

## ORIGINAL ARTICLE

# Histochemical Changes in Placenta of Diabetic Pregnant Women

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### ABSTRACT

Placenta is the most important and vital organ of intrauterine life. It shows various histological changes in case of diabetic mothers. In the present study we have studied the histological changes in placenta of diabetic pregnant females and compared them with normal placenta. A total of 40 placentas were collected immediately after delivery for the study. Out of these 40 placentas, 30 were from diabetic pregnant females (as experience group) and 10 from normal pregnant females (as control group). Specimens of placenta were obtained after delivery, and then the placenta put to the gross examination includes the fetal and maternal surface. For light microscopy tissue samples was taken and fixed in formalin then followed by embedding in paraffin and 5-6 micron serial sections were generated with the microtome. The tissue sections were stained with H&E, PAS and van Gieson methods. Observations were carried out under a light microscope and differences between both control and diabetic group were studied. The results obtained from this study emphasized that on histological examination the diabetic placenta showed increased: Syncytial knots, Glycogen deposition in the axis of villi, collagen deposition and structural changes as compared to normal placenta. The mean blood sugar in the baby groups were significantly different between groups ( $p < 0.05$ ), and in babies of diabetics the levels of blood sugar were lower than control babies. Based on the findings of this study and the previous, it is concluded that several histological changes could be seen in placenta of diabetic pregnant females.

**Key words:** Placenta, Diabetes Mellitus, Collagen, Glycogen, Syncytial knot.

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### INTRODUCTION

Despite progress in recent years, particularly in the field of diabetes mellitus and gestational diabetes, again diabetes is a serious threat in pregnancy and increase the risk of fetal malformations, and also increases the risk Birthdays [11].

Placenta is a fetal organ that is located between the mother and fetus is essential for embryo development. In addition to food transfer, other functions such as the synthesis of a variety of hormones and growth factors are performed by placenta. Mothers Diabetes causes many changes in a variety of hormones, cytokines and maternal metabolites and also same in fetal blood circulation. The diabetes-related changes also affects placenta, as receptors, enzymes and transporters are often existence at two sides of the maternal and fetal placental. Varieties of structural and functional changes in the placenta due to mother's diabetes were reported. Although not all of these changes occurs in all placenta, but it seems that it may depend on the type and severity of maternal diabetes [15].

Recent studies suggest that placenta changes even in providing optimal control of blood sugar levels also occurs [5]. These changes have been observed in one third of diabetes cases prior pregnancies, it seems there has been a relation between these changes and poor control of diabetes. However still it is not clear that how the proper control of diabetes is effective in the improvement of these changes [11].

Diabetes mellitus is a common endocrine disorder that can cause significant complications of pregnancy. Congenital malformations, premature birth, fetal macrosomia and intrauterine growth restriction, are common complications associated with gestational diabetes [18]. Microscopic and macroscopic pathology of placenta during pregnancies complicated by diabetes vary to the extent that there is disagreement among the

results[24]. Perhaps the reason of difference in results is due to different ages of mothers that diabetes occurs, either due to the severity of patient diabetes[18].

The exact pathogenesis of placental dysfunction is still unknown, But there is a strong suspicion that the severity of the abnormalities not only depends on the severity of the mother's diabetes, rather than it is dependent of mothers diabetes control during her pregnancy, In the case of women who have mild diabetes or diabetes that is well controlled, Such anomalies are not seen, or it is very mild [25].

Because in case of changes in the placenta in diabetics, there were not general agreements, the aim of this study is to investigate the microscopic structure of the placenta in diabetic patients.

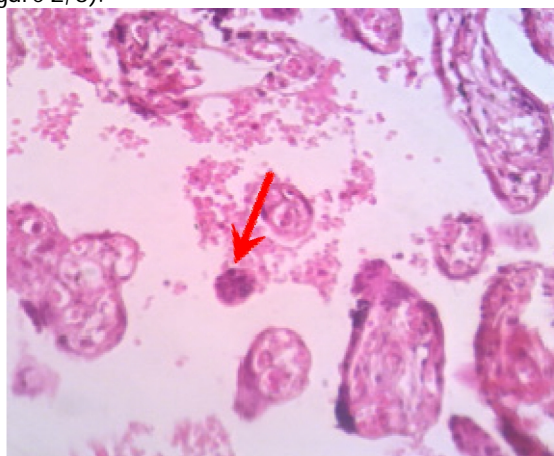
### MATERIALS ANDMETHODS

The study was conducted in two groups. The control group consisted of individuals who were not diabetic and diabetic in the experimental group were investigated. After childbirth in diabetic cases and delivery of the placenta, apparent status of maternal and fetal placental was examined.

Then samples taken from the placenta, and the samples obtained were studied by light microscope. For studying by light microscopy, samples were fixed in formalin and paraffin and then were prepared for sectioning. From the samples 5-6micronthicksections were prepared. The sections were transferred on glass slides and by 25% alcohol was wide open as possible. The slides has been moved to a warm house and for examining placental morphology H&E staining, to investigation rate of glycogen precipitation PAS staining, and for evaluation of collagen fibers van Gieson staining was used. Histopathological changes in diabetic groups were compared with control groups. The results of maternal and fetal blood sugar and placental weight were analyzed by using Independent t-test of PASW SPSS software (18<sup>th</sup> version).

