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Does treatment modality affect measures of arterial stiffness in women with gestational diabetes?

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CONTRIBUTION

What are the novel findings of this work?

Brachial and aortic augmentation index (Aix) are increased amongst women with GDM from 24+0 to 35+6 weeks of gestation compared to healthy controls, regardless of treatment modality. We observed a decrease in mean AIx in all GDM treatment groups after initiation of treatment and an altered longitudinal pattern of AIx in the groups treated with metformin.

What are the clinical implications of this work?

Maladaptation of the cardiovascular system may underlie the association between GDM and gestational hypertensive disorders. Our findings also suggest that treatment with metformin could possibly attenuate the increase in augmentation index, although this finding requires further investigation in larger studies.

ABSTRACT

Objectives: The incidence of gestational diabetes mellitus (GDM) is increasing and is associated with adverse maternal, fetal and neonatal outcomes. Arterial stiffness (AS) is raised in pregnancies complicated by placental-mediated diseases such as pre-eclampsia. We investigated if AS is different between healthy pregnancies and women with GDM on different treatment modalities**.**

Methods: We conducted a prospective longitudinal cohort study to assess and compare AS in pregnancies complicated by GDM with low-risk controls. AS, measured by pulse wave velocity (PWV) and brachial (BrAIx) and aortic (AoAIx) augmentation Index, was recorded using the Arteriograph® at four gestational windows: 24+0 to 27+6; 28+0 to 31+6; 32+0 to 35+6 and ≥36+0 weeks of gestation (windows W1-W4, respectively). Women with GDM were considered both as a single group, and as subgroups defined by treatment modality. Data were analyzed using a linear mixed model on each AS variable (log-transformed) with group, gestational windows, maternal age, ethnicity, parity, body mass index, mean arterial pressure and heart rate as fixed effects and individual as a random effect. We compared the group means including relevant contrasts and adjusted the p-values using the Bonferroni correction.

Results: The study population comprised 155 low-risk controls and 127 with GDM, of whom 59 were treated with dietary intervention, 47 with metformin alone and 21 with metformin plus insulin. The two-way interaction term of study group and gestational age was significant for BrAIx and AoAIx ($p<0.001$), though there was no evidence ($p=0.729$) that mean AoPWV was different between the study groups. Women in the control group demonstrated significantly lower BrAIx and AoAIX at gestational windows W1-3 compared to the combined GDM group, but not at W4. Mean (95% CI) difference in log adjusted BrAIx was -0.37 (-0.52,

0.22), -0.23 (-0.35, -0.12), and -0.29 (-0.40, -0.18) at W1, W2 and W3, respectively. Mean (95% CI) difference in log adjusted AoAIx was -0.49 (-0.69, -0.3), -0.32 (-0.47, -0.18) and -0.38 (-0.52, -0.24) at W1, W2 and W3, respectively. Similarly, women in the control group also demonstrated significantly lower BrAIx and AoAIx compared with each of the GDM treatment subgroups (diet, metformin and metformin plus insulin) at W1-3. The increase in mean BrAIx and AoAIx seen between W2 and W3 in the women with GDM treated with dietary management was attenuated in the metformin and metformin with insulin groups, however the mean differences in BrAIx and AoAIx between these treatment groups were not statistically significant at any gestational window.

Conclusions: Pregnancies complicated by GDM demonstrate significantly higher AS compared to low-risk pregnancies regardless of treatment modality. Our data provides a basis for further investigation into the association of metformin therapy with changes in AS and risk of placental-mediated diseases.

INTRODUCTION

The incidence of gestational diabetes mellitus (GDM) is rising, driven largely by an increase in maternal obesity.[1] GDM is associated with a variety of adverse maternal, fetal and neonatal outcomes,[2] including an increased risk of placental-mediated diseases such as maternal hypertensive disorders of pregnancy,[3] and development of cardiovascular disease in the future life.[4]

Only a handful of studies have investigated changes in central hemodynamics in pregnancies complicated by GDM and have produced conflicting results. Three studies[5-7] have all reported no difference in pulse wave velocity (PWV) between women with GDM and low risk controls in the third trimester, whereas Mansukhani *et al.*[8] found PWV to be 3.7% higher amongst women with GDM at 35-37 weeks gestation. Similarly, three studies found no difference in third trimester AIx between women with GDM and healthy controls[6, 8, 9], whereas Savvidou *et al.*[7] and Kintiraki *et al.*[10] both reported increased AIx among women with GDM [13.1 ± 8.9% *vs* 0.7 ± 11.4% ($p<0.001$) and 3.77 ± 2.22 *vs* 1.51 ± 3.35% ($p = 0.021$), respectively]. A further limitation of these studies is that all have evaluated AIx at a single time point only, rather than throughout the third trimester. With the exception of Mansukhani *et al*.,[8] the studies involved fewer than 50 women in each group, and have presented the pregnant women with GDM as a single group without considering the potential impact of different treatment modalities on maternal cardiovascular adaptation.

