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Maternal obesity and sex-specific differences in placental pathology

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ABSTRACT

Objective: Adverse effects of obesity have been linked to inflammation in various tissues, but studies on placental inflammation and obesity have demonstrated conflicting findings. We sought to investigate the influence of pregravid obesity and fetal sex on placental histopathology while controlling for diabetes and hypertension.

Methods: Placental histopathology focusing on inflammatory markers of a cohort of normal weight (BMI = 20–24.9) and obese (BMI ≥ 30) patients was characterized. Demographic, obstetric and neonatal variables were assessed.

Results: 192 normal and 231 obese women were included. Placental characteristics associated with obesity and fetal sex independent of diabetes and hypertension were placental disc weight >90th percentile, decreased placental efficiency, chronic villitis (CV), fetal thrombosis, and normoblastemia. Additionally, female fetuses of obese mothers had higher rates of CV and fetal thrombosis. Increasing BMI increased the risk of normoblastemia and CV. The final grade and extent of CV was significantly associated with obesity and BMI, but not fetal gender. Finally, CV was less common in large-for-gestation placentas.

Conclusions: Maternal obesity results in placental overgrowth and fetal hypoxia as manifested by normoblastemia; it is also associated with an increased incidence of CV and fetal thrombosis, both more prevalent in female placentas. We have shown for the first time that the effect of maternal obesity on placental inflammation is independent of diabetes and hypertension, but significantly affected by fetal sex. Our data also point to the intriguing possibility that CV serves to normalize placental size, and potentially fetal growth, in the setting of maternal obesity.

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1. Introduction

Over the last decade, studies have established that maternal obesity increases morbidity and mortality for the maternal–fetal dyad [1,2]. Maternal morbidities include gestational diabetes mellitus (GDM), gestational hypertension (GHTN), preeclampsia/eclampsia [1,3], labor dystocia [4], operative and Cesarean delivery, and postpartum hemorrhage [5]. Offspring of obese mothers are at a higher risk of macrosomia (birth weight > 4 kg) as well as intrauterine fetal demise (IUFD) [6,7]. Furthermore, these neonates are at increased risk of obesity and metabolic disease later in their own adult life [8,9].

Several studies support the association between pregravid obesity and placental pathology. These include an association with increased placental weight and reduced efficiency, the latter measured as a ratio of birthweight to placental weight [10,11]. Likewise, inflammation in the form of chorioamnionitis has been documented in placentas of obese women [11,12]; this lesion is most often a mixed infiltrate composed of neutrophils and lymphocytes and is thought to be maternal in origin, although it can be accompanied by a fetal response [13]. This is not surprising given the increased risk of labor dystocia and prolonged ruptured membranes [4].

However, the more common inflammatory cell type seen infiltrating tissues in the setting of obesity is the macrophage [14], a cell type often seen in a placental disc lesion called “chronic villitis” [13,15]. Chronic villitis (CV) is defined by the presence of a mixture of macrophages and lymphocytes—mostly T cells—infiltrating

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chorionic villi [15]. While some are secondary to infection (i.e. with TORCH-related microorganisms such as CMV), the majority cannot be linked to any pathogen and are therefore thought to represent a transplantation rejection reaction, otherwise known as villitis of unknown etiology or VUE [13,15]. Where this lesion involves large areas of the placenta, it has been associated with intrauterine growth restriction and recurrent pregnancy loss [13,15]. In addition, and particularly when associated with obliterative fetal vascular lesions ("obliterative fetal vasculopathy"), CV is also known to be a risk factor for neonatal encephalopathy and cerebral palsy [15–17].

Evaluations of CV in the setting of maternal obesity have shown conflicting results; however, these studies have been relatively small in sample size (<50 per group): Challier et al. noted an increase in the number of resident macrophages in the placentas of obese women [18], while Roberts et al. were unable to see major differences in immune cell infiltration within the chorionic villi [19], instead, only noting increased expression of pro-inflammatory cytokines in such placentas. Larger studies have not addressed this question [11].

Pregnancy outcomes and placental pathology are also dependent on fetal sex [20]. Male neonates are often larger than females and have an increased risk of preterm birth and stillbirth, while pregnancies with female fetuses tend to be at a higher risk of preterm preeclampsia [21–23]. Relatively few studies have evaluated sex-related differences in placental pathology. Walker et al. reported placentas of male fetuses having significantly higher rates of velamentous insertion of umbilical cord and chronic deciduitis, and lower rates of villous infarction compared to those of female fetuses [24]. A relationship with fetal sex has not been described for either CV or fetal thrombosis.

