WORLD JOURNAL OF PHARMACY AND PHARMACEUTICAL SCIENCES
SJIF Impact Factor 7.421
Volume 7, Issue 12, 40-68 Research Article ISSN 2278 – 4357

PLACENTAL PATHOLOGY IN WOMEN COMPLICATED BY GESTATIONAL DIABETES A THESIS

***Dr. Maha Radh Ismaiel and Dr. Sajwa Qais Mahmood**

Consultant Obstetrician & Gynaecologist Assistant Professor in Department of Obstetrics & Gynaecology College of Medicine, Al Mustansiriya University Al-Yarmouk Teaching Hospital.

Article Received on
30 September 2018,

Revised on 21 October 2018,
Accepted on 11 Nov. 2018,

DOI: 10.20959/wjpps201812-12699

***Corresponding Author**
Dr. Maha Radhi Ismaiel
Consultant Obstetrician & Gynaecologist Assistant Professor in Department of Obstetrics & Gynaecology College of Medicine, Al Mustansiriya University Al-Yarmouk Teaching Hospital.

ABSTRACT

Back Around: Diabetes mellitus is major complication of pregnancy regardless of whether it is pre-pregnancy (overt)or gestational in onset. In such pregnancies, the fetus is exposed to increased morbidity & mortality associated with hypoxic stress & various metabolic abnormalities. Placentae from diabetic pregnancies present morphological abnormalities independent of the level of glycemic control. One may hypothesise that diabetes results in impaired placental function, weight, circulation& histology, accounting for these phenomena. **Objectives:** The objective of this study is to investigate the histopathological effect of diabetes on the placenta in pregnancies complicated by gestational diabetes compared to uncomplicated pregnancies (control). **Study design:** A Case — Control study. ***Settings:*** The study involve 40 singleton pregnancies complicated by gestational diabetes who are attending Al Yarmouk Teaching Hospital & compared with group of 40 uncomplicated pregnancies (control) over a period of 12 months from the first of April 2008 to the end of March 2009 by studying samples taken from their placentae & send for histopathological study with regard to the following changes chorangiosis, ischemia, infarction, fetal vessel thrombosis, villous immaturity, presence fetal nucleated red blood cells, vascular villi and lymphohistocytotic. ***Results:*** The presence of degenerative lesions such as fibrinoid necrosis and vascular lesions like chorangiosis was apparent, mainly in the diabetes group. Villous immaturity and the presence of NFRBC as an indication of chronic fetal hypoxia were significantly increased in the placentas of women with diabetes compared with the

control group. Fetal/placental weight ratio was significantly lower in the diabetic group. ***Conclusion:*** Histological abnormalities were observed more frequently in the diabetic placentas compared to the controls. These findings support the hypothesis that impaired placental function is one of the main reasons for the increased frequency of fetal complications in diabetic pregnancies.

KEYWORDS: Placenta; pathology; gestational diabetes; pregnancy.

INTRODUCTION

Gestational Diabetes Mellitus

Diabetes in pregnancy encompasses a range of disease entities including gestational diabetes and overt diabetes mellitus. Nonpregnant diabetics are divided into two types based on the pathophysiology of their disease, whereas diabetes in pregnancy is usually divided into pregestational and gestational diabetes.

Pregesational diabetics include all patients with type 1 and type 2 diabetes diagnosed prior to pregnancy. Gestational diabetics are those diagnosed in pregnancy. Because of lack of screening in many nonpregnant women, this latter group may occasionally contain undiagnosed pregestational diabetic women.[1]

Definition

Gestational Diabetes Mellitus is defined as carbohydrate intolerance that first recognized during pregnancy and in most cases resolves after pregnancy. In the broader sense of this definition, therefore, women with previously unrecognized (pregestational diabetes mellitus) type 2 diabetes mellitus and newly presenting type 1 diabetes mellitus during pregnancy ought to be considered as gestational diabetes mellitus.[2]

Epidemiology

The incidence of gestational diabetes ranges from 2%-5% of pregnant women depending on various demographic populations, including age, ethnic group, family history of diabetes, previous infant weighing >4000 g and previous still birth. As many as 50% of women with gestational diabetes will go on to develop overt (type 2) diabetes mellitus.[1,2]

Screening

Internationally, there remain no agreement on how and whom to screen for gestational diabetes mellitus.

The decision to screen for GDM universally or to only target those women with risk factors has also not been resolved.[3]

The classical risk factors which increase the incidence for developing GDM which should be screened for:-

1. A previous history of GDM, impaired glucose tolerance test or impaired fasting glycemia.
2. A family history in first degree relative of diabetes.
3. Advancing maternal age especially > 35 years.
4. Body mass index >35 or weight >85 kg).
5. A previous pregnancy with macrosomic baby (weighing above 95th centile for gestational age, race, sex or >4000 g).
6. Glycosuria or obesity first trimester.
7. Glycosuria on two occasions in either second or third trimester.
8. Polyhydromnias in current pregnancy.
9. Previous unexplained still birth.
10. Recurrent miscarriages.
11. Ethnic groups African-American, Asian, Hispanics and Afro-Caribbean's.

In addition to this, statistics showed a double risk of GDM in smokers. Polycystic ovarian syndrome is also a risk factor. Some studies have looked at more potential risk factors, such as shortstature. About 40%-60% of women with GDM have no demonstrable risk factor, for this reason many advocate to screen all women. Typically women with GDM may exhibit no symptoms, but some women may demonstrate increase thirst, fatigue, nausea and vomiting, bladder infection, yeast infection and blurred vision these symptoms are another reason for universal screening.[3,4]

The screening test is performed between 24 — 28 weeks of gestation, some authorities advocate it at booking visit, others at 28 weeks and others suggest that blood glucose level should be checked whenever there is >+ glycosuria on dipstick test.[3,4]

Diagnosis

The 75 gram OGTT is advocated by the World Health Organization in the "one-tiered" approach but is less well validated than the 100 gram test. The World Health Organization uses cutoffs of fasting plasma glucose > 126 mg/dl (7.0 mmo1/1) or 2 hour post load

glucose > 140 mg/dl (7.8 mmo1/1). The American Diabetes Association, in contrast, requires that at least 2 of the 3 venous plasma glucose levels be attained or exceeded to diagnose gestational diabetes as shown below. A large observational study found no difference in adverse pregnancy outcomes when gestational diabetes was diagnosed using the American Diabetes Association or World Health Organization criteria.

Table 6: Diagnosis of Gestational Diabetes.

75 g glucose load test	American Diabetes Association	World Health Organization
Fasting glucose	>95 mg/dl (5.3 mmo1/1)	>126 mg/di (7.0 mmol/l)
1 hour glucose	>180 mg/d1 (10.0 mmo1/1)	----------
2 hour glucose	>155 mg/di (8.6 mmo1/1)	>140 mg/di (7.8 mmol/l)

When using an OGTT, the criteria for the diagnosis of diabetes is a 2 h glucose >200 mg/di (11.1 mmo1/1) after a 75 gram oral glucose load (American Diabetes Association and World Health Organization criteria). The 75 gram glucose load should be administered when the patient has ingested at least 150 grams of carbohydrate for the 3 days preceding the test and after an overnight fast.[5,6]

	2-h Plasma Glucose (after 75 gram Glucose Load)
Normal glucose tolerance	<140 mg/dl (7.8 mmol/l)
Impaired glucose ("pre-diabetes")	140-199 mg/dl (7.8-11.1 mmol/l)
Diabetes mellitus	>200 mg/dl (11.1 mmo1/1)

Complications of Gestational Diabetes

Despite improvement in pregnancy outcome, women with both gestational and pregestational diabetes at greater risk for a number of pregnancies related complications. These include preterm labour, Other infectious morbidities such as Candida vulvovaginitis, urinary tract infection, respiratory tract infection, and puerperal pelvic infection. Polyhydromnias occurs as a result of fetal hyperglycemia and the amniotic fluid index parallels the amniotic fluid glucose level among women with diabetes. Ketoacidosis and preeclampsia are other complications of GDM, the incidence of preeclampsia and nonproteinuric hypertension is 6% -20% of pregnancies complicated by GDM.[7,8,9]

