variability in pregnant women. Increased BMI, and advanced age as well, prior to pregnancy are risk factors for pregnancy complications, with maternal obesity being the most important predictor of pregnancy complications in women with GDM [40].

Obesity constitutes an independent factor associated with foetal macrosomia, in light of which we believe it could make sense to further investigate our findings in different subjects. Pregnancy outcome can be good for both mothers with GDM and children with a timely and adequate approach [41]. With regard to the relationship between macrosomia or LGA, and maternal obesity, we examined maternal birth weight in the three cases of LGA or Macrosomia in the light of IOM recommendations. Case 2 (obese patient before pregnancy) and case 5 (overweight patient before pregnancy) presented a gestational weight gain greater than the IOM recommendations, whereas case 11 (obese patient before pregnancy) manifested a gestational weight gain less than the IOM recommendations. Gestational weight gains greater than or less than guideline recommendations, compared with weight gains within recommended levels, is associated with higher risk of adverse maternal and infant outcomes [41]. Based on limited sample size, further conclusions on the relationship between macrosomia or LGA, and maternal obesity cannot be drawn. However, we carefully speculate on how GDM contributes to predict macrosomia or LGA through a specific trend of glycaemic variability.

Another limitation of this study involves the lack of data from CGM. In fact, the EasyGV software used in this study typically calculates parameters of glycaemic variability from CGM data, rather than glucometers. Unfortunately, CGM data are not available for patients included in this study for reason independent of authors' intention. In fact, in our clinical practice the development of GDM does not constitute an indication *per se* requiring CGM. As consequence, validation of results with different methods are warranted in order to expand the role of glycaemic variability as a tool in patients with transitional diabetic conditions, such as pregnant women, and to explore the most optimal method to measure glycaemic variability. Although found of interest in other related studies (see e.g. [42–47]), we also did not measure HOMA-IR, insulin levels and "time in range" in our cohorts of pregnant women with GDM.

Conclusions

Our study shows the presence of differentiated trends of glycaemic measures when the birth weight was above the 90th percentile, and with cases of hypoglycaemia and of hyperbilirubinemia. We found spikes in the values of some parameters (particularly GM, HBGI and J-index) with adverse foetal outcomes particularly around weeks 30–31 of gestation. We are confident that any future insight will contribute to improve GDM management and treatment, as the relationship between glycaemic variability and gestational complications constitutes a novel and intriguing field of research.

Supporting information

S1 Fig. Unadjusted trends in parameters of glycaemic measures over time, as spotted (grey bullets) and fitted values (segmented lines). GM = Glycaemic Mean; GMV = Glycaemic Mean Value; HBGI = High Blood Glucose Index; LBGI = Low Blood Glucose Index; MAG = Mean Absolute Glucose; MAGE = Mean Amplitude of Glucose Excursions; SD = Standard Deviation.
(PDF)

S1 Table. Mean values of parameters of glycaemic variability. * = Cases with obese pre-pregnancy BMI delivering LGA/macrosomic fetuses, whose mean of GM was compared with that of other cases to assess an eventual difference of statistical significance between the group of cases.
(PDF)

S2 Table. STROBE checklist.
(PDF)

Author Contributions

Conceptualization: Immacolata Blasi.

Data curation: Immacolata Blasi, Valeria Pugni.

Formal analysis: Marcello Morciano.

Methodology: Valeria Pugni, Marcello Morciano.

Supervision: Giuseppina Comitini, Marcello Morciano, Giorgio Grassi, Tullia Todros, Giancarlo Gargano, Lorenzo Aguzzoli.

Writing – original draft: Jessica Daolio.

Writing – review & editing: Immacolata Blasi, Jessica Daolio, Marcello Morciano, Giorgio Grassi.

References

1. Engelgau MM, Herman WH, Smith PJ, German RR, Aubert RE. The epidemiology of diabetes and pregnancy in the U.S., 1988. Diabetes Care. 1995; 18:1029–33. https://doi.org/10.2337/diacare.18.7.1029 PMID: 7555537

2. Kjos SL, Buchanan TA. Gestational diabetes mellitus. N Engl J Med. 1999; 341:1749–56. https://doi.org/10.1056/NEJM199912023412307 PMID: 10580075

3. HAPO Study Cooperative Research Group, Metzger BE, Lowe LP, Dyer AR, Trimble ER, Chaovarindr U, et al. Hyperglycaemia and adverse pregnancy outcome. N Engl J Med. 2008; 358: 1991–2002. https://doi.org/10.1056/NEJMoa0707943