This study aimed to evaluate placental pathology in the setting of pregravid obesity in patients delivering at or near term, with a focus on CV, as this is the main lesion involving infiltration of macrophages in the placental villi. Given the links between maternal obesity with diabetes and hypertensive diseases of pregnancy, as well as the association between fetal sex and pregnancy outcomes, this study explored the influence of these variables on placental pathology in the setting of maternal obesity. The hypothesis was that maternal obesity is linked to specific pathologic findings of the placenta, which differ based on fetal sex.

2. Materials and methods

The study, approved by the Human Research Protections Program Committee of the University of California San Diego (UCSD) Institutional Review Board, is part of a Perinatal Biospecimen Banking study, where both low- and high-risk pregnant patients are consented, and both clinical data as well as various biospecimens, including placental tissue, are collected. The subjects for this study were selected from amongst this study population based on the following criteria: delivery of a singleton, live-born infant at or after 35 weeks gestational age between January 1, 2010 to April 30, 2013; and documented maternal pregravid or early pregnancy weight (obtained prior to 14 weeks). Maternal demographic, anthropometric, and obstetric data, as well as neonatal outcomes data were abstracted from the electronic medical record. Placental examination, including gross and histologic examinations, were performed either through the hospital (for those with a clinical indication for placental exam) or through the research core (for those placentas without such indications).

2.1. Maternal data

Specific maternal factors included maternal age, ethnicity, pregravid BMI, BMI at delivery, gestational age, diabetes, hypertension and mode of delivery. The majority of Non-Hispanic women were White Non-Hispanic. Asian and Black participants comprised 4.4% and 2.4% of the study population, respectively. For calculation of all BMI measured weight and height were used. For pregravid BMI, measured weight under 14 weeks gestation was used. For BMI at delivery, measured weight at either the last prenatal visit or at admission for delivery was used. Maternal BMI was classified as normal weight (20–24.9 kg/m^2), class I obesity (30.0–34.9 kg/m^2), class II obesity (35.0–39.9 kg/m^2), or class III obesity (≥40.0 kg/m^2). For the majority of analyses, maternal obesity was used as a dichotomous variable. For dichotomous analyses, class 1 to 3 obesity were aggregated into one group. The diagnosis of diabetes included pre-gestational type 2 diabetes (T2DM) or gestational diabetes type 2 (GDMA2); patients with type 1 diabetes (T1DM) or gestational diabetes type 1 (GDMA1) were excluded. Patients with GDMA2 were identified by an abnormal 2-h or 3-h glucose tolerance test, and treated with oral medication or insulin therapy for adequate glycemic control. Hypertensive disorders were classified based on the following: Chronic Hypertension (CHTN), defined by high blood pressure persistently at or above 140/90 mmHg, diagnosed before 20 weeks gestation without proteinuria. Gestational hypertension (GHTN), defined by elevated blood pressure at or above 140/90 mmHg on at least two occasions at least 6 h apart after the 20th week of gestation in women known to be normotensive before pregnancy and before 20 weeks gestation without other signs or symptoms of preeclampsia based on the 2013 American Congress of Obstetricians and Gynecologists guidelines [25]. Preeclampsia or preeclampsia with severe features was defined based on ACOG criteria [25].

2.2. Neonatal data

Neonatal gender, birthweight (grams) and the APGAR scores at 1 and 5 min were recorded. Birthweight percentile (BW%) was adjusted for gender and gestational age based on the 1999–2000 US Natality Datasets [26,27]; category of growth was classified by the following defined as follows: small for gestational age (SGA) as birth weight of ≤10th percentile, normal for gestational age as birth weight between 11th and 89th percentile, and large for gestational age (LGA) as birth weight ≥ 90th percentile. Macrosomia was defined as birth weight greater than 4000 g [6,7,28,29].

2.3. Placental histopathology data

Placental examination was performed by a single pathologist (MMP) using standard protocols [13].

2.3.1. Gross examination

Size, weight, and lesions of the placental disc were recorded. Specifically, the weight of the unfixed disc was measured without attached cord and membranes. Placental weight percentile was adjusted for gestational age [30], and designated as "large" if it was ≥90th percentile for gestation. Placental efficiency was defined by the ratio of fetal to placental weight at delivery.

Fetal membrane (color and insertion) and umbilical cord (length, insertion site, and lesions) were also evaluated. "Umbilical cord abnormality" was defined based on factors associated with umbilical cord obstruction, and included abnormally long cord, hypertwisting (>3 twists per 10 cm), or the presence of true knot(s) [31].

For each placenta, the following sections were examined histologically: one section each of umbilical cord and membrane roll; any disc lesions; and two full-thickness sections of grossly-normal placental disc. All these sections were processed, embedded in paraffin, sectioned, and stained with hematoxylin and eosin (H&E)

using standard protocols.