Metformin is a glyburide recommended for treatment of hyperglycemia in GDM that does not respond to dietary and lifestyle changes.[11] Its use is associated with a reduction in the risk of gestational hypertension amongst pregnant women with GDM and polycystic ovarian syndrome (PCOS), [12-14] and a reduction in the incidence of pre-eclampsia in obese pregnant

women.[14] In non-pregnant populations, metformin has been shown to improve markers of arterial stiffness (AS) in populations with non-alcoholic fatty liver disease[15] and PCOS[16-18] though the effect of metformin on AS in pregnant women has not yet been fully established. In a pilot study, comparing longitudinal changes in AS in pregnancies complicated by GDM,[19] brachial and aortic AIx were significantly higher amongst women with GDM managed with dietary intervention at 32-34 weeks of gestation, and amongst those managed with metformin at 26-28 weeks of gestation compared to women with low-risk pregnancies, but there was no difference at other gestations. Although this study was limited by the small number of women included, it raised the possibility that different treatment modalities for GDM may have a differential effect on AS.

The aim, therefore, of our study was to examine longitudinal changes in AS among women with GDM and to investigate if any changes were affected by treatment modality.

METHODS

This was a prospective longitudinal cohort study of central hemodynamics in women with GDM, compared to low-risk controls. Pregnant women were recruited from the antenatal and ultrasound clinics at the Leicester Royal Infirmary, a tertiary-level maternity unit in the United Kingdom. Ethical approval was obtained from the East Midlands Research Ethics Committee (15/EM/0469, IRAS 182250) and the University Hospitals of Leicester (UHL) NHS Trust Research and Innovation Department prior to commencement. The study was conducted in accordance with the principles of Good Clinical Practice, and the Declaration of Helsinki.[20] All women provided written consent to take part.

Study population

Women over 16 years of age, with a viable singleton pregnancy, were eligible for inclusion in the study. We excluded women with pre-existing diabetes and cardiovascular disease, and with pregnancies complicated by aneuploidy or fetal abnormality. Since funding for translation services was not available, we also excluded women who did not speak or read English. In addition, women in the low-risk control group were excluded if they developed any hypertensive disorder of pregnancy, delivered prior to 37 completed weeks of gestation, or delivered a neonate with birth weight <10th centile according to population-based growth charts.[21]

GDM was defined as fasting glucose of ≥5.6 mmol/L, and/or a serum glucose level of ≥7.8mmol/L 2 hours after a 75g oral glucose load. The screening protocol for GDM used was adapted from UK national guidance[11] (Table S1). Women with GDM were managed by a multi-disciplinary team of midwives, obstetricians, and endocrinologists at UHL. Adequate glucose control was defined as a fasting glucose of <5.3mmol/L and 1 hour post prandial glucose levels

of <7.8mmol/L[11]. In accordance with UK national guidance[11], women who did not achieve and subsequently maintain these levels within 2 weeks of commencement of dietary interventions were commenced on metformin therapy by the managing physicians, starting at 500mg and increasing up to a maximum of 2000mg daily. Supplemental insulin therapy was commenced if adequate glycemic control was not achieved within 2 weeks of commencing metformin.

Power calculation, based on the data available from Osman *et al*.,[19] identified that for >80% statistical power, a sample size of 100 was required to detect ≥35% difference in AoAIx and ≥7% difference in PWV between women with GDM and healthy controls.

Study measurements

All women attended a minimum of two study visits during the third trimester. We collected demographic details including maternal age, ethnicity, height, parity, body mass index (BMI) and smoking status at booking. Gestational age at each visit was calculated based on the dating scan performed between 11^{+0} and 13^{+6} weeks gestation. For women in the GDM group, treatment modality (diet, metformin or metformin and insulin) and gestation at commencement of treatment were also recorded. Women who changed treatment modality after recruitment to the study were excluded, so that only women who remained in the same treatment group at each study visit were included in the analysis.

Maternal hemodynamics were measured at 4 gestational windows: 24+0 to 27+6 weeks gestation (W1), 28+0 to 31+6 weeks (W2), 32+0 to 35+6 weeks (W3) and ≥36+0 weeks (W4). Assessments were performed in a temperature-controlled room, free from noise or any other distractions. Patients were positioned in the semi-recumbent position and were asked not to move or talk during the assessment. All measurements were performed by a researcher who

had received appropriate training. The assessments were performed at scheduled appointments between 0900 and 1700. Our group has previously shown that measurements of PWV and AIx are not significantly affected by the time of day at which they are measured.[22]

Brachial (BrAIx) and aortic AIx (AoAIx), and PWV were measured using the Arteriograph® (TensioMed Ltd, Budapest, Hungary), which estimates AS oscillometrically, using a single, non-invasive blood pressure cuff. The Arteriograph® has been validated against invasive assessment of AS in a non-pregnant population undergoing cardiac angiography,[23] and shown to have good to excellent repeatability amongst healthy pregnant subjects in the third trimester.[22] Recruits had a minimum of two Arteriograph® readings taken at each visit. Measurements with a standard deviation of ≥1.0 m/s were excluded, as recommended by the Arteriograph® user manual,[24] and an average taken of the remaining readings.