4. Monnier L, Colette C. Glycemic variability. Should we and can we prevent it? Diabetes Care 2008; 31: S150–4. https://doi.org/10.2337/dc08-s241 PMID: 18227477

5. Kerenyi Z, Tamas G, Kivimaki M, Peterfalvi A, Madarasz E, Bosnyak Z, et al. Maternal glycemia and risk of large-for-gestational age babies in a population based screening. Diabetes Care. 2009; 32:2200–5. https://doi.org/10.2337/dc09-1088 PMID: 19729526

6. Henriksen T. The macrosomic fetus: a challenge in current obstetrics. Acta Obstet Gynecol Scand. 2008; 87:134–45. https://doi.org/10.1080/00016340801899289 PMID: 18231880

7. Herranz L, Pallardo LF, Hillman N, Martin-Vaquero P, Villarroel A, Fernandez A. Maternal third trimester hyperglycaemic excursions predict large for gestational age infants in type 1 diabetic pregnancy. Diabetes Res Clin Pract. 2007; 75:42–6. https://doi.org/10.1016/j.diabres.2006.05.019 PMID: 16837097

8. Person B, Hanson UF. Size at birth in relation to quality of blood glucose control in pregnancies complicated by pregestational diabetes mellitus. Br J Obstet Gynecol. 1996; 10:427–33. https://doi.org/10.1111/j.1471-0528.1996.tb09768.x

9. Kerssen A, de Valk HW, Visser GHA. Increased second trimester maternal glucose levels are related to extremely large-for-gestational age infants in women with type 1 diabetes. Diabetes Care. 2007; 30:1069–74. https://doi.org/10.2337/dc05-1985 PMID: 17468376

10. Mazze R, Yogev Y, Langer O. Measuring glucose exposure and variability using continuous glucose monitoring in normal and abnormal glucose metabolism in pregnancy. J Matern Fetal and Neonat Med. 2012; 25:1171–5. https://doi.org/10.3109/14767058.2012.670413 PMID: 22384865

11. Alberti KG, Zimmet PZ. Definition, diagnosis and classification of diabetes mellitus and its complications. Part 1: diagnosis and classification of diabetes mellitus provisional report of a WHO consultation. Diabet Med. 1998; 15:539–53. https://doi.org/10.1002/(SICI)1096-9136(199807)15:7<539::AID-DIA668>3.0.CO;2-S PMID: 9686693

12. Clinical Practice Committee. Guidelines for the Management of Hypoglycaemia. (2018). Available online at: http://www.adhb.govt.nz/newborn/Guidelines/Nutrition/HypoglycaemiaManagement.htm [accessed 10 April 2020].

13. Dixon KC, Ferris RL, Marikar D, Chong M, Mittal A, Manikam L, et al. Definition and monitoring of neonatal hypoglycaemia: a nationwide survey of NHS England Neonatal Units. Arch Dis Child Fetal Neonatal Ed. 2017; 102:F92–3. https://doi.org/10.1136/archdischild-2016-311473 PMID: 27758930

14. Harris D, Weston PJ, Battin MR, Harding JE. A survey of the management of neonatal hypoglycaemia within the Australian and New Zealand Neonatal Network. J Paediatr Child Health. 2014; 50:E55–62. https://doi.org/10.1111/j.1440-1754.2009.01599.x PMID: 19863712

15. European Standards of Care for Newborn Health: Neonatal jaundice. (2018). Available online at: https://www.efcni.org/wp-content/uploads/2018/11/2018_11_16_ESCNH_Report_final.pdf. [accessed 28 June 2022].

16. Bertino E, Spada E, Occhi L, Coscia A, Guliani F, Gagliardi L, et al. Neonatal Antropometric Charts: The Italian neonatal study compared with other European study. JPGN. 2010; 51:353–61. https://doi.org/10.1097/MPG.0b013e3181da213e

17. Hill NR, Oliver NS, Choudhary P, Levy JC, Hindmarsh P, Matthews DR. Normal reference range for mean tissue glucose and glycemic variability derived from continuous glucose monitoring for subjects without diabetes in different ethnic groups. Diabetes Technol Ther. 2011; 13:921–8. https://doi.org/10.1089/dia.2010.0247 PMID: 21714681

18. Schlichtkrull J, Munch O, Jersild M. The M-value, an index of blood sugar control in diabetics. Acta Med Scand. 1965; 177:95–102. https://doi.org/10.1111/j.0954-6820.1965.tb01810.x

19. Service FJ O'Brien PC, Rizza RA. Measurement of glucose control. Diabetes Care. 1987; 10:225–37. https://doi.org/10.2337/diacare.10.2.225