2.3.2. Histologic examination

Each subject had a detailed placental pathology examination by a single pathologist (MMP). The pathologist was blinded to fetal-sex and maternal BMI. Only minimal obstetric information was recorded in the pathology request form, including gestational age and indication for placental examination (i.e. diabetes, pre-eclampsia, intrauterine growth restriction). The presence or absence of the histopathologic findings listed in Table 1 was documented. These findings were defined by previously-established criteria [13].

Placentas with CV were graded jointly by one board-certified anatomic pathologist (MMP) and one pathologist-in-training (XL), blinded to clinical information, using a modified version of the Redline method [15], where a focus of CV was classified as low grade (<10 inflamed villi per focus), high grade (>10 inflamed villi per focus but each focus still fits into a 10x field), or diffuse (focus of villitis fills up more than a 10x field). For placentas with only low and/or high grade foci, the number of CV foci were counted; then the following formula was used to determine the final grade of the entire case:

$$[(\text{Number low grade foci} \times 1.0) + (\text{number of high grade foci} \times 2.0)]/\text{total number of foci}$$

Each case was graded as low grade (grade = 1) if the above value was <1.5 and high grade (grade = 2) if the value was >1.5; if even one focus of diffuse CV was identified, it was considered diffuse (grade = 3).

For evaluating extent of CV, the numerator of the above formula was used [(Number low grade foci × 1.0) + (number of high grade foci × 2.0)]. Among those with only low and/or high grade CV, the highest score was 11; therefore, all "diffuse" cases were arbitrarily assigned an "extent" score of 12.

A total of 20 cases (5 from each of 4 categories: normal vs. obese, with and without CV) were also evaluated by immunohistochemistry for T-cells (CD3; rabbit anti-CD3 antibody, clone #EP449E, Abcam) and macrophages (CD68; mouse anti-CD68 antibody, clone #KP1, Dako), using a Ventana automated stainer.

2.4. Statistical analysis

Statistical analysis was performed using SPSS software (SPSS release 21.0, Chicago, IL, USA). Bivariate analyses were performed to examine maternal and perinatal differences between normal and obese groups. Student's t-test and Chi-square were used for analyses of continuous and categorical variables, respectively. Goodman's Gamma test for ordinal variables was performed for birth weight percentile classifications. The same approach was applied to evaluate maternal and perinatal differences between CV, fetal thrombosis, normoblastemia, and large placental disc groups.

Placental pathologic outcomes, including gross and histologic findings were compared between maternal weight (lean/obese) and fetal sex groups in bivariate and multivariate analyses. Placental findings (placental large disc, normoblastemia, CV, and fetal thrombosis) were adjusted for maternal, placental and gender covariates in the multivariate analysis. Only variables with a p value of <0.2 were considered candidate covariates. The choice of relevant covariates for adjustment was done by backward model selection. The candidate covariates were fetal sex, parity, maternal obesity, maternal hypertensive disorder, preterm delivery, and placental findings (large disc weight, abnormal umbilical cord, CV, fetal thrombosis, intervillous thrombus, decidual vasculopathy, infarcts, normoblastemia, fetal vasculitis, and retroplacental hematoma). Finally, multivariate logistic regression models were used to compare the proportion of placental findings that were associated with maternal obesity and fetal sex. Adjusted odd ratios (aOR) and 95% confidence intervals (95% CI) were reported. All statistical tests were two-sided, with p-values less than 0.05 considered statistically significant.

3. Results

3.1. Maternal and perinatal characteristics in relation to maternal BMI

Four hundred and twenty-three women were included in the study (192 normal weight and 231 obese). Maternal and fetal characteristics are summarized in Table 2. Mean maternal age and mode of delivery were similar between groups. Obese women were more likely to be Hispanic, multiparous, diabetic, and hypertensive, with a lower gestational age at delivery. Fetal gender and APGAR scores were not different between the two groups. As anticipated, birthweight demonstrated a significant positive association with maternal obesity.

3.2. Placental findings by maternal characteristics and fetal sex

The mean disc weight of placentas from obese patients was 44 g greater than normal-weight patients (Table 3). Other significant placental pathology associated with obesity included CV, normoblastemia, and decidual vasculopathy (Table 3). Interestingly, CV was more common in female placentas, but only in the setting of maternal obesity (p = 0.018). In the bivariate analysis, decidual vasculopathy was associated with obesity, but multivariate analysis

Table 1
Histologic findings in relation to maternal/fetal/placental conditions.

Maternal/fetal/placental condition	Histologic finding(s) in the placenta
Intrauterine hypoxic stress	Meconium-laden macrophages in fetal membranes
	Normoblastemia
Amniotic fluid infection	Chorioamnionitis
	Fetal vasculitis (umbilical cord or chorionic plate)
Abnormalities of fetal circulation	Fetal thrombosis (fetal thrombotic vasculopathy, with or without upstream thrombosis in fetal blood vessels)
	Patchy villous edema
Placental Inflammation	Chronic villitis (of unknown etiology)
Maternal vascular abnormalities and related changes	Decidual vasculopathy
	Placental infarction
	Villous hypermaturity
	Intervillous thrombus
	Perivillous fibrin deposition involving >20% of the placental disc
	Retroplacental hemotoma

Table 2
Demographic and clinical characteristics in relation to maternal BMI.