Statistical analysis

All AS variables (BrAIx, AoAIx and AoPWV) were log-transformed. We fitted a linear mixed model on each log transformed AS variable with group, gestational window (W1, W2, W3 and W4), maternal age, ethnicity, parity, BMI, mean arterial pressure (MAP) and heart rate (HR) as fixed effects and the individual participant as a random effect. We evaluated each model for the two-way interaction effect of group and gestational window and retained the interaction term when it was statistically significant ($p<0.05$). All models estimated the variance at different gestational windows to account for the heterogeneity of variance at different periods. For comparison of AS between low-risk controls and women with GDM, the contrast estimated the difference in means between the control group and all women with GDM (i.e., those on the diet, metformin and metformin plus insulin combined into a single

group), and between the control group and the women with GDM in individual treatment sub-groups.

All statistical tests were two-sided with the type 1 error rate (p-value) of 0.05 to determine statistical significance. The p-values obtained from all group comparisons were adjusted using the Bonferroni correction to account for multiple comparisons. All statistical analyses were performed using the R software version 4.0.3 with the R packages nlme, emmeans, and ggplot2 (R Core Team, 2020).

RESULTS

In total, 211 women with GDM were recruited to the study. Among them, 84 of these women were later excluded; 45 changed treatment modalities during the study period, 31 attended only a single study visit, 5 patients did not meet the study criteria (later diagnosed with having a fetal abnormality or pre-existing diabetes), and 3 women were treated with insulin only. After exclusion of these women, 127 women with GDM and 155 low risk controls were included in the analysis. Of the women with GDM, 59 were treated with dietary management, 47 with metformin alone, and 21 with metformin in combination with insulin.

Baseline characteristics and pregnancy outcomes of the control group, and the women with GDM are described in Table 1. Compared with the low-risk controls, pregnant women in the combined GDM group were older, had higher BMI, with higher proportion of non-White ethnicity (all $p<0.05$). Amongst the women with GDM, fasting oral glucose tolerance test (OGTT) levels were significantly lower in the women controlled by dietary management compared to those requiring metformin and metformin plus insulin ($p<0.001$ for both comparisons). 2 hour OGTT levels were significantly lower in the diet group compared to the metformin plus insulin group ($p=0.002$), but not the metformin group ($p=0.07$).

Women with GDM delivered at an earlier gestation than the women in the control group ($p<0.001$). After adjusting for gestation at delivery, women with GDM controlled by dietary intervention delivered neonates with a smaller birth weight centile than the low-risk controls ($p<0.001$), but there was no statistically significant difference in neonatal birthweight centile between the controls and women with GDM controlled pharmacologically ($p>0.05$).

Amongst the women treated pharmacologically, all women were first commenced on metformin, and then insulin later. The mean gestation at the commencement of metformin

therapy was significantly earlier in the metformin plus insulin group compared to the metformin only group: 21.1 ± 6.3 *vs* 27.1 ± 5.6 weeks, p<0.001. Median duration of metformin treatment prior to the first and final hemodynamic assessment was significantly longer in the metformin with insulin group compared to the metformin only group: 3.1 (interquartile range (IQR), 1.7 – 5.7) *vs* 7.3 (IQR, 5.0 – 11.3) weeks, p<0.001, and 8.1 (IQR, 6.0 – 13.1) *vs* 12.4 (IQR, 10.1 – 22.3) weeks, p<0.001, respectively.

Changes in arterial stiffness

a. Association of arterial stiffness with maternal characteristics

Maternal BMI at booking was negatively associated with BrAIx (p=0.012) and AoAIx (p=0.010) but positively associated with PWV (p=0.029). All AS variables (BrAIx, AoAIX, PWV) demonstrated a positive association with maternal MAP at booking (p<0.001). Both maternal age (p=0.008) and HR (p<0.001) showed a positive association with PWV. Maternal age was positively associated with BrAIx (p=0.003). Maternal HR was negatively associated with AoAIx (p<0.001). There was no association between maternal ethnicity (White *vs* Non-white) or parity (nulliparous *vs* multiparous) with AS variables (p>0.05). Detailed outputs of the fitted linear mixed models are presented in Tables S2-S4.

b. Low-risk controls vs total women with GDM

The two-way interaction term of study group and gestational age was significant for BrAIx and AoAIx (p<0.001), however, there was no evidence (p=0.729) that mean AoPWV was different between the study groups. Mean difference between the control groups and all women with GDM (as a combined group) for log adjusted BrAIx and AoAIx at each gestational window are demonstrated in Table 2. Women in the total GDM group demonstrated significantly higher

mean BrAix and AoAIx compared to low-risk controls at W1, W2 and W3 ($p<0.05$) but not at W4.

c. Low-risk controls vs women with GDM by treatment modality

Adjusted predicted mean and 95% CI for BrAIx, AoAIx and PWV for control and individual treatment groups at each gestational window are presented in Table 3, and Figures 1-3. Women in each of the GDM treatment groups (dietary management, metformin alone and metformin with insulin) demonstrated significantly higher mean BrAIx and AoAIx compared to the control group at three gestational windows: W1, W2 and W3 ($p<0.05$). There was no difference between any of the GDM treatment groups and low-risk controls at W4 for in BrAIx and AoAIx ($p>0.05$). There was no difference in PWV between the control group and any of the GDM treatment groups at any gestational window ($p>0.05$).

d. Women with GDM managed with dietary intervention vs metformin

The difference in BrAIx, AoAIx and PWV between the women with GDM on diet, metformin or metformin with insulin was not statistically significant at any gestational window. However, we did observe a contrasting pattern of BrAIx and AoAIx across advancing gestation. Amongst the women with GDM controlled with dietary intervention, the predicted mean BrAIx fell between W1 and W2, from -60.71% to -62.64%, and then rose to -53.07% at W3. In the women with GDM taking metformin, predicted mean BrAIx also fell between W1 and W2 from -57.25% to -58.14%, but then remained stable at -57.89% in W3 before rising to -53.28% at W4. In the women with GDM taking metformin with insulin, predicted mean AoAIx fell between W1 and W2 from -50.53% to -58.87%, and then remained stable at -56.74% in W3 and -56.97% in W4.