20. Kovatchev BP, Clarke WL, Breton M, Brayman K, McCall A. Quantifying temporal glucose variability in diabetes via continuous glucose monitoring: mathematical methods and clinical application. Diabetes Technol Ther. 2005; 7: 849–62. https://doi.org/10.1089/dia.2005.7.849 PMID: 16386091

21. Wojcick JM. "J"-index. A new proposition of the assessment of current glucose control in diabetic patient. Horm Metab Res. 1995; 27:41–2. https://doi.org/10.1055/s-2007-979906

22. Hermanides J, Vriesendorp TM, Bosman RJ, Zandstra DF, Hoekstra JB, Devries JH. Glucose variability is associated with intensive care unit mortality. Crit Care Med. 2010; 38: 838–42. https://doi.org/10.1097/CCM.0b013e3181cc4be9 PMID: 20035218

23. Physical status: the use and interpretation of anthropometry. Report of a WHO Expert Committee. World Health Organ Tech Rep Ser. 1995; 854:1–452. PMID: 8594834.

24. Gonzalez-Quintero VH, Istwan NB, Rhea DJ, Rodriguez LI, Cotter A, Carter J, et al. The impact of glycemic control on neonatal outcome in singleton pregnancies complicated by gestational diabetes. Diabetes Care. 2007; 30:467–70. https://doi.org/10.2337/dc06-1875 PMID: 17327306

25. Langer O, Yogev Y, Most O, Xenakis EM. Gestational diabetes: the consequences of not treating. Am J Obstet Gynecol. 192;2005:989–97. https://doi.org/10.1016/j.ajog.2004.11.039 PMID: 15846171

26. Crowther C, Hiller J, Moss J, McPhee A, Jeffries W, Robinson J. Effect of treatment of gestational diabetes mellitus on pregnancy outcomes. N Engl J Med. 2005; 352:2477–86. https://doi.org/10.1056/NEJMoa042973 PMID: 15951574

27. Yu W, Wu N, Li L, OuYang H, Qian M, Shen H. A Review of Research Progress on Glycemic Variability and Gestational Diabetes. Diabetes Metab Syndr Obes. 2020; 13:2729–41. https://doi.org/10.2147/DMSO.S261486 PMID: 32801819

28. Yu F, Lv L, Liang Z, Wang Y, Wen J, Lin X, et al. Continuous glucose monitoring effects on maternal glycemic control and pregnancy outcomes in patients with gestational diabetes mellitus: a prospective cohort study. J Clin Endocrinol Metab. 2014; 99:4674–82. https://doi.org/10.1210/jc.2013-4332 PMID: 25057872

29. Dalfrà MG, Chilelli NC, Di Cianni G, Mello G, Lencioni C, Biagioni S, et al. Glucose Fluctuations during Gestation: An Additional Tool for Monitoring Pregnancy Complicated by Diabetes. Int J Endocrinol. 2013; 2013:279021. https://doi.org/10.1155/2013/279021 PMID: 24319455

30. HAPO Study Cooperative Research Group. Hyperglycemia and Adverse Pregnancy Outcome (HAPO) Study: associations with neonatal anthropometrics. Diabetes. 2009; 58:453–9. https://doi.org/10.2337/db08-1112 PMID: 19011170

31. Lapolla A, Dalfrà MG, Di Cianni G, Bonomo M, Parretti E, Mello G. Scientific Committee of the GISOGD group: a multicentre Italian study on pregnancy outcome in women with diabetes. Nutr Metab Cardiovasc Dis. 2008; 18:291–7. https://doi.org/10.1016/j.numecd.2006.12.001

32. Mazze RS, Strock E, Wesley D, Borgman S, Morgan B, Bergenstal R, et al. Characterizing glucose exposure for individuals with normal glucose tolerance using continuous glucose monitoring and ambulatory glucose profile analysis. Diabetes Technol Ther. 2008; 10: 149–59. https://doi.org/10.1089/dia.2007.0293 PMID: 18473688

33. Dalfrà MG, Sartore G, Di Cianni G, Mello G, Lencioni C, Ottanelli S, et al. Glucose variability in Diabetic pregnancy. Diabetes Technol Ther. 2011; 8: 853–9. https://doi.org/10.1089/dia.2010.0145 PMID: 21751862

34. McDonnel CM, Donath SM, Vidmar SI, Werther GA, Cameron FJ. A novel approach to continuous glucose analysis utilizing glycemic variation. Diabetes Technol Ther. 2005; 7:253–63. https://doi.org/10.1089/dia.2005.7.253 PMID: 15857227