Further more, women with pregestational diabetes are at risk for the acute complications of diabetes because of the metabolic alterations associated with pregnancy, as well as the effects of strict glycemic control. Current data would seem to indicate that pregnancy is an independent risk factor for diabetic retinopathy.[7]

In women with GDM is at increased incidence of fetal macrosomia 20% -30%, as they give birth to infants heavier than 4000g. It has been estimated that about 4% of women with GDM will give birth to infant weighing over 4500g. Macrosomia cause increase risk of cesarean section where risk increase up to 20%, shoulder dystocia and instrumental delivery.[8]

Maternal complications include miscarriages, sudden intra uterine death, still birth which increase 10 folds in GDM.[7]

The offspring of women with GDM at increase risk for number of complications, which include congenital malformations that occurs at about 2-3 folds over that of normal pregnancy. The most frequent types of malformations involve centralnervous system, cardiovascular system, gastrointestinal system and skeletal system, with cardiac malformations being the most frequent.[7]

Metabolic abnormalities affecting infants of GDM women are hypoglycemia, that affecting about 25%-40% where plasma glucose level below 35mg/d1 in term infants and 25mg/d1 in the preterm infants, and it develop in the first few hours of life, particularly in cases of poor glycemic control. Hypocalcaemia that generally occurs in association with hypophosphotemia and occasionally with hypomagnesaemia. Neonatal hypocalcaemia occurs when calcium level below 7mg/dl where serum calcium levels are lowest on the second day or third day of life.[7]

Polycythemia occurs when venous hematocrit exceeds 65% and it believed to occur as result of chronic intrauterine hypoxia, which leads to an increase in erythropoietin and results in an increase in red blood cell production. Neonates born to women with GDM also may have a higher incidence of hyperbilirubinemia as compared with nondiabetics. It develops in approximately 20%-25% of cases.[7] Lastly, there are long term consequences of diabetic pregnancy. These include obesity, neuropsychological defects, and an increased tendency to develop overt diabetes in about 1-3% later in life.[7,9]

Prognosis

Gestational diabetes generally resolves once the baby is born. Based on different studies, the chances of developing GDM in a second pregnancy are between 30 and 84%, depending

on ethnic background. A second pregnancy within 1 year of the previous pregnancy has a high rate of recurrence.

Women diagnosed with gestational diabetes have an increased risk of developing diabetes mellitus in the future. The risk is highest in women whoneeded insulin treatment, women with more than two previous pregnancies complicated by GDM, and women who were obese (in order of importance). Women requiring insulin to manage gestational diabetes have a 50% risk of developing diabetes within the next five years. Depending on the population studied, the diagnostic criteria and the length of follow-up, the risk can vary enormously. The risk appears to be highest in the first 5 years, reaching a plateau thereafter. Another study found a risk of diabetes after GDM of more than 25% after 15 years. In populations with a low risk for type 2 diabetes, in lean subjects and in patients with auto-antibodies, there is a higher rate of women developing type 1 diabetes.

Children of women with GDM have an increased risk for childhood and adult obesity and an increased risk of glucose intolerance and type 2 diabetes later in life. This risk relates to increased maternal glucose values. It is currently unclear how much genetic susceptibility and environmental factors each contribute to this risk, and if treatment of GDM can influence this outcome.[7,8,9]

White's classification of GD.[10]

Class	Description	Therapy
A 1	GestationalDiabetes; glucose intolerance developing during pregnancy; fasting blood glucose normal	Diet only
A2	Gestational Diabetes with fasting plasma glucose>105mg; or 2-h postprandial glucose>120mg	Diet and insulin
B	Overt Diabetes developing after age 20 or duration <10	Diet and insulin
C	Overt Diabetes developing before age 20 or duration >10 years	Diet and insulin
D	Overt Diabetes developing between age 10-19, or duration 10-19, or background retinopathy	Diet and insulin
F	Overt Diabetes at any age or duration with nephrapathy	Diet and insulin
R	Overt Diabetes at any age or duration with proliferative retinopathy	Diet and insulin
H	Overt Diabetes at any age or duration with arteriosclerotic heart disease	Diet and insulin

Placental Development

After conception in the outer third of fallopian tube, rapid cellular division occurs in the fertilized ovum. By the time of implantation into endometrium, the morula (ball of cells) has

changed to a blastocyst, with the beginning of the major subdivision into embryo and placenta. At this stage, a fundamental change occurs in cell mass. A fluid space appears and a division occurs into an inner cell mass (embryoblast), which will form embryo, and an outer cell mass (trophoblast), which will form placenta. This is the blastocyst and it is in this form the embedding takes place at about 6-8 days after fertilization.[11]

At the eight day of development, the trophoblast has differentiated into two layers:

1. An inner layer of mononucleated cells, the cytotrophoblast.
2. An outer multinucleated zone with out distinct cell boundaries, the syncytiotrophoblast.

Mitotic figures are found in the cytotrophoblast but not in the syncytiotrophoblast. Thus, cells in the cytotrophoblast divide and migrate into the syncytiotrophoblast, where they fuse and lose their individual cell membranes.[12]

At the implantation pole, the syncytiotrophoblast becomes very thick and extends finger-like branches deep into the endometrium. As the blastocyst sinks deeper and deeper into the endometrium, as more and more of the outer trophoblast layer comes in contact with the endometrium, more and more of the trophoblast shell is transformed into syncytiotrophoblast. Eventually, when the blastocyst is entirely covered by endometrium, the entire outer trophoblast shell is transformed into syncytiotrophoblast. Later, when the blastocyst grows bigger, up and out of the endometrium, the syncytiotrophoblast at the anti-implantation pole loses contact with the endometrium and atrophies.[3]

By the 11^{th} to 12^{th} day of development, the trophoblast is characterized by lacunar spaces in the syncytium that form an intercommunicating net work. This network is particularly evident at embryonic pole, at the abembryonic pole, the trophoblast still consist mainly of cytotrophoblastic cells.

The syncytiotrophoblast penetrate deeper into stroma and erode the endothelial lining of the maternal capillaries, which are congested and dilated, are known as sinusoids.

The syncytial lacunae become continuous with the sinusoids and maternal blood enters the lacunar system. As the trophoblast continues to erode more sinusoids, maternal blood begins to flow through the trophoblastic system, establishing the uteroplacental circulation.

By day 13th, the trophoblast characterized by villous structure, where the cells of the cytotrophoblast proliferate locally and penetrate into the syncytiotrophoblast, forming cellular columns surrounded by syncytium. Cellular columns with the syncytial covering are known as primary villi.

During further development, mesodermal cells penetrate the core of the primary villi and grow toward the deciduas. The newly formed structure is known as secondary villi.

By the end of the third week, mesodermal cells in the core of the villous begin to differentiate into blood cells and small blood vessels, forming the villous capillary system. The villous system now is known as a tertiary villous or definitive placental villous. Capillaries in tertiary villi make contact with capillaries that developing in mesoderm of the chorionic plate and in the connecting stalk. These vessels, in turn, establish contact with the intra-embryonic circulatory system connecting the placenta and the embryo. Hence, when the heart begins to beat in the fourth week of development, the villous system is already to supply the embryo proper with essential nutrient and oxygen.