Characteristics	Normal (n = 192)	Obese (n = 231)	P-value
	(Mean ± SD or n(%))		
Maternal characteristics			
Age	32.2 ± 5.5	31.4 ± 5.9	0.162
Parity			<0.001
0	100 (58.1)	72 (41.9)	
1-4	77 (36.8)	132 (63.2)	
≥5	0 (0)	9 (4.2)	
Ethnicity			<0.001
Hispanic	57 (26.4)	159 (73.6)	
Non-Hispanic	132 (65.7)	69 (34.3)	
Pregravid BMI	21.9 ± 1.5	38.2 ± 7.1	<0.001
BMI at delivery	27.6 ± 2.3	42.3 ± 6.8	<0.001
Gestational age	38.6 ± 2.4	37.9 ± 2.6	0.003
Gestational age			0.032
Preterm (<36.6 weeks)	24 (12.5)	47 (20.0)	
Term (>37 weeks)	168 (87.5)	184 (80.0)	
Mode of delivery			0.071
Vaginal	89 (46.4)	87 (37.7)	
Cesarean	103 (53.6)	144 (62.3)	
Diabetes	14 (7.3)	115 (49.8)	<0.001
Any hypertensive disorder	35 (18.2)	122 (52.8)	<0.001
Preeclampsia	21 (10.9)	62 (26.8)	<0.001
Perinatal characteristics			
Fetal sex			0.306
Male	94 (49.0%)	124 (53.7%)	
Female	98 (51.0%)	107 (46.3%)	
Apgar score <7 at 1 min	21 (10.9)	37.0 (16.1)	0.126
Apgar score <7 at 5 min	4 (2.1)	2.0 (0.9)	0.294
Birth weight percentile			<0.001[a]
Small for Gestational Age	34.0 (17.7)	28.0 (12.1)	
Normal for Gestational Age	149.0 (77.6)	164.0 (71.0)	
Large for Gestational Age	9.0 (4.7)	39.0 (16.9)	
Birth weight grams	3124.8 ± 678.3	3283.0 ± 810.2	0.032
Birth weight percentile	38.9 ± 26.9	52.6 ± 31.2	<0.001

[a] Overall difference in birthweight percentile.

Table 3
Placental Histopathologic characteristics by maternal BMI.

Characteristics	Normal (n = 192)	Obese (n = 231)	P-value
	(Mean ± SD or n(%))		
Gross examination			
Disc weight	456 ± 109.9	500 ± 143.2	0.001
Abnormal umbilical cord insertion	30/190 (15.8)	31/230 (13.5)	0.503
Umbilical cord abnormality	14/191 (7.3)	26/230 (11.3)	0.166
Intrauterine hypoxic stress			
Meconium staining of fetal membranes	16 (8.3)	24 (10.3)	0.472
Normoblastemia	29 (15.1)	79 (34.2)	<0.001
Amniotic fluid infection			
Chorioamnionitis	52 (27.1)	53 (22.9)	0.326
Fetal vasculitis	22 (11.5)	20 (8.7)	0.338
Fetal vascular abnormalities			
Fetal thrombosis	33 (17.2)	51 (22.1)	0.209
Villous edema	8/185 (4.3)	11/220 (5.0)	0.749
Placental inflammation			
Chronic villitis	31 (16.1)	60 (26.0)	0.014
Maternal vascular abnormalities			
Decidual vasculopathy	14 (7.3)	43 (18.6)	<0.001
Villous hypermaturity	5 (2.6)	12 (5.2)	0.177
Increased perivillous fibrin deposition	5 (2.6)	7 (3.0)	0.793
Infarcts	26 (13.5)	35 (15.0)	0.639
Intervillous thrombus	22 (11.5)	36 (15.6)	0.219
Retroplacental hematoma	9 (4.7)	9 (3.9)	0.688

did not confirm this association, and instead revealed a persistent relationship between decidual vasculopathy and maternal hypertensive disease with an aOR of 2.87 (95% CI 1.47, 5.58; Table 5). Other placental findings were not significantly different between sex-specific groups in lean vs. obese patients (Tables 3 and 4). GWG data was available in 222 of 231 obese women. In obese women, GWG of less than 11, 11–20, and greater than 20 lbs (occurring in 24.3%, 23.4% and 52.3% of the women, respectively) was not associated with placental disc size (p = 0.178), villitis (p = 0.658) or normoblastemia (0.436).

Table 4
Placental Histopathologic characteristics by maternal BMI and fetal sex.