Similarly, amongst the women with GDM controlled with dietary intervention, both the predicted mean AoAIx fell between W1 and W2, from 6.77% to 5.52%, and then rose to 9.86% at W3 and 10.4% at W4. In the women with GDM taking metformin, predicted mean AoAIx also fell between W1 and W2 from 8.7% to 8.3%, but then remained stable at 8.49% in W3 before rising to 10.88% at W4. In the women with GDM taking metformin with insulin, predicted mean AoAIx fell between W1 and W2 from 14.31% to 8.28%, and then remained stable at 8.63% in W3 and 7.95% in W4.

DISCUSSION

Summary of main findings

In this prospective longitudinal study, we observed increased AoAIx and BrAIx amongst women with GDM compared to low-risk controls, irrespective of the treatment modality. BrAIx and AoAIx did not differ between individual treatment groups, but there was a trend towards an altered longitudinal pattern of AS between the metformin and diet-controlled groups.

Interpretation of main findings and comparison with literature

The finding of a negative association between maternal BMI and AIx was unexpected, given that systemic vascular resistance (SVR) has previously been reported to be higher in morbidly obese women compared to non-obese pregnant women – although the difference was only significant when reported as the systemic vascular resistance index, and not SVR[25]. In the current study, the majority (84.4%) of participants were not obese, and hence, the observed effect is limited to our study population and may not be applicable to a wider population. It should also be noted that the effect size for BMI is reasonably small compared to the effect of the study groups and therefore the variability of the AIx due to variation in maternal BMI is minimal. A negative association between BMI and AIx has previously been reported in certain populations[26, 27]. The relationship between these variables may therefore not be fully understood and warrants further investigation with a large dataset and adequate distribution of participants across all BMI categories.

In keeping with several previous studies,[5-7] we found no difference in PWV between women with GDM and healthy pregnancies. The only study[8] to have reported higher PWV amongst

women with GDM had a larger population (218 women) and examined PWV at a single time point (35-37 weeks gestation) only.

Our finding of increased AIx amongst the women with GDM is in agreement with a number of previous studies,[7, 10] but at odds with three others[6, 8, 9]. This contrast in findings might be explained by the differences in study design. All three examined AS at a single time point in third trimester, and two[6, 9] included fewer than 35 women with GDM. Mansukhani et al[8] included a larger number of women, but examined AIx at 35-37 weeks' gestation, which straddles two gestational windows in our study, including the ≥36+0 window at which we also found no significant difference between the women with GDM and healthy controls.

An important finding of our study is that BrAIx and AoAIx were higher in the women with GDM when compared to healthy controls, regardless of their treatment type. A previous longitudinal pilot study[19] found a significant difference in BrAIx and AoAIx between healthy controls and women with GDM treated with metformin at 26-28 weeks only and at 32-34 weeks only for those managed with dietary intervention. The current study was larger, and we found a significant difference between the controls and women with GDM on all treatment modalities at three gestational windows, from 24+0 through to 35+6 weeks gestation.

Monitoring of glycemic response to treatment for GDM at each gestational window was beyond the scope of this study. However, since pharmacological treatment was initiated based on the participants self-monitored blood glucose levels, the different treatment groups as well as the birthweight and centiles of the neonates can be interpreted as indirect markers of glycemic control. There was also no difference in rates of large for gestational age babies between the controls and any of the GDM groups, implying that our cohort of women with

GDM were generally well controlled. It is therefore interesting to note that we found increased BrAIx and AoAIx amongst the women with GDM, despite their apparently being adequately treated.

The increase in BrAIx and AoAIx seen in the GDM dietary management and healthy control groups was suppressed until 35+6 weeks of gestation for those with GDM treated with metformin alone, and until ≥36+0 weeks gestations for those treated with metformin with insulin. Neonatal birth weight centile was significantly higher in the metformin treated groups compared to the dietary management group, and we therefore consider it unlikely that this observation represents tighter glycemic control in the groups treated pharmacologically. Several studies conducted in non-pregnant populations have reported a significant reduction in AIx associated with treatment with metformin[15-17]. At a molecular level, metformin treatment decreases inflammation and promotes angiogenesis in pregnant populations,[28] and improves oxidative stress and endothelial function in animal models[29]. Our findings suggest that metformin may play a role in attenuating the physiological increase in AS seen during pregnancy in healthy controls and GDM diet groups in the mid-to-late third trimester. It is however important to note that the difference in BrAIx and AoAIx between any of the GDM groups was not significant at any of the gestational windows, and that the study was underpowered for this comparison. This observation therefore remains a hypothesis which would warrant further investigation.