Mean while, cytotrophoblastic cells in the villi penetrate progressively in to the overlying syncytium until they reach the maternal endometrium. Here they establish contact with similar extension of neighboring villous stems, forming a thin outer cytotrophoblastic shell. This shell gradually surrounds the trophoblast entirely and attaches the chorionic sac firmly to the maternal endometrial tissue and villi will extend from the chorionic plate to the decidua basalis, they are called stem villi or anchoring villi. Those villi that branching from the sides of stem villi are free villi (terminal villi) through which nutrient and other factors will occur.[13]

There are 2 waves for development of uteroplacental circulation, the first wave occurs in the first trimester and consist of invasion and modification of the spiral arteries of the decidua, reaching its border with the myometrium.

the second wave occurs between 12-16 weeks of gestation which involves invasion of the intramyometrial part of the spiral arteries, converting narrow — lumen muscular spiral arteries into dilated, low resistance uteroplacental vessels.[11]

In the first trimester, growth of the placenta is more rapid than the fetus, by the 17th weeks of gestation; placenta and fetal weights are approximately equal. At term the placental weight may be roughly one Sixth that of the fetal weight.[13]

Full Term Placenta

At full term, the placenta is discoid with diameter of 15 — 25 cm, and its 3 cm in thickness, and weighs 500 — 600 g and 500 ml in volume. After birth, when the placenta is viewed from maternal side, consist of 15 — 20 large lobules or called cotyledons, that covered by thin layer of decidua basalis, grooves between the cotyledons are formed by decidual septa.

The fetal surface of the placenta is covered entirely by the chorionic plate. A number of large vessels, the chorionic vessels, converge toward the umbilical cord. The chorion, in turn, is covered by the amnion. Attachment of the umbilical cord is usually eccentric and occasionally even marginal.[13]

Circulation of the Placenta

Cotyledons receive there blood through 80 — 100 spiral arteries and pierce the decidual plate and enter the intervillous spaces, which contain approximately 150 ml of blood which is replenished about 3 or 4 times per minute. The lumen of the spiral artery is narrow, so blood pressure in the intervillous spaces is high. This pressure forces the blood deep into the intervillous spaces and bathes the numerous small villi of the villous tree in oxygenated blood. As the pressure decreases, blood flows back from the chorionic plate toward the deciduas, where it enters the endometrial veins. Hence, blood from the intervillous lakes drains back into the maternal circulation through endometrial veins.[12]

The effect of Diabetes on the Placenta

Pathological examination of the placenta is probably the most underutilized pathologic assessment of any human tissue. The placenta provides a wealth of information, retrospectively about the fetus and prospectively regarding the infant.[14]

The histopathology of the placenta can answer specific questions about in utero insults, give insight into management of subsequent pregnancies, and provide an assessment of the newborn risk. Placental pathology has been a key litigious informant in inferring timing of insult.[15]

Diabetes affects the placenta in several ways macroscopically, microscopically and affects its weight.

Classical morphologic investigations of placental structure in diabetic pregnancies have shown a varying degree of changes in the syncytiotrophoblast, cytotrophoblast, trophoblastic basement membrane, and fetal vessels. Some authors found no major differences in microscopic villous changes, in particular in women with good glycaemic control.

Overall, most authors reported a relative placental immaturity, due probably to a high proportion of villi with stromal oedema and focal fibrinoid necrosis.[16]

Diabetes is a state of chronic oxidative stress, and the responses of the fetal—placental vasculature of diabetic placentas to vasoconstrictor and vasodilator agents are significantly attenuated when compared to those in normal control placentas. Gestational diabetes produces an enhancement of the observed relaxation caused by hypoxia and the contraction produced by reoxygenation or hydrogen peroxide. These data suggest that in patients with vasculopathy, the pathophysiology of the placental changes is secondary to a phenomenon of hypoxia—reoxygenation similar to that found in preeclampsia. There have been few investigations of the utero-placental circulation in diabetic pregnancies.[17]

The placental weight and neonatal weight were increased which will result in increased perinatal jeopardy the rate of operative delivery was higher in diabetic mothers.

The placental weight and neonatal weight were increased resulting in increase rate of operative delivery.[18]

GDM affect feto- placental ratio, as it causes decrease of this ratio if it compared to uncomplicated pregnancy.[19,20]

Placenta from diabetic patients showed immaturity of the villi, hypertrophy of the capillaries and thickening of the basement membrane of the tropho- blastic villi and the amniotic membrane. Focal fibrinoid necrosis, an increase in the number of Hofbauer cells and dilatation of villi capillaries were also commonly observed in placentae from diabetic mothers, and the normal cuboidal cells lining the amniotic membrane tended to become tall columnar with distally located nuclei.[20,21]

Placentas from diabetic pregnancies present similar morphological abnormalities independent of the level of glycemic control. The incidence of increased in these pregnancies despite the quality of blood glucose control. One may hypothesize that diabetes results in impaired placental function, weight, circulation and histology, accounting for these phenomena.[22,23]

Other placental abnormalities associated with diabetes are chorangiosis and increased presence of nucleated fetal red blood cells as a sign of chronic fetal hypoxia.[24]

GDM causes hypoxia that affect endothelial proliferation and vessel maturation. In preplacental hypoxia, the mother, placenta and fetus are hypoxic, there is excessive branching angiogenesis and growth of capillaries and villi is impoverished.[25]

Maternal diabetes is associated with extensive placental abnormalities such as placentomegaly, infarcts, dysmaturity, angiopathies, abnormalities of the basement membrane and abnormalities of placental villi such as hydrops, fibrosis and increased glycogen content. The most consistent of these features is the enlargement of the placenta, which is usually associated with fetal macrosomia.[26]

An increased placental weight has been well documented in pregnancies complicated with GDM, and the placental size is related to the duration and control of diabetes mellitus. However, in these reports the study subjects had either preexisting diabetes or GDM and required insulin, and there is limited data on the placental weight and ratio in mothers with mild gestational glucose intolerance on diet control only.[27,28]

In pregnancies complicated by diabetes, IUGR, and preeclampsia in association with increased trophoblast apoptosis and deportation and altered placental vascular reactivity such as villous immaturity and hydropic villi.[29]

The presence of villous immaturity, chorangiosis and ischemia were significantly increased in the placentas of women with GDM.

The placenta that shows abnormalities in angiogenesis and maturation may lead to fetal hypoxia and compromise.[30,31]

Vascular surface area available for absorption of oxygen from maternal blood in the intervillous. The occurrence of capillary hyperplasia in the chorionic villi is a pathological finding indicative of long-standing and significant fetal hypoxia. It is a compensatory

mechanism for increasing oxygen delivery to a hypoxic fetus by increasing the villous space.[32]

In placentas of women with GDM and fetal macrosomia there was a significant hickening of basal membranes of the trophoblast, and structural abnormalities in perivascular space (stromal oedema) with proliferation of collagen in terminal villi. Intensity of these changes was related to the degree of hyperglycemia and affected fetal and neonatal wellbeing.[33]

The cytotrophoblast is unusually prominent and the trophoblastic basement membranes shows irregular thickening. Sometimes deposits of fibrinoid material can be seen between the trophoblast and the basement membrane.[34]

In pregnancies complicated by fetal hypoxia, such as GDM, deferent categories of hypoxia can be identified based on observed patterns of villous development and companion changes in growth factors associated with villous macrophages, trophoblast and capillary endothelium.[35]

Chorangiosis indicates abnormalities in fetal micro-circulation with resultant placental fibrosis and infarction. All of these lead to chronic fetal hypoxia.[36]

Aim of the study

The aim of this study is to investigate pathological difference of placenta in pregnancies complicated by gestational diabetes compared to uncomplicated pregnancies (control).

Patient and Methods

Prospective case — control study was conducted on 80 pregnant women attending AL Yarmouk Teaching Hospital over a period of 12 months from the first of April 2008 to the end of March 2009.

The study included Forty singleton pregnancies complicated by gestational diabetes were recruited at the time of diagnosis between 24 and 32 weeks of gestation and compared to 40 consecutive normal pregnancies (control group) matched for their age, gestational age and parity.

GDM was diagnosed by doing a 75-g oral glucose tolerance test (OGTT) at 24-32 weeks according to WHO criteria for diagnosis of GDM.

Inclusion Criteria

- All were diagnosed with GDM by OGTT at 24 -28 weeks of gestation.
- Singleton pregnancy.
- Gestational age ranging from 37 — 41 weeks of gestation.

Exclusion criteria

- Presence of Multiple pregnancies.
- Presence of Hypertension, andPreeclampsia.
- Presence of chorioamnionitis.
- Presence of Intrauterine growth retardation.
- Presence of fetal congenital anomalies.
- Smoking.