35. Gupta R, Khoury J, Altaye M, Dolan L, Szczesniak RD. Glycemic excursions in type 1 diabetes in pregnancy: a semiparametric statistical approach to identify sensitive time points during gestation. J Diabetes Res. 2017; 2017:2852913. https://doi.org/10.1155/2017/2852913 PMID: 28280744

36. Bapayeva G, Terzic S, Dotlic J, Togyzbayeva K, Bugibaeva U, Mustafinova M, et al. Pregnancy outcomes in women with diabetes mellitus—the impact of diabetes type and treat-ment. Prz Menopauzalny. 2022; 21:37–46. https://doi.org/10.5114/pm.2022.113781 PMID: 35388283

37. Dinicola S, Unfer V, Facchinetti F, Soulage CO, Greene ND, Bizzarri M, et al. Inositols: From Established Knowledge to Novel Approaches. Int J Mol Sci. 2021; 22:10575. https://doi.org/10.3390/ijms221910575 PMID: 34638926

38. Facchinetti F, Appetecchia M, Aragona C, Bevilacqua A, Bezerra Espinola MS, Bizzarri M, et al. Experts' opinion on inositols in treating polycystic ovary syndrome and non-insulin dependent diabetes mellitus: a further help for human reproduction and beyond. Expert Opin Drug Metab Toxicol. 2020; 16:255–74. https://doi.org/10.1080/17425255.2020.1737675 PMID: 32129111

39. Ceriello A, Prattichizzo F, Phillip M, Hirsch IB, Mathieu C, Battelino T. Glycaemic management in diabetes: old and new approaches. Lancet Diabetes Endocrinol. 2022; 10:75–84. https://doi.org/10.1016/S2213-8587(21)00245-X PMID: 34793722

40. Bapayeva G, Terzic S, Dotlic J, Togyzbayeva K, Bugibaeva U, Mustafinova M, et al. The influence of advanced age and obesity on pregnancy course and outcome in patients with diabetes mellitus. Prz Menopauzalny. 2022; 21:170–9. https://doi.org/10.5114/pm.2022.116351 PMID: 36254135

41. Goldstein RF, Abell SK, Ranasinha S, Misso M, Boyle JA, Black MH, et al. Association of gestational weight gain with maternal and infant outcomes: a systematic review and meta-analysis. JAMA. 2017; 317:2207–25. https://doi.org/10.1001/jama.2017.3635 PMID: 28586887

42. Feig DS, Donovan LE, Corcoy R, Murphy KE, Amiel SA, Hunt KF, et al. Continuous glucose monitoring in pregnant women with type 1 diabetes (CONCEPTT): a multicentre international randomised controlled trial. Lancet. 2017; 390:2347–2359. https://doi.org/10.1016/S0140-6736(17)32400-5 PMID: 28923465

43. Kristensen K, Ögge LE, Sengpiel V, Kjölhede K, Dotevall A, Elfvin A, et al. Continuous glucose monitoring in pregnant women with type 1 diabetes: an observational cohort study of 186 pregnancies. Diabetologia. 2019; 62:1143–1153. H https://doi.org/10.1007/s00125-019-4850-0 PMID: 30904938

44. Battelino T, Danne T, Bergenstal RM, Amiel SA, Beck R, Biester T, et al. Clinical Targets for Continuous Glucose Monitoring Data Interpretation: Recommendations From the International Consensus on Time in Range. Diabetes Care. 2019; 42:1593–1603. https://doi.org/10.2337/dci19-0028 PMID: 31177185

45. Advani A. Positioning time in range in diabetes management. Diabetologia. 2020; 63:242–252. https://doi.org/10.1007/s00125-019-05027-0 PMID: 31701199

46. Zhang W, Tang Y, Huang J, Yang Y, Yang Q, Hu H. Efficacy of inulin supplementation in improving insulin control, HbA1c and HOMA-IR in patients with type 2 diabetes: a systematic review and meta-analysis of randomized controlled trials. J Clin Biochem Nutr. 2020; 66:176–183. https://doi.org/10.3164/jcbn.19-103 PMID: 32523243

47. González-González JG, Violante-Cumpa JR, Zambrano-Lucio M, Burciaga-Jimenez E, Castillo-Morales PL, Garcia-Campa M, et al. HOMA-IR as a predictor of Health Outcomes in Patients with Metabolic Risk Factors: A Systematic Review and Meta-analysis. High Blood Press Cardiovasc Prev. 2022; 1. https://doi.org/10.1007/s40292-022-00542-5