Strengths and limitations

This is the largest study to investigate maternal hemodynamics in GDM throughout the third trimester, rather than at a single time-point. We have also considered women with GDM on different treatments as separately, rather than as a combined group, allowing us to

characterize a more detailed picture of AS in pregnancies complicated by GDM than has previously been described in the literature.

A limitation of the study is that we were unable to assess hemodynamics prior to the initiation of treatment or collect data regarding the glycemic control after diagnosis. Whilst the need for pharmacological treatment and the neonatal birthweight act as indirect markers of glycemic control, we are, therefore, unable to conclude or exclude that glycometabolic decompensation accounted for the difference in hemodynamics between the groups. Secondly, whilst this study is amongst the largest to look at AS amongst women with GDM, the treatment subgroups were still relatively small. Finally, we were restricted from inviting postnatal women for follow-up by the COVID-19 pandemic, and so our data does not assess if the differences in AS observed in pregnancy persist after delivery.

Clinical and research implications

Our findings demonstrate that women with GDM have altered measures of AIx during the third trimester compared to healthy pregnancies. Women with metabolic syndrome have increased measures of AIx,[30] and so it is possible that our findings reflect the pre-clinical risk factors and predisposition for development of GDM. Since increased AS predicts the development of gestational hypertensive disorders,[31, 32] our findings also suggest that maladaptation of the cardiovascular system in pregnancies complicated by GDM may underlie the association between GDM and PIH and PET. Further work is needed which will correlate increased AIx in pregnant women with GDM these clinical outcomes.

Our findings also suggest that treatment with metformin could possibly attenuates the changes in BrAIx and AoAIx in the mid-to-late third trimester in low-risk and GDM diet-controlled pregnancies, which may explain why metformin therapy has been associated with

decreased rates of PET and PIH in obese pregnant women.[12] Prospective interventional studies are therefore needed to further investigate this effect of metformin and its correlation with the risk of development of placental-mediated diseases.

DISCLOSURE

T. R. is a NIHR Senior Investigator.

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FIGURE LEGENDS

Figure 1: Brachial augmentation index by gestational window and study group

Figure 2: Aortic augmentation index by gestational window and study group

Figure 3: Pulse wave velocity by gestational window and study group

Table 1: Baseline characteristics and pregnancy outcomes of study population

Baseline characteristics	Controls (n = 155)	Total GDM (n = 127)	Diet (n = 59)	Metformin Only (n = 47)	Metformin + Insulin (n = 21)
Maternal age (years)	29.5 ± 5.32	32.0 ± 5.19	31.5 ± 5.45	32.0 ± 4.96	33.7 ± 4.83
Maternal height (cm)	164.6 ± 7.07	161.8 ± 6.09	161.2 ± 5.67	161.2 ± 6.12	164.9 ± 6.54
Maternal weight (kg)	68.7 ± 15.67	77.1 ± 21.58	69.9 ± 16.16	81.4 ± 24.7	89.7 ± 19.36
Maternal BMI at booking (kg/m^2)	25.3 ± 5.03	29.3 ± 7.54	26.5 ± 5.60	31.2 ± 9.03	32.8 ± 5.92
Parity					
Nulliparous	64 (41.3)	52 (40.9)	24 (40.7)	22 (46.8)	6 (28.6)
Multiparous	91 (58.7)	75 (59.1)	35 (59.3)	25 (53.2)	15 (71.4)
Maternal ethnicity					
White British/ European	124 (80.0)	55 (43.3)	30 (50.8)	16 (34.0)	9 (42.9)
Non-white	31 (20.0)	72 (56.7)	29 (49.2)	31 (66.0)	12 (57.1)
Current smoker	5 (3.2)	3 (2.4)	2 (3.4)	1 (2.1)	0 (0)
OGTT fasting glucose (mmol/L)	n/a	4.8 ± 0.84	4.4 ± 0.71	5.0 ± 0.72	5.5 ± 0.9
OGTT 2 hour glucose (mmol/L)	n/a	8.8 ± 1.51	8.4 ± 0.97	8.8 ± 1.35	9.7 ± 2.52
Pregnancy Outcomes					
Gestational age at delivery (weeks)	39.8 ± 1.1	38.8 ± 1.1	39.1 ± 1.3	38.8 ± 0.9	38.3 ± 0.7
Hypertensive disorder of pregnancy	0 (0)	10 (7.9)	5 (8.5)	2 (4.3)	3 (14.3)
Birth weight (g)	3558 ± 419	3280 ± 468	3190 ± 463	3320 ± 459*	3446 ± 467
Birth weight centile	58.4 ± 26.7	49.5 ± 31.9	40.5 ± 30.5	53.2 ± 30.8	66.5 ± 31.1
Birth weight category					
Small for gestational age	0 (0)	20 (15.7)	13 (22.0)	6 (12.8)	1 (4.8)
Large for gestational age	23 (14.8)	13 (10.2)	3 (5.1)	5 (10.6)	5 (23.8)