Gestational age was assessed using the date of the last menstrual period in case of a regular menstrual cycle and was confirmed by first trimester ultrasound.

In case of an irregular menstrual cycle gestational age was determined by first trimester ultrasound. At birth, whether vaginal delivery or by caesarean section, and after clamping of the cord, birth weights were collected.

The placentas were rinsed in water and left to drain of blood for at least half hour, then weighed without the umbilical cord and membranes.

We calculated the ratio (placental ratio) between infant birth weight and placental weight, corrected for gestational age. Gross examination of the placenta was done, looking for ischemia and infarction.

Samples taken from the placenta about 5 cm in diameter, were embedded in formaldehyde and send for Histopathological examination A pathologist blinded to all clinical data except gestational age (in order to assess the villous maturation) reviewed all histological samples.

All samples were embedded in paraffin and stained with hematoxylin and eosin, then it sliced to samples about lmm thickness. The histological assessment was carried out with regard to the following aspects: fetal vessel thrombosis, villous immaturity, chorangiosis, presence of nucleated fetal red blood cells (NFRBC), mural thrombosis,

ischemia, ischemic villitis, infarction, presence of hydropic or avascular villi, massive perivillous fibrin deposition and villous fibrinoid necrosis.

The following are histological definitions;

- Lymphohistiocytic villitis was diagnosed by the presence of numerous lymphocytes and macrophages in the villous stroma. The extent of lymphohistiocytic villitis was graded according to. Grade 1 and 2 were considered as moderate villitis and grade 3 and 4 as severe villitis.
- Ischemia.
- Infarction, assessed at gross examination, was present and classified as moderate when <10 per cent of the placental volume and severe when >10 per cent of the placental volume was infracted.
- The presence of nucleated fetal red blood cells (nRBCs), as a sign of chronic fetal hypoxia, was defined as moderate when present in the umbilical cord only and severe when also present in the peripheral villi.
- Villous fibrinoid necrosis, a condition where villous stroma is replaced by fibrinoid, was defined as moderate if only a few foci were seen in a small number of low power (10 x) fields and severe when some villi with fibrinoid necrosis were found in most low power fields examined.
- Degree of villous immaturity was defined as moderate when there was a relatively decreased formation of terminal villi and a relatively increased presence of mature intermediate villi in relation to gestational age in each slide. Villous immaturity was defined as severe when there was a relatively decreased formation of terminal villi and a relatively increased presence of immature and mature intermediate villi in each slide.
- Chorangiosis, i.e. vascular hyperplasia of the chorionic villi, was defined as the occurrence of 10 or more villi with 10 or more capillaries in 10 or low power microscopic field.
- Hydropic villi were diagnosed when large terminal villi were present with edematous fluid and with an increase of villous macrophages and with an artifactual separation of the trophoblast lining from the underlying stroma.
- Fetal vessel thrombosis was diagnosed when a large fetal stem villous vessel was partially or completely occluded by a thrombus.

Statistical Analysis

Results expressed in number/percent with odds ratios, 95% confidence intervals and P value calculated for each variable using Pearson chi-square (X^2) test and t-test to determine the relative importance of various variables. P value < 0.05 was considered as statistically significant, and P value < 0.01 was considered to be highly significant. The data were analyzed epidemiologically using SSPS, version 16.0 for windows.

RESULTS

No differences existed between the groups regarding maternal age and parity. The patients' age ranged 24-40 years with mean age 30.63±4.37 years and the parity ranged 0-6 with mean 2.30±1.59, as shown in table 1 and table 2.

Table 1: The age distribution.

Age groups (years)	Cases		Controls	
	No	%	No	%
<20				-
20--24	1	2.5	1	2.5
25--30	16	40.0	16	40.0
30--34	15	37.5	15	37.5
=>35	8	20.0	8	20.0
Mean±SD (Range)	30.63±4.37 (24-40)		30.62±4.37 (24-40)	

Table 2: The parity distribution.

Parity	Cases		Controls	
	No	%	No	%
Para 0		10.0	4	10.0
Para 1	9	22.5	9	22.5
Para 2-3	19	47.5	19	47.5
=>Para 4	8	20.0	8	20.0
Parity	Cases	Controls	Cases	Controls
		No		No
Mean±SD	2.30±1.59		2.30±1.59	
(Range)	(0-6)		(0-6)	

Regarding table 3: the gestational age was matched in both groups. In GDNI group the gestational age ranged 37 — 41 with mean 38.67±1.23, while in control group the range was 37 — 41 with mean 38.62 ±1.23.

Table 3: The gestational age (weeks) distribution.

Gestational age (weeks)	Cases		Controls	
	No	%	No	%
37	10	20.0	8	20.0
38	9	27.5	11	27.5

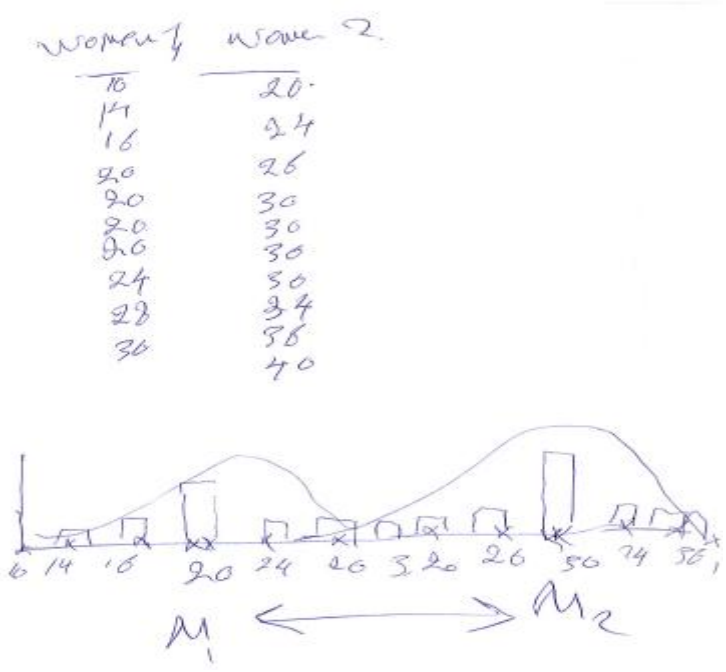
39	10	25.0	10	25.0
40	8	20.0	8	20M
41	3	7.5	3	7.5
Mean±SD (Range)	38.62±1.23 (37-41)		38.67±1.23 (37-41)	

Fetal weight differs between control and GDM fetuses. The mean of fetal weight in control was 3556.25±310.02, while the mean fetal weight in GDM was 3831.25±339.15 as shown in table 4.

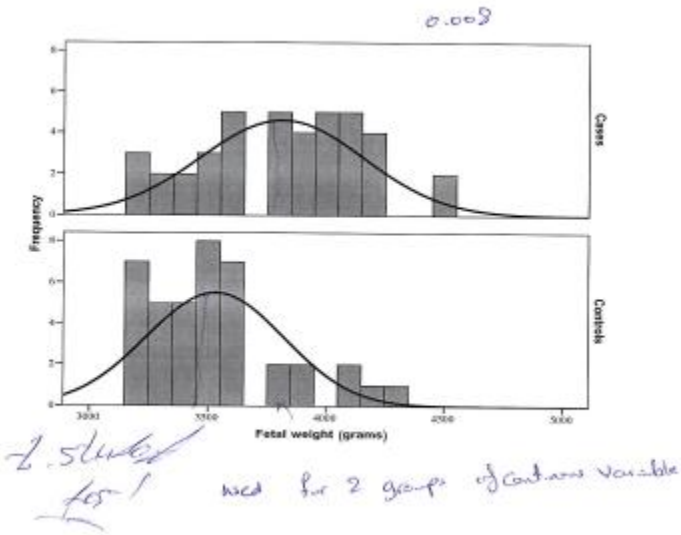
Table 4: The fetal weight (grams) distribution.