Characteristics n(%)	Normal (n = 192)		Obese (n = 231)		P-value
	Male n = 94(%)	Female n = 98(%)	Male n = 124(%)	Female n = 107(%)	
Gross examination	**n (%)**				
Large disc weight	10/93 (10.8)	7/97 (7.2)	32/124 (25.8)	20/106 (18.9)	<0.001
Abnormal umbilical cord insertion	15/94 (16.0)	15/96 (15.6)	19/123 (15.4)	12/107 (11.2)	0.734
Umbilical cord abnormality	6/94 (6.4)	8/97 (8.2)	14/123 (11.4)	12/107 (11.2)	0.550
Intrauterine hypoxic stress					
Meconium staining of membranes	9 (9.6)	7 (7.1)	11 (8.9)	13 (12.1)	0.666
Normoblastemia	13 (13.8)	16 (16.3)	41 (33.1)	38 (35.5)	<0.001
Amniotic fluid infection					
Chorioamnionitis	27 (28.7)	25 (25.5)	34 (27.4)	19 (17.8)	0.251
Fetal vasculitis	14 (14.9)	8 (8.2)	12 (9.7)	8 (7.5)	0.301
Fetal vascular abnormalities					
Fetal thrombosis	11 (11.7)	22 (22.4)	18 (14.5)	33 (30.8)	0.002
Villous edema	7/91 (7.7)	1/94 (1.1)	7/118 (5.9)	4/102 (3.9)	0.162
Placental inflammation					
Chronic villitis	15 (16.0)	16 (16.3)	25 (20.2)	35 (32.7)	0.010
Maternal vascular abnormalities					
Decidual vasculopathy	9 (9.6)	5 (5.1)	26 (21.0)	17 (15.9)	0.003
Villous hypermaturity	2 (2.1)	3 (3.1)	6 (4.8)	6 (5.6)	0.568
Increased fibrin deposition	2 (2.1)	3 (3.1)	6 (4.8)	1 (0.9)	0.334
Infarcts	11 (11.7)	15 (15.3)	18 (14.5)	17 (15.9)	0.846
Intervillous thrombus	16 (17.0)	6 (6.1)	19 (15.3)	17 (15.9)	0.096
Retroplacental hematoma	2 (2.1)	7 (7.1)	7 (5.6)	2 (1.9)	0.162

Table 5
Multivariate logistic regression final model for risk of abnormal placental findings by fetal sex, maternal and placental variables.

Placental findings	aOR (95% CI)
Gross examination	
Large disc weight	
Maternal obesity	2.47 (1.29, 4.76)
Maternal diabetes	1.70 (0.94, 3.06)
Placental Chronic villitis	0.51 (0.22, 1.06)
Intrauterine hypoxic stress	
Normoblastemia	
Maternal obesity	2.42 (1.47, 3.97)
Placental Umbilical cord abnormality	2.70 (1.33, 5.52)
Placental Decidual vasculopathy	3.09 (1.69, 5.66)
Fetal vascular abnormalities	
Fetal thrombosis	
Female baby	2.48 (1.47, 4.17)
Placental large disc	2.59 (1.37, 4.89)
Placental Chronic villitis	3.28 (1.88, 5.70)
Villous edema	
Male baby	2.52 (0.88, 7.20)
Pre-term delivery (<36.6 weeks)	2.83 (1.06, 7.56)
Placental inflammation	
Chronic villitis	
Maternal obesity	1.96 (1.18, 3.27)
Placental large disc	0.49 (0.24, 1.02)
Maternal vascular abnormalities	
Decidual vasculopathy	
Any maternal hypertensive disorder	2.87 (1.47, 5.58)
Pre-term delivery (<36.6 weeks)	2.99 (1.53, 5.87)
Normoblastemia	3.15 (1.69, 5.84)

There were no statistically significant interactions. Candidate covariates included fetal sex, maternal obesity, maternal hypertensive disorder, preterm delivery, and placental findings.

After controlling for specific maternal and placental characteristics, as well as fetal sex, *large placental disc weight* was significantly associated with maternal obesity (Table 5). Parity was shown to have a modest influence on placental disc weight (Para 0 = 456.0 ± 122.6 g; Para 1–4 = 497.3 ± 138.4 g; Para ≥ 5 = 483.0 ± 97.7 g; p = 0.006); however after controlling for maternal BMI, this association was no longer statistically significant. The only placental pathologies associated with parity included fetal vasculitis (p = 0.017), maternal infarcts (p = 0.019), and a trend in chorioamnionitis (p = 0.052). Parity did not influence any other placental histopathologic findings. The presence of normoblastemia was associated with obesity, gross umbilical cord abnormalities, and decidual vasculopathy. These associations were not found with maternal diabetes. Inflammation in the placental disc (CV) was also more common in the obese group, independent of maternal diabetes and hypertensive disease (Table 5). We further stratified maternal obesity by pre-pregnancy BMI category, including normal weight (20–24.9 kg/m^2), class I obesity (30.0–34.9 kg/m^2), class II obesity (35.0–39.9 kg/m^2), or class III obesity (≥40.0 kg/m^2). Normoblastemia and CV increased with worsening obesity (p < 0.001 and 0.034 respectively; Fig. 1).