Data are given as mean ± SD or n (%). OGTT = oral glucose tolerance test. Small for gestational age = birthweight <10th centile; large for gestational age = birthweight >90th centile – both according to population-based growth charts.[22]

Table 2: Mean difference in log adjusted BrAIx and AoAIx between controls and all women with gestational diabetes

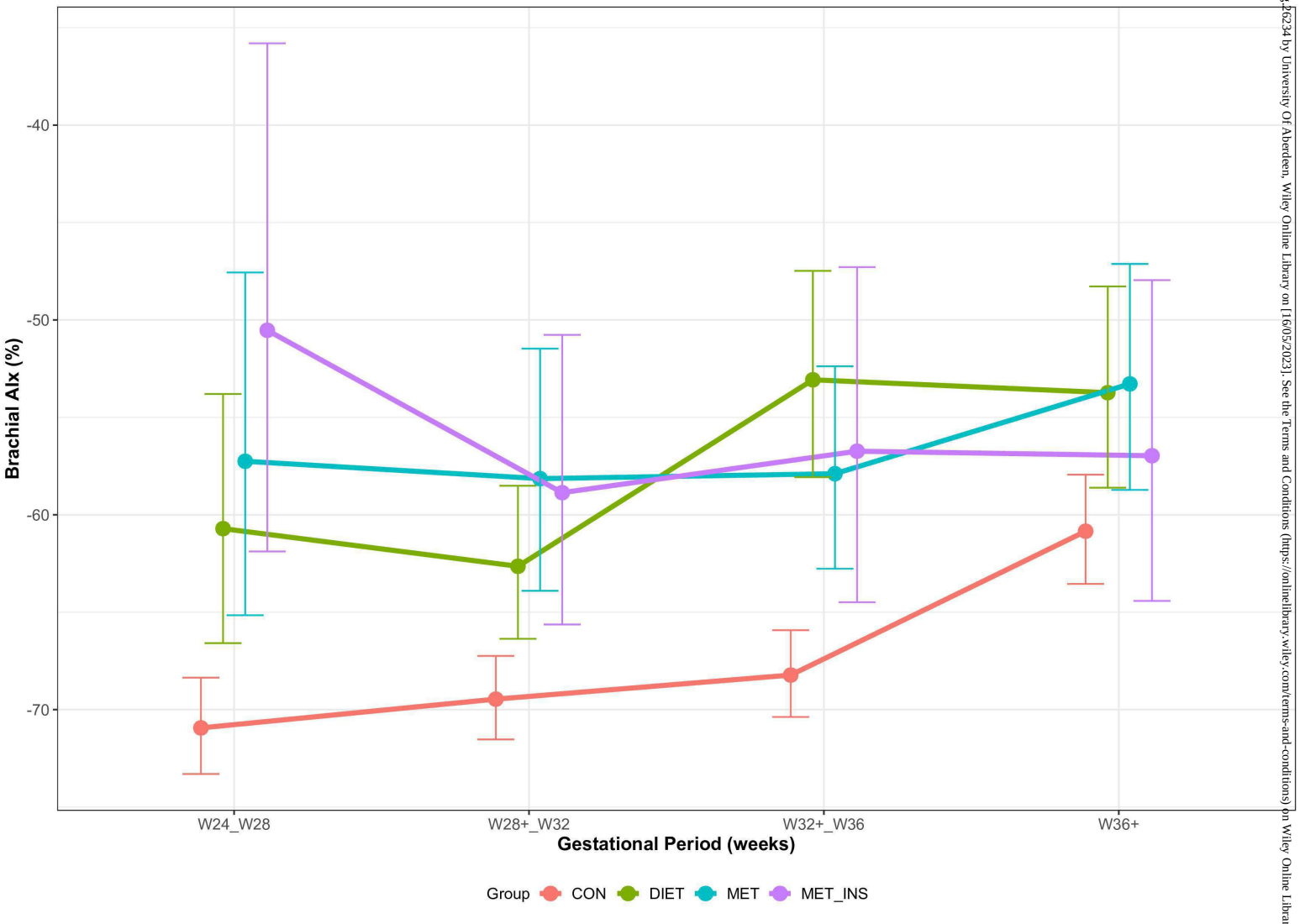
	24+0 to 27+6		28+0 to 31+6		32+0 to 35+6		≥36+0	
	Mean difference	**Adjusted P value**	**Mean difference**	**Adjusted P value**	**Mean difference**	**Adjusted P value**	**Mean difference**	**Adjusted P value**
BrAIx	-0.37 (-0.52, -0.22)	<0.001	-0.23 (-0.35, -0.12)	0.001	-0.29 (-0.40, -0.18)	<0.001	-0.11 (-0.22, 0.00)	>0.999
AoAIx	-0.49 (-0.69, -0.3)	<0.001	-0.32 (-0.47, -0.18)	<0.001	-0.38 (-0.52, -0.24)	<0.001	-0.17 (-0.32, -0.03)	0.405

Data are given as mean difference (95% CI). P-values were adjusted using the Bonferroni correction.