Fetal weight (grams)	Cases		Controls	
	No	%	No	%
3000--	3	7.5	7	17.5
3250--	4	10.0	10	25.0
3500--	8	20.0	15	37.5
3750--	9	22.5	4	10.0
4000--	14	35.0	3	7.5
4250--	2	5.0	1	2.5
Mean±SD (Range)	3831.25±339.15 (3200-4500)		3556.25±310.02 (3200-4300)	

P=0.008*



This graph shows the distribution of fetal weight in both groups. In GDM group the curve shifted to the left because of heavier fetal weight, while in control group the curve shifted to the right.



Graph 3: fetal weight distribution.

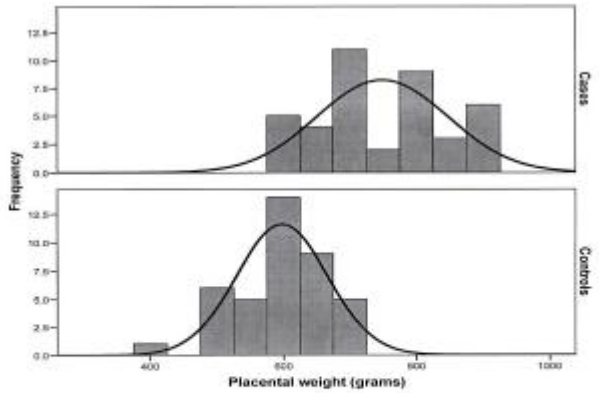
The placentae of GDM were heavier than the placetae of uncomplicated pregnancy. The placental weight of GDM ranged 600-900 with mean 748.75 ±97.06 g, while the placental weight of uncomplicated pregnancy ranged 400-700 g with mean 597.50±68.83g.

Table 5: The placental weight (grams) distribution.

Placental weight (grams)	Cases		Controls	
	No	%	No	%
400--	-		1	2.5
500--		-	11	27.5
600--	9	22.5	23	57.5
700--	13	32.5	5	12.5
800--	12	30.0	-	-
900--	6	15.0	-	-
Mean±SD (Range)	748.75±97.06 (600-900)		597.50±68.83 (400-700)	

P=0.0001*

This graph shows distribution of the placental weight. The placental weight in cases shifted to the right as it's heavier than the placentae of control, in control group it shifted to the left.



Graph 1: Placental weight distribution.

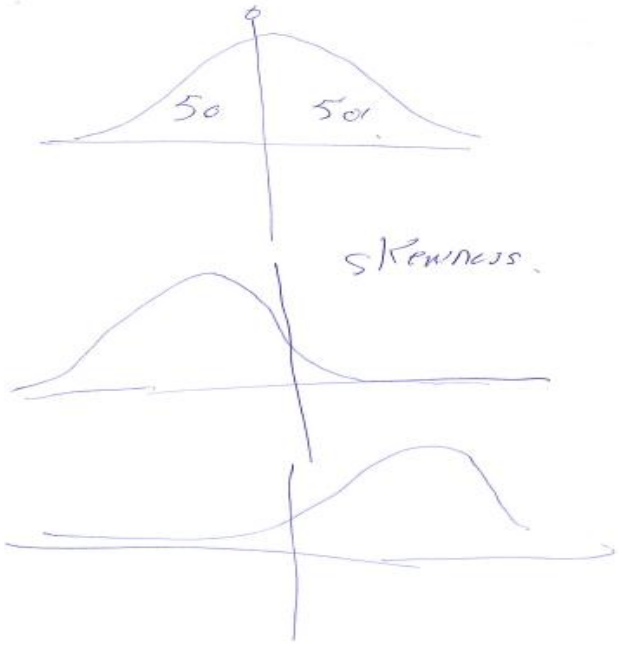
Graph 1: Placental weight distribution.

As result of higher placental weights, the feto/placental weight ratios were lower in pregnancies complicated by GDM compared to uncomplicated pregnancies.

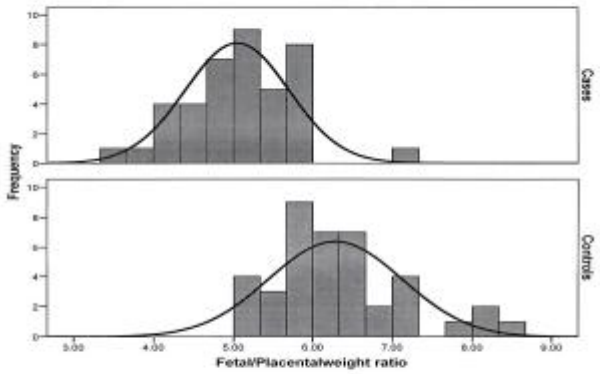
Table 6: The fetal/placental weight ratio distribution.

Fetal/Placental weight ratio	Cases		Controls	
	No	%	**No**	**%**
	2	5.0		
		12.5		
	10	25.0	-	-
5.00--	12	30.0	7	17.5
5.50--	10	25.0	9	22.5
6.00--	-	-	13	**32.5**
		-	3	**7.5**
7.00--	1	2.5	4	**10.0**
7.50--				2.5
8.00--				7.5
Mean±SD (Range)	5.05±0.66 (3.56-7.00)		6.28±0.83 (5.14-8.50)	

P=0,0001*



This graph shows feto-placental ratio; the ratio was shifted to the left in cases because of lower feto-placental ratio, in control group the ratio shifted to the right.



Graph 2: feto-placental ratio.

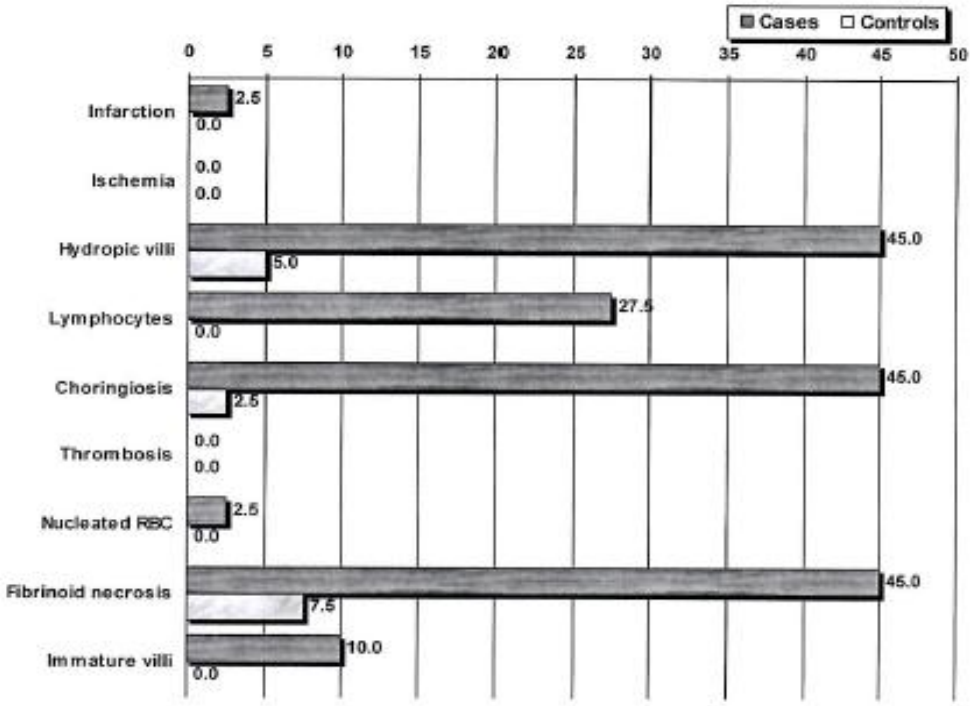
Table 7 shows histopathological findings that were seen in both groups. The presence of fibrinoid necrosis, hydropic villi and chorangiosis were mainly seen in GDM group. The presence of infarction, lymphocytes, nucleated RBC and immature villi were only seen in the placentae of GDM group. There were no cases of ischemia and thrombosis found in both groups.

Table 7: The histopathological findings.