Two placental lesions were found to be associated with fetal sex: fetal thrombosis was more common in female, and villous edema was more common in the male placentas (Table 5). Fetal thrombosis was also associated with both large placental disc and CV, while villous edema was also associated with preterm placentas (Table 5).

The association of CV with a decreased risk of large placental disc nearly reached significance (aOR of 0.49, with CI [0.24, 1.02]). This association persisted when the cases were stratified based on maternal obesity: 14.9% of obese women with CV had large placental discs as opposed to 25.7% of obese women without CV (aOR of 0.51, with CI [0.22, 1.06]).

When evaluating CV with respect to grade and extent, almost half of the CV (28/57) in obese placentas, but only 14% of CV (4/28) in lean placentas, demonstrated a diffuse distribution (X^2 = 9.71, p < 0.01). Using logistic regression analysis, the final grade (low, high, diffuse) and extent (sum of foci with villitis) of CV was significantly associated with obesity and BMI (p = 0.001), but not fetal gender. Immunohistochemical studies confirmed the mixture of T cells (CD3+) and macrophages (CD68+) in foci of CV. The intensity of CD3 and CD68 stains correlated well with the severity of inflammation, with diffuse inflammation always associated with stronger CD3/CD68 staining compared to low- or high-grade inflammation, in both lean and obese placentas (Fig. 2).

Placental efficiency was measured by the ratio of fetal to placental weight; the entire cohort was normally distributed with a mean of 6.83 ± 1.27. Placental efficiency was decreased in the obese

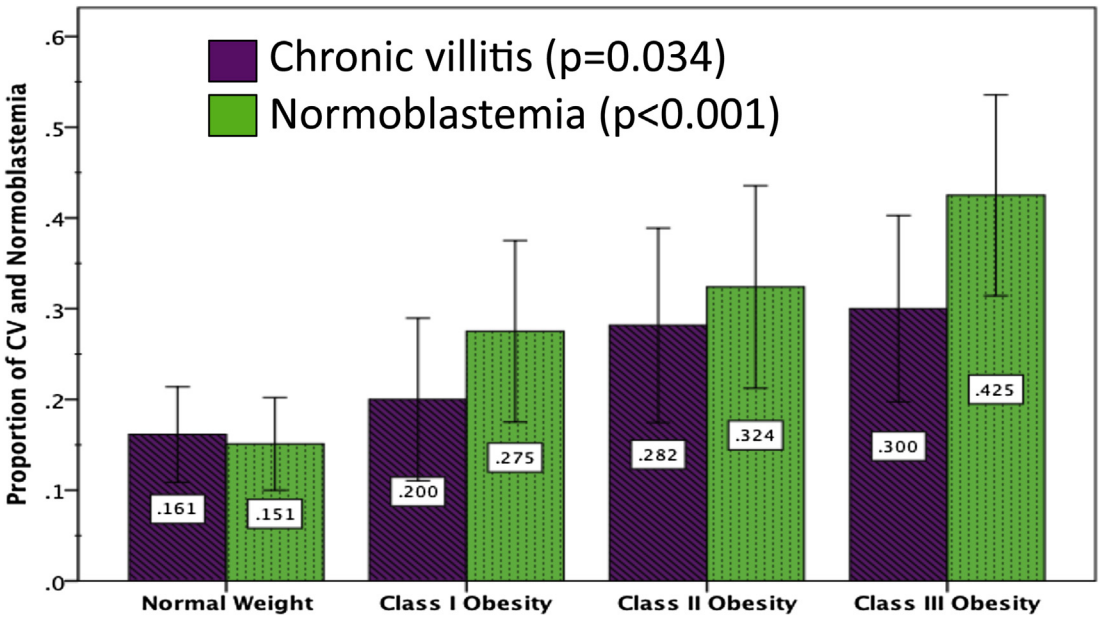

Fig. 1. Proportion of chronic villitis and normoblastemia in different categories of obesity. The risk of CV and normoblastemia is increased with increasing maternal BMI. BMI, body mass index; CV, chronic villitis. Error bars represent standard error of the mean.

group compared to normal-weight women (6.71 ± 1.29 vs. 6.97 ± 1.25, p = 0.042). Bivariate analysis revealed no significant difference in placental efficiency between groups with and without CV (p = 0.113), diabetes (p = 0.347) and any hypertension (p = 0.240); there was also no significant difference in placental efficiency between male and female placentas (p = 0.892). The association between obesity and placental efficiency aOR persisted after controlling for villitis: for every 1 unit increase in BMI, placental efficiency decreased by 0.1. The presence of gender, diabetes and hypertension did not impact the model.