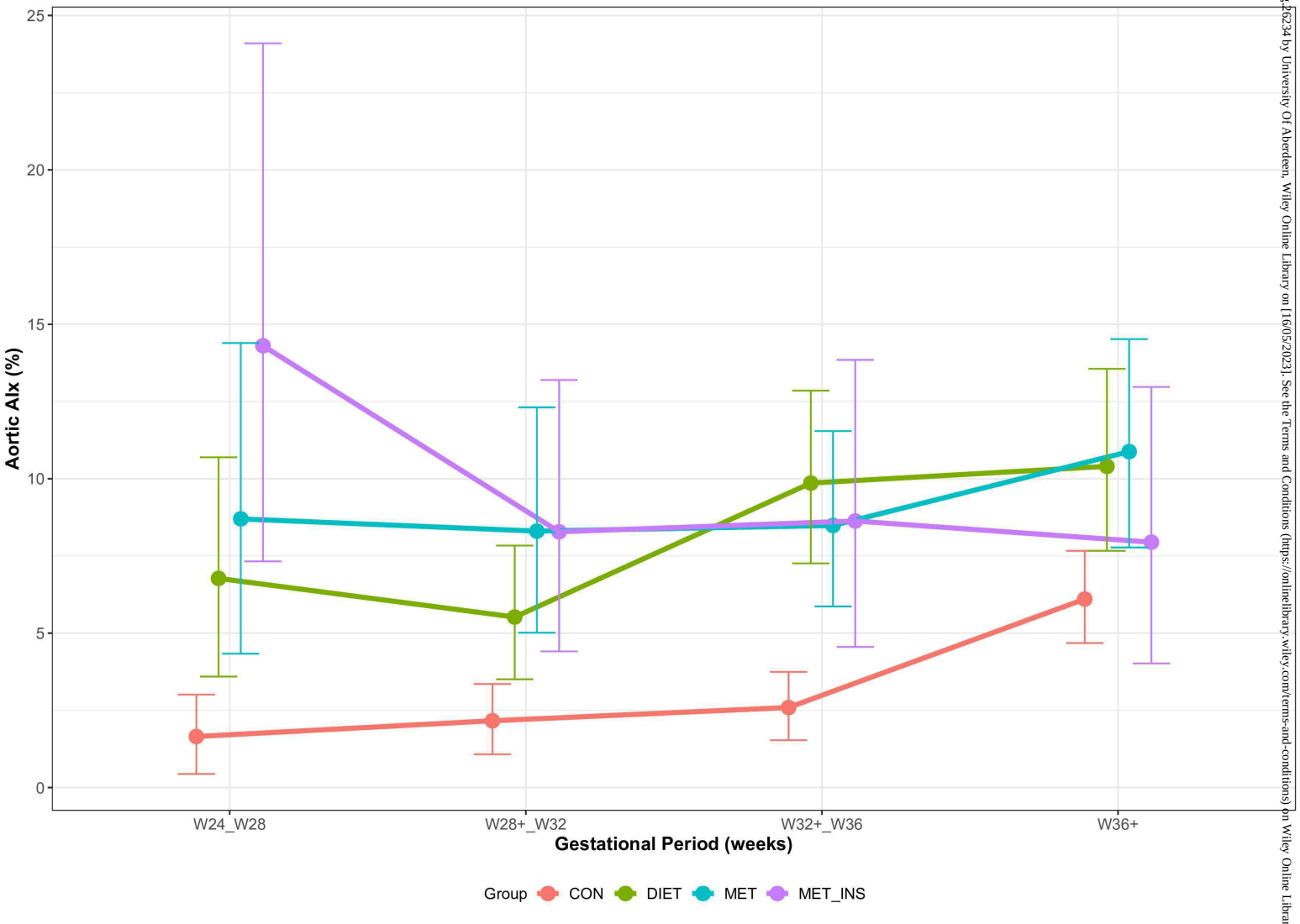
Table 3: Brachial and aortic augmentation index and pulse wave velocity by gestational window and treatment group

Group	24+0 to 27+6 weeks (W1)	28+0 to 31+6 weeks (W2)	32+0 to 35+6 weeks (W3)	≥36+0 weeks (W4)
Brachial Augmentation Index (BrAIx)				
Control	-70.94 (-73.31, -68.36)	-69.46 (-71.53, -67.25)	-68.23 (-70.38, -65.92)	-60.84 (-63.55, -57.94)
Diet	-60.71 (-66.59, -53.80) *	-62.64 (-66.37, -58.51) *	-53.07 (-58.07, -47.48) *	-53.73 (-58.61, -48.28)
Metformin	-57.25 (-65.15, -47.56) *	-58.14 (-63.90, -51.47) *	-57.89 (-62.77, -52.38) *	-53.28 (-58.72, -47.12)
Metformin + Insulin	-50.53 (-61.88, -35.80) *	-58.87 (-65.63, -50.77) *	-56.74 (-64.49, -47.29) *	-56.97 (-64.42, -47.96)
Aortic Augmentation Index (AoAIx)				
Control	1.65 (0.44, 3.01)	2.16 (1.08, 3.35)	2.59 (1.53, 3.75)	6.10 (4.68, 7.67)
Diet	6.77 (3.59, 10.69) *	5.52 (3.51, 7.84) *	9.86 (7.26, 12.85) *	10.40 (7.67, 13.56)
Metformin	8.70 (4.33, 14.39) *	8.30 (5.02, 12.31) *	8.49 (5.86, 11.54) *	10.88 (7.78, 14.52)
Metformin + Insulin	14.31 (7.32, 24.10)*	8.28 (4.41, 13.20) *	8.63 (4.56, 13.85) *	7.95 (4.02, 12.97)
Aortic Pulse Wave Velocity (PWV)				
Control	7.73 (7.51, 7.96)	8.01 (7.83, 8.19)	8.19 (8.00, 8.38)	8.43 (8.24, 8.62)
Diet	7.76 (7.47, 8.06)	8.04 (7.79, 8.29)	8.22 (7.96, 8.49)	8.46 (8.20, 8.72)
Metformin	7.90 (7.57, 8.24)	8.18 (7.88, 8.49)	8.37 (8.07, 8.68)	8.61 (8.31, 8.92)
Metformin + Insulin	7.89 (7.46, 8.34)	8.17 (7.76, 8.61)	8.35 (7.93, 8.80)	8.59 (8.16, 9.05)