Placental weight (grams)	Cases		Controls		P value
	No	%	No	%	
InfarctionPositive	1	2.5	-	-	-
Negative	39	97.5	40	100	
Ischemia Positive	-	-	-	-	-
Ne ative	40	100—	40	100	
Hydro.ic villiPositive	18	45.0	2	5	0.0001*
Negative	22	55.0	39	95	
LymphocytesPositive	I11	27.5	-	-	-
Negative	29	72.5	40	100	
ChorangiosisPositive	18r	45.0	1	2.5	0.0001*
Negative	22	55.0	39	97.5	
ThrombosisPositive	-	-	-	-	-
Negative	40	100	40	100	
Nucleated RBCPositive	1	2.5	-	-	-
Negative	39	97.5	40	100	
Fibrinoid necrosis Positive	18	45.0	3	7.5	0.0001*
Negative	22	55.0	39	92.5	
Immature villiPositive	4	10.0	-	-	-
I Negative	i36	90.0	t40	,100	

*Significant at 0.05 level of significance using Pearson chi-squared test.

This graph shows the distribution of histopathological findings in both groups.



Graph 3: Histopathological findings,

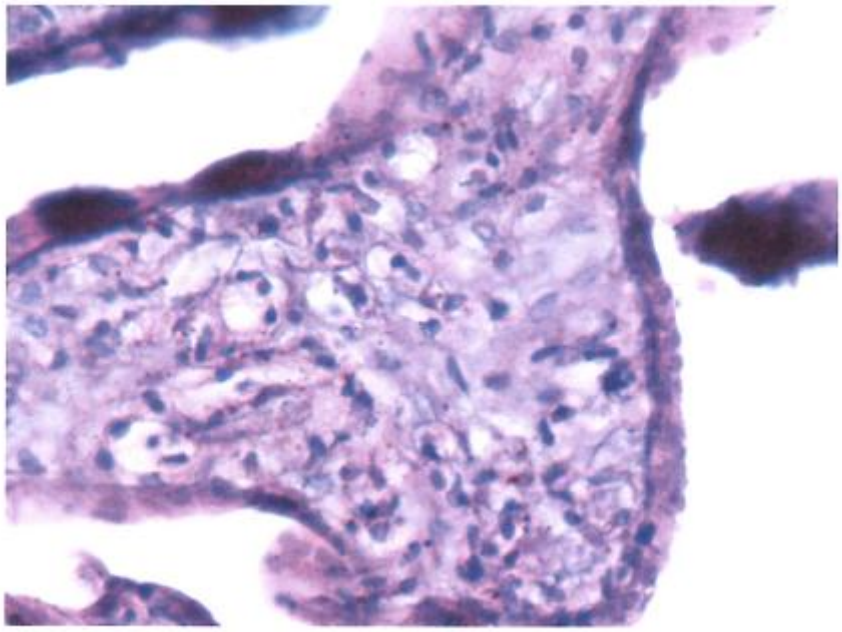


figure (1) shows lymphohistocytes

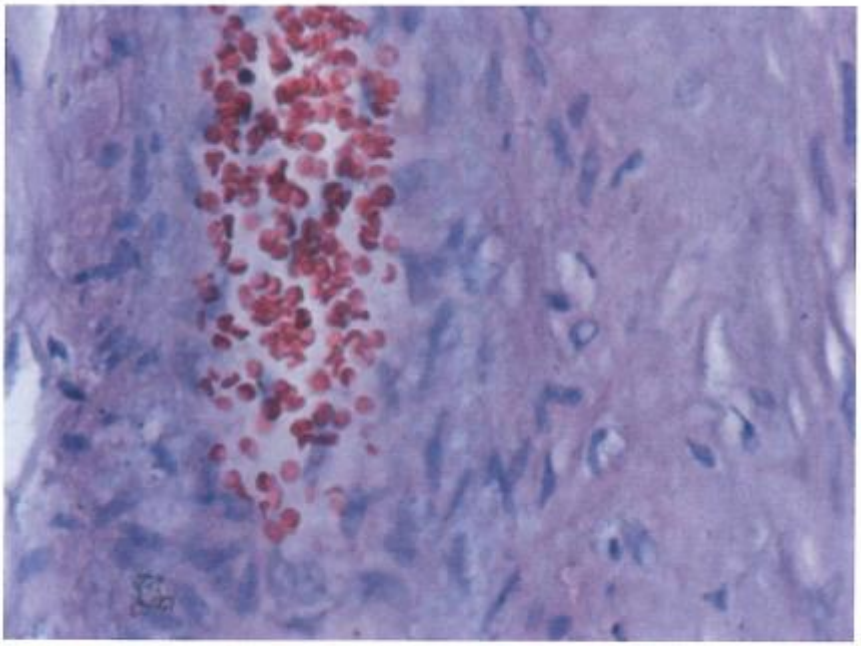


figure (2) shows NRBC

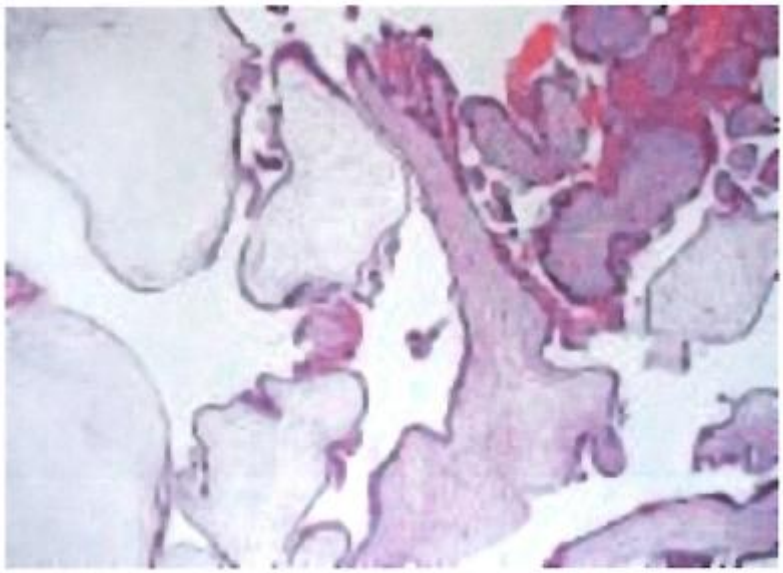


figure (3) shows hydropic villi

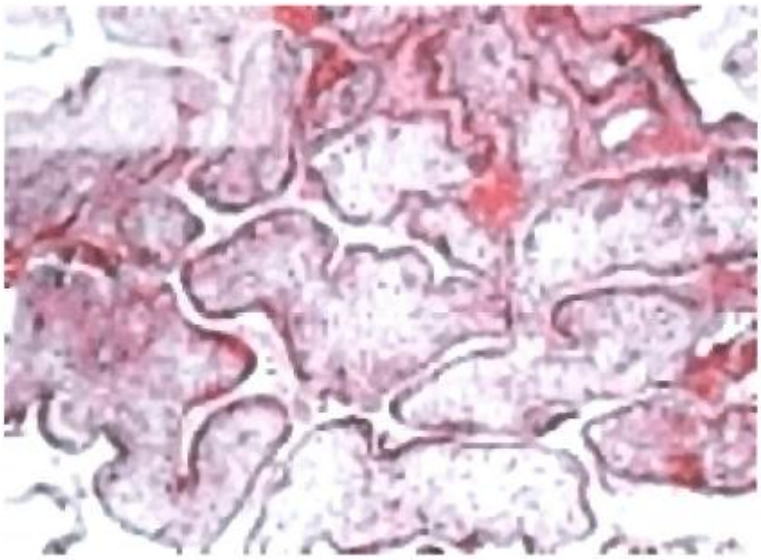


figure (4) shows immature villi

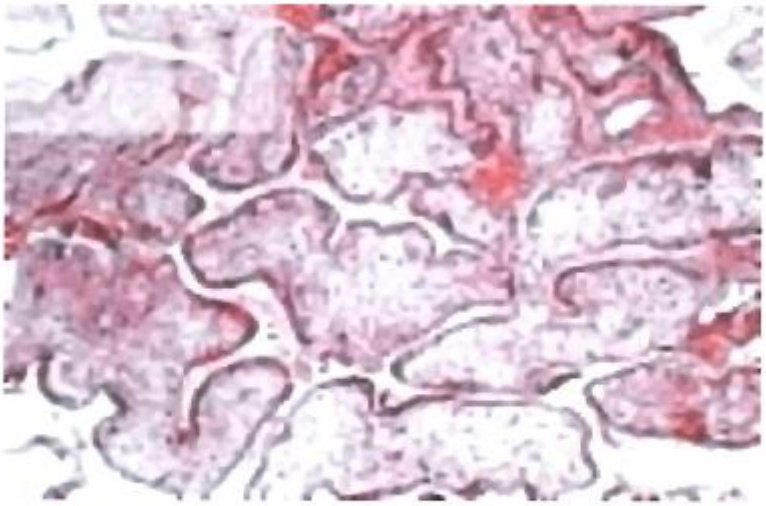


Figure (7) Shows Placental infarction

DISCUSSION

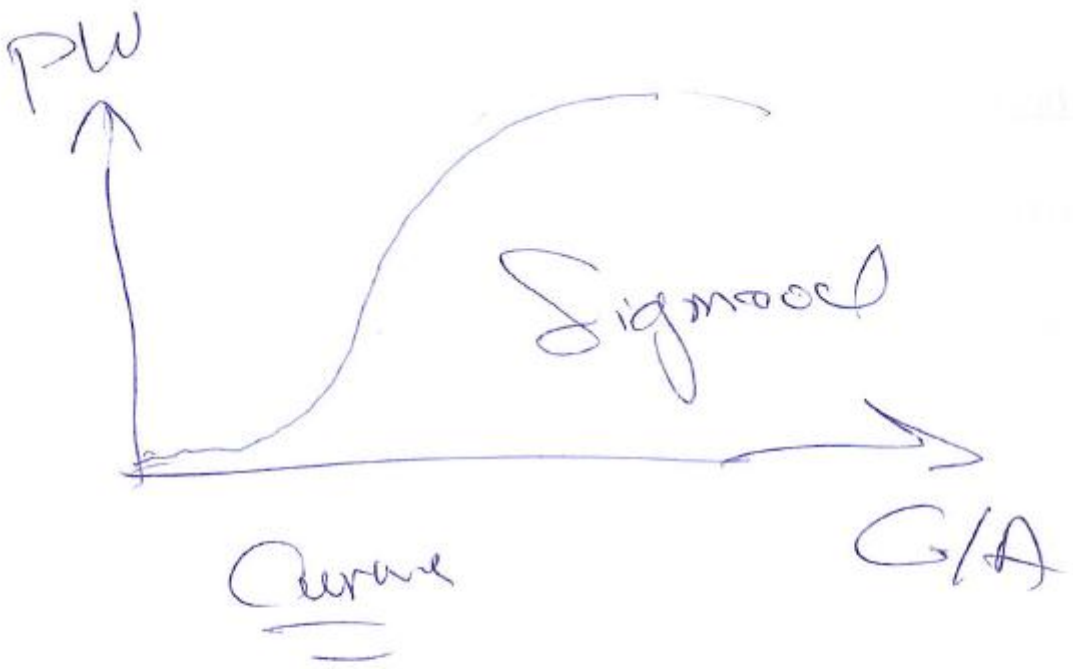
G.D.M considered one of the factors that make pregnancy at high risk, as maternal, fetal or neonatal complications are increased in diabetic pregnancies. Maternal hyperglycemia results in fetal hyperglycemia that stimulates fetal insulin over production, which causes fetal macrosomia, various congenital anomalies and fetal hypoxia. The placenta act as barrier that transport nutrients and oxygen, which are necessary for fetal development and wellbeing. It's believed that impaired placental function (in terms of abnormal weight or histology) can demonstrates the duration, severity or the onset of various conditions affecting the mother or the fetus.[24,25,32]

Our study included 80 singleton pregnancies, 40 complicated by GDM and 40 with uncomplicated pregnancies. Both groups were matched regarding age, parity and gestational age.

Fetal weight in GDM group was higher than that of the control group (which is explained by fetal insulin over production in GDM) as was found in our study with a p value < 0.008 which is statistically significant.

These results were consistent with the results of Daskalakis et al, with a p value < 0.005.[24]

Our study demonstrates that the placental weight increased in GDM comparable to the placental weight of control group, this increase in the placental weight resulted from compensation for oxygen and nutrient diffusion by increasing exchange surface area of the placenta.



Sigmoid Curve its acurve headed shape
is which had typical question in meth

Our results agreed with the results of Tericco et al who also reported a higher placental weight in GDM, when compared to the placental weight of uncomplicated pregnancies.[37]

In our study we observed lower feto-placental weight ratio, because the increased placental weight was proportionally greater than the increased fetal weight; leading to a decreased feto-placental weight ratio.

Terrico et al and Lao et al explained this by that most of placental growth occurs in the first half of gestation, well before GDM diagnosis and beginning of treatment, while the growth of the fetus occurs mainly in the third trimester as showed Lao et al in 1997 and Tericco et al in