4. Discussion

This study confirms that maternal obesity is associated with placental overgrowth and fetal hypoxic stress as manifested by normoblastemia, the histopathologic correlates to prior studies reporting the association of maternal obesity with macrosomia and fetal distress [6,7]. Likewise, placental efficiency, as measured by fetal to placental weight ratio, was decreased in obese women [10,11]. The average increase in placental weight in our obese population (44 g) was similar to the increase in placental weight associated with multiparity (27–41 g); however, the latter finding was not significant when controlling for BMI.

Novel and more significant to our study was the association of maternal obesity with CV. It is well known that obesity is associated with inflammation, particularly macrophage infiltration, in other tissues and organs, including adipose tissue [14,32]. We found both the prevalence, as well as the grade and extent, of CV to increase with increasing maternal BMI. CV was specifically associated with maternal pre-pregnancy BMI and not gestational weight gain. Although maternal diabetes and hypertension were positively associated with obesity in our study population, as expected [1,3], we found that CV was independent of these maternal comorbidities. Very few previous studies had investigated the association between maternal obesity and macrophage infiltration of the placenta, and these studies had shown mixed findings [18,19]. One explanation for such mixed results is the small number of cases used; another may be the lack of consideration of fetal sex as a variable. In our study, CV was seen at a higher frequency in placentas of female fetuses, but only in the setting of maternal obesity. The pathophysiology of this finding is unclear and warrants further investigation. Interestingly, we also found an increased prevalence of fetal thrombosis in female placentas, not previously reported; this is likely secondary to the increased prevalence of CV in the same placentas, as fetal thrombosis-associated changes, such as avascular villi, can occur in the setting of villous inflammation [16]. However, fetal thrombosis was also increased in female placentas of lean women, who did not have an increased rate of CV.

An additional unexpected finding was the higher prevalence of patchy villous edema in male placentas. This lesion has been associated with fetal vascular malperfusion, and was also seen more commonly in preterm placentas, as previously reported [33]. Its higher prevalence in male placentas may therefore be a reflection of a higher proportion of male placentas amongst the preterm deliveries.

It is known that CV can be associated with fetal growth restriction, particularly when the associated lesion of fetal thrombosis is also present in the placenta [13,15–17]. As previously reported, placentas with CV were less likely to be large for gestational age. This finding was also seen in obese women, where CV was less commonly seen in large placentas. This could suggest that, in the setting of an otherwise destined-to-be-large placenta, villitis serves to normalize placental size. Given this, and the finding of increased risk of CV in female placentas of obese mothers, we expected a decreased risk of fetal macrosomia in female babies born to obese women; however, we did not see such a decrease. It is possible that given the rates of macrosomia in this study, we may not be adequately powered to see such an effect. The presence of CV also did not impact the duration of pregnancy or placental efficiency. It is therefore unclear as to what degree these variables (CV, fetal growth, and gestational age at delivery) are connected by the same pathophysiologic mechanism. Nevertheless, the possibility that CV serves to normalize placental size and may potentially also normalize birthweight is intriguing.

The largest previous study of placental pathology among obese women had shown an association of maternal obesity with