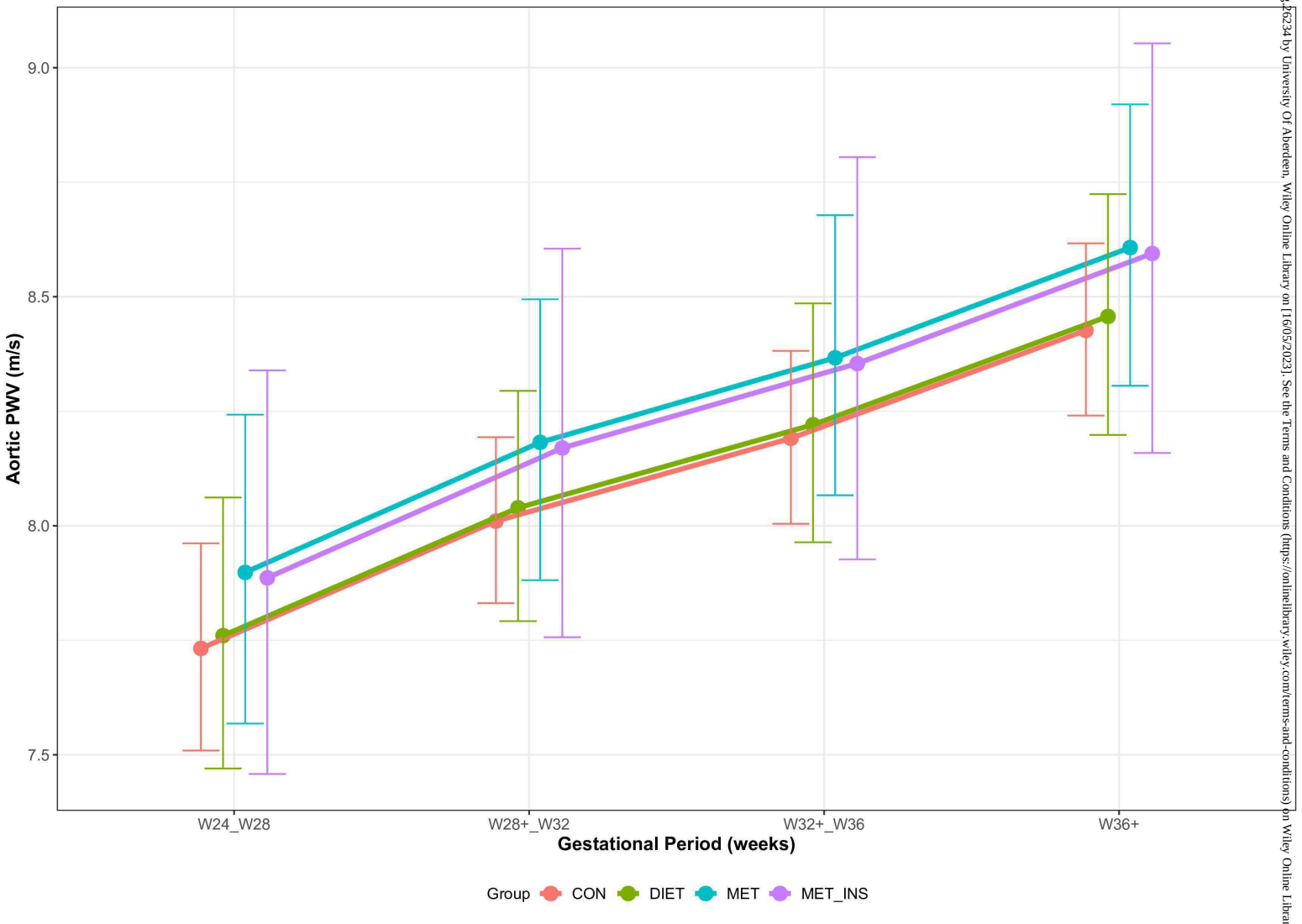
Data are given as mean (95% CI). *Indicates significant difference from control group within the same gestational window ($P<0.05$).



UOG_26234_FIGURE 1 BrAIx.jpg



UOG_26234_FIGURE 2 AoAIx.jpg



UOG_26234_FIGURE 3 PWV.jpg