2003.[34,35] They both stated that fetal growth is regulated by the balance between the fetal nutrient demand and the maternal-placental supply that is strictly related to utero-placental blood flow, placental size and its transfer capability. However, placental growth follows an S-curve regression while fetal growth follows an exponential pattern with maximum growth in the third trimester when there is also a very significant increase in fetal body fat mass. As a consequence, the feto-placental weight ratio decreased significantly during pregnancy.[37,38]

Various histopathological changes were observed in our study by examination the placentae of both groups; these are infarction, chorangiosis, fibrinoid necrosis, hydropic villi, immature villi, lymphohistocytes and nucleated RBCs.

Regarding infarction, lymphohistocytes, NRBCs and immature villi were only seen in the placentae of GDM, while chorangiosis, fibrinoid necrosis and hydropic villi were seen in the placentae of both groups but with a lower percentage in the control group.

Regarding the presence of ischemia and fetal vessel thrombosis were not observed in the placentae of both groups in our study.

Our results inconsistent with Saldeen et al who reported the presence of ischemia and fetal vessel thrombosis in his study (this could be explained by involvement of a large study groups and for a longer period of time), while the other hstopathological changes were reported in the placentae of GDM of his study.[39]

Saldeen et al also reported the presence of immature villi, ischema and thrombosis in the placentae of both groups (with a p value <0.05), this disagree with our finding as we observed the presence of immature villi in the placentae of GDM only.

Saldeen et al explain the presence of placental histopathological changes in the placenta of uncomplicated pregnancies, as the pregnancy may pass without a well-established clinical significance of diabetes.[37]

The presence of immature villi in our study was 4% of GDM group. Our findings disagreed with Jauniaux and Burton study, as they observed the presence of immature villi in the placentae of GDM and uncomplicated pregnancies, the volume of immature villi was greater in the diabetic group, but the difference were insufficiently large to reach statistical significance.[17] This could be explained by higher study group of our thesis and different

methodologies in the studies, in our study we use paraffin while in their study, they use resin section.

Regarding NRBCs, in our study we observed their presence in the placentae of GDM only about 2.5%.Our results were inconsistent with the results of Daskalakis et al, who found NRBCs in the placentae of both groups.

Daskalakis et al hypothesized that presence of NRBC, chorangiosis and villous immaturity suggests fetal hypoxia. These finding results from capillary hyperplasia and impaired trophoblastic invasion to the maternal spiral arteries in the intervillous spaces, this result in increased diffusion distance between intervillous spaces and fetal capillary that result in increase of exchange surface area.[24]

Evers et al observed histopathological changes in the placentae of diabetic pregnancies and uncomplicated pregnancies in various percentages; also he observed the presence of these histopathological changes in the placentae of pregnancies complicated by type 1 diabetes mellitus.

Evers et al suggests that the presence of histopathological changes in the placentae of uncomplicated pregnancies may be because of undiagnosed diabetes or passed of little clinical compliance.[25]

In conclusion, the treatment of gestational diabetes, by achieving optimal glycemic control, is able to modulate fetal weight proportionally more than placental weight. Infant of diabetic mothers may be protected against chronic hypoxemia because of a relatively high placental weight corresponding to an increase exchange surface as.