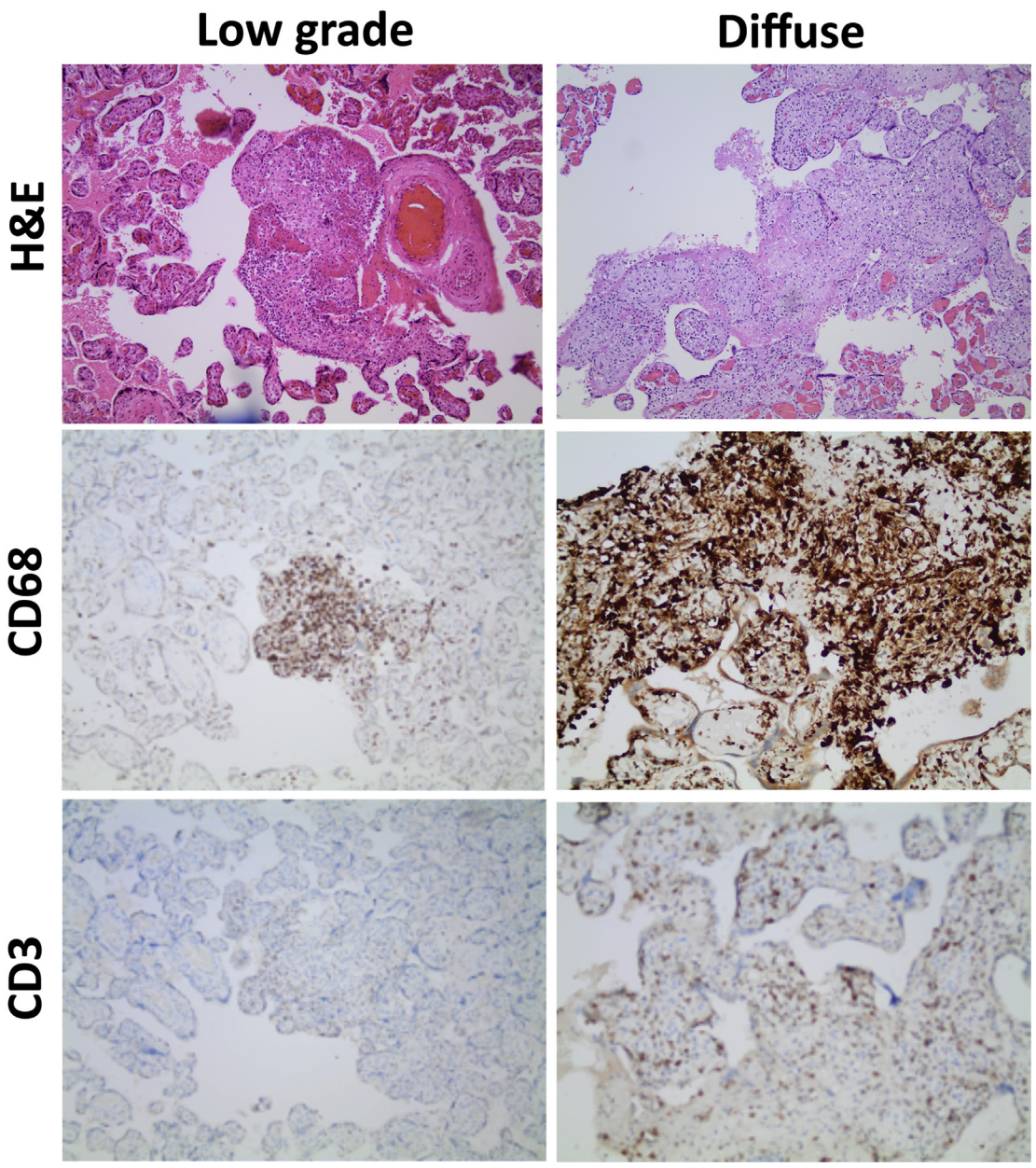

Fig. 2. Macrophage and T cell infiltration in chronic villitis. Examples at either end of the grade spectrum (low grade and diffuse) of chronic villitis are shown, with infiltration of macrophages (shown by immunohistochemistry for CD68) and T cells (shown by immunohistochemistry for CD3). Magnification of all panels: 100X.

maternal vascular lesions (decidual vasculopathy and atherosis), villous infarcts, fetal neutrophilic infiltration, and meconium staining of fetal membranes [11]. Among these four lesions, we only noted an association of maternal obesity with maternal vascular lesions; even there, multivariate analysis revealed these lesions to be related to maternal hypertensive disease. The different results are likely secondary to differences in the study populations. The above study used the Collaborative Perinatal Project data from a cohort of over 54,000 pregnancies delivering at 12 centers across the U.S., dating back to the 1960s. Also, the majority (59.3%) of the obese population is the CPP study was black [11], compared to our mostly Hispanic obese population.

The strengths of this study are the large sample size, which permits stratification of the data. Likewise the amount of clinical data available for each case was robust and allowed the analyses to be controlled for potential confounders such as diabetes and hypertension. The evaluation of the placentas was uniform and performed by an expert in placental evaluation who was blinded to most clinical information, except for gestational age and indication for placental examination. Nevertheless, several limitations exist. Women delivering before 35 weeks were excluded due to the relatively small number in our cohort (<15); for this reason, the rate of preterm delivery in relation to maternal obesity is small. Also, we had very high rates of diabetes and hypertension among our patient population, typical of those seen at an academic tertiary care center; while efforts were made to control for these variables, our findings may not be fully generalizable to all populations. Finally, our numbers were not large enough to allow for evaluation of pregestational vs. gestational maternal disease, including hypertension and diabetes. Therefore, future studies should include larger cohorts in order to be better able to address the effect of multiple covariates simultaneously.

Overall, our data underscore the importance of placental histopathology in evaluating effects of fetal sex and maternal obesity on pregnancy outcomes. This study was not designed to evaluate long-term outcomes, such as development of childhood obesity or diabetes. Future studies, including follow-up of the neonates in our series, could yield valuable information, specifically since it is now recognized that factors *in utero* can affect the health of the individual later in life. More research on the impact of maternal obesity, with consideration of placental pathology, is needed to address these questions.

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