The histological lesions that associated with impaired glucose metabolism may persist despite of tight metabolic control and may explain the steady occurrence of fetal and neonatal complications.

CONCLUSIONS

Histological abnormalities were observed more frequently in the diabetic placentae compared to the controls. These findings support the hypothesis that impaired placental function is one of the main reasons for the increased frequency of fetal complications in diabetic pregnancies.

RECOMMENDATIONS

Large group study, wider time of study are recommended in order to search for other various histopathological changes in placentae of GDM (which were not found in our study) and compare it with placentae of uncomplicated pregnancies.

Also further studies are recommended to search these histopathological changes in placentae of pregnancies complicated by type 1 and type 2 diabetes mellitus and compare it with placentae of uncomplicated pregnancies.

ACKNOWLEDGEMENT

I would like to express my sincere attitude in deep appreciation to my teacher and supervisor Assistant professor Dr.Lilyan Sersam for her great help, kind advice and enthusiastic support throughout the period of this study.

Placental Pathology in women

Complicated by Gestational Diabetes

REFERENCES

1. Callahan .T.L, Caughey.A.B. Diabetes in pregnancy In: Blueprints Obstetrics & Gynecology 4th edition, 2007; 105-112.
2. Konje.J.C. Gestational Diabetes In: Obstetrics & Gynecology, An Evidence — Based text book for MRCOG, 2004; 172-178.
3. Dornhorst.A, Williamson.C. Diabetes and endocrine diseases In: pregnancy in Dewhurst 's Textbook of Obstetrics & Gynaecology. 7th edition, 2007; 246-253.
4. Moore. T.R, Metzger.B.E. et al Gestational Diabetes from Wikipedia, 2008.
5. Weinstock. R.S, Zygmont.S.V Pancreatic Islet Function Test. In: Endocrine Book, 2007; 123-130.
6. World Health Organization. Definition and Diagnosis of diabetes mellitus and Intermediate Hyperglycemia, report of WHO/ IDF, Geneva. WHO, 2006.
7. Reece.E.A, Homko.C.J. Diabetes Mellitus and Pregnancy In: Danforth's Obstetrics & Gynecology, 2004; 247-255.
8. James.D.K, Weinerant.C.P et al. Diabetes Mellitus In: High Risk Pregnancy. 3d edition, 2006; 996-1000.
9. Gary.F, Leveno.K.J et al. Medical and Surgical Complications In: Williams Obstetrics'. 22nd edition, 2005; 2: 1172-1183.

10. Koos.B, Moore.J.G, Gambonb.J.C. Common Medical and Surgical Conditions Complicating Pregnancy In: Essentials of Obstetrics & Gynecology, 2004; 216-221.
11. Altshuler, Benirschke, Driscoll. et al. Placental Triage, 2001.
12. Sadler.T.W. Langman's Medical Embryology, 2004; 2-6: 31-148.
13. Gary.F, Leveno.K.J et al. Anatomy and Phisiology In: Williams Obstetrics'. 22nd edition, 2005; 1: 54-64.
14. Beischer.N.A. Fetal and Placental Development In: Care of Pregnant Women and her Baby, 1989; 52-60.
15. Roberts. D. J. Histopathology of Placental Disorders, 2008.
16. Roberts.D.J, Oliva.E. Clinical Significance of Placental Examination in Perinatal Medicine. J Matern Fetal Neonatal Med., 2006; 19(5): 255-64.
17. Jauniaux. E, Burton. GJ. Villous histomorphology and placental bed biopsy investigation in type 1 diabetic pregnancies.
18. Makhseed M, Musini V.M, et al. Placental Pathology in Relation to the White,s classification of diabetes mellitus. Arch Gynecol Obstet, 2002; 266(3): 136-40.
19. Rhman.M.H, Sultana.S.Z, et al. Vascular Changes in the Placenta of Diabetic mothers and Hypertensive Diabetic mothers. Mymensingh Med J., 2006; 15(2): 176-9.
20. Kucuk.M, Doymaz.F. Placental weight and placental weight to birth weight ratio are increased in diet and exercise treated gestational diabetes subjects but not in subjects with one abnormal value on 100-g OGTT. J Diabetes, 2009; 23(1): 25-31.
21. Rahman.MH, Sultana.SZ.et al. Vascular changes in the placenta of diabetic mothers and hypertensive diabetic mothers. Mymensingh Medical J, 2006; 15(2): 176-179.
22. Younes.B, Al-Nuaim.L et al. Basement membrane thickening in the placentae from diabetic women. J pathology international, 2008; 46(2): 100-104.
23. Giachini. F. R. C, Carriel. V. et al. Maternal diabetes affects extracellular matrix components during placentation. Journal of Anatomy, 2008; 212(1): 31-41.
24. Daskalakis G., Marindpoulos.S. et al. Placental pathology in women with Gestational Diabets. J Acta Obstetricia et Gynecologica Scandinavica, 2008; 4(87): 403 — 407.
25. Evers.I.M, Nikkles. P.G.J, et al. Placental Pathology in Women with Type 1 Diabetes and in a Control Group with Normal and Large-for Gestational-Age Infants. J Placenta, 2003; 24(8-9): 819-825.
26. Ahmed. M.A, Makhseed. M.A, Musini. V.M. Impaired Gestational glucose tolerance. It's effect on Placental Pathology. Saudi Medical J., 2004; 25(9): 1241-1244.

27. Clarson. C, Teveaarwerk. G.J, et al. Placental weight in diabetic pregnancies. J Placenta, 1989; 10(3): 275-281.
28. M0ndestin.M.A.J, Ananth.C.V. et al. Birth weight and fetal death in the United States: The effect of maternal diabetes during pregnancy. Am J Obstetric & Gynecology, 2002; 187(4): 922-926
29. Myatt.L, Cui.X. Oxidative stress in the placenta. J Histochemistrey and Cell Biology, 2004; 122(4): 369-382.
30. Tutin. A, Madazl. R, et al. The Incidence of Placental Abnormalities, Maternal and Cord Plasma Malondialdehyde and Vascular Endothelial Growth Factor Levels in Women with Gestational Diabetes Mellitus and Nondiabetic Controls. J Gynecologic and Obestetric Investigation, 2008; 65: 227-232.
31. Mayhew. T. M, Sisley. I. Quantitative Studies on the Villi, Trophoblast and Intervillious spaces of Placentae from Women with well —controlled Diabetes Mellitus. J Placenta, 1998; 19: 371-377.
32. David. A, Schwart. Z. Placental pathology and obstetrical out come. J Healthcare Risk Manger, 2000; 4: 7-10.
33. Pietryga. M, Biczysko. W, et al. Ultrastructural examination of the placenta in pregnancy complicated by diabetes mellitus. J Ginekol Po1, 2004; 75(2): 111-118.
34. Huettner. PH.C. Placental Development, Indications for and Methods of Examination. The United States & Canada Pathology Acadamy. Annual Meeting, 2008.
35. Mayhew.T.M. Enhanced fetoplacental angiogenesis in pre-gestational diabetes mellitus: the extra growth is exclusively longitudinal and not accompanied by microvascular remolding. J Diabetologia, 2002; 45: 1434-1439.
36. Campbell.I.W, et al. Placental dysfunction and stillbirth in gestational diabetes mellitus. The British Journal of Diabetes and Vascular Disease, 2009; 9: 3-8.
37. Terrico. E. et al. Foetal and Placental Weight in Relation to Maternal Characteristics in Gestational Diabetes. J Placenta, 2003; 24: 343-347.
38. Lao. T.T et al. Placental Weight to Birthweight Ratio is increased in Mild Gestational Glucose Intolerance. J Placenta, 1997; 18: 227-230.
39. Saldeen. P et al. Structural, functional and circulatory placental changes associated with impaired glucose metabolism. J European J of Obestetrics & Gynecology and Reproductive Biology, 2002; 105: 136-142.