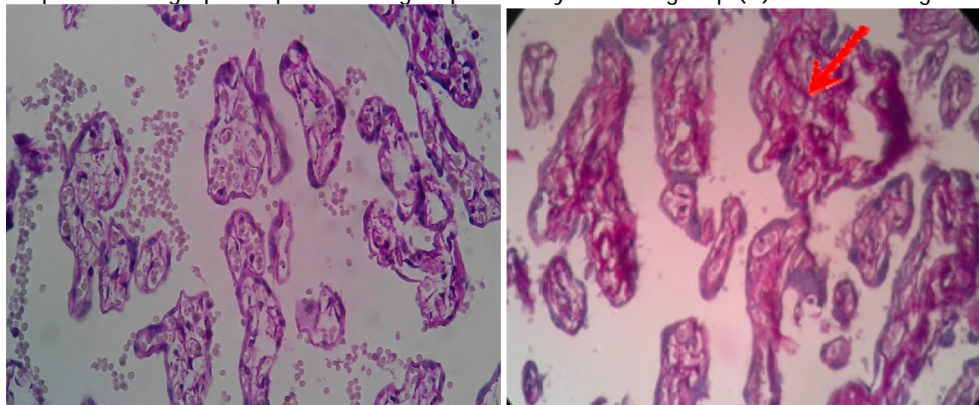
### RESULTS AND DISCUSSION

The results obtained from this study were presented into two parts: light microscopy and statistical analysis. Histological analysis of the diabetic placenta in current study revealed increased syncytia I knot (figure 1). Histochemical analysis showed that PAS and Van Gieson reactivity for glycogen and collagen was much stronger in diabetic group (figure 2, 3).



**Control group (A)**

Figure 1. The photomicrographs of placenta in groups of study: Control group (A) H & E Staining.



**Control group (A)**

**Experimental group (B)**

Figure 2. The photomicrographs of placenta in groups of study: Control group (A), Experimental group (B), PAS Staining.

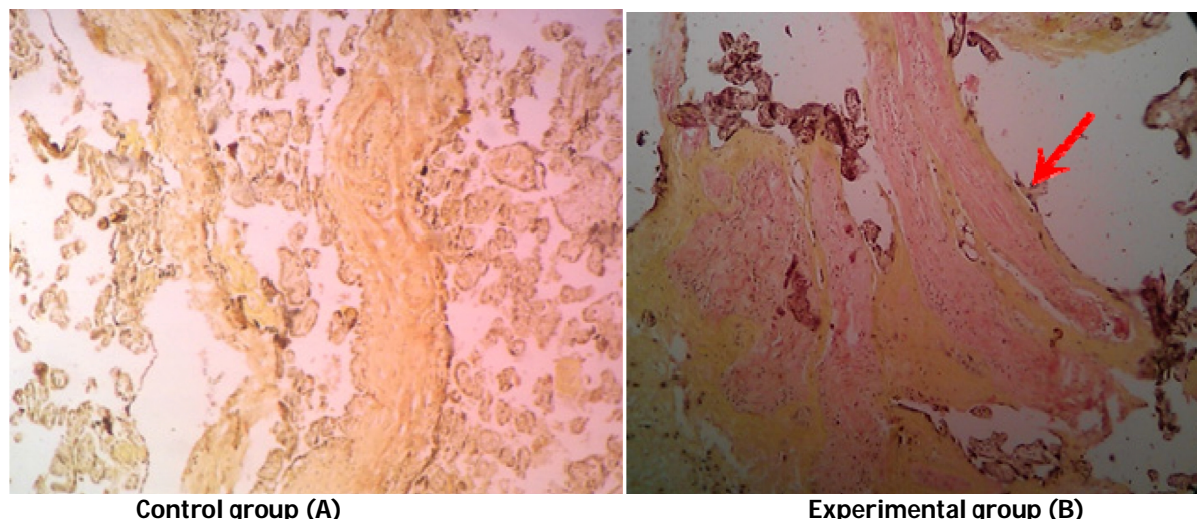


Figure 3. The photomicrographs of placenta in groups of study: Control group (A), Experimental group (B), Van Gieson Staining. Pay attention to collagen deposition in diabetic group

The results of maternal and fetal blood sugar, placental and baby weight and mother age in two groups were given in table-1.

Table1: Maternal and fetal blood sugar, placental and baby weight and mothers age in two groups (Mean±SE)

	Mothers blood sugar	Baby blood sugar	Baby weight	Placenta weight	Mothers age
<b>Control</b>	97.30±2.82	68.00±2.00	3618.66±79.61	585.00±32.53	28.40±1.81
<b>Diabetics</b>	100.86±2.23	62.76±1.24	3850.00±385.35	620.70±23.31	32.13±0.92
<b>P value</b>	0.402	0.039	0.374	0.438	0.058

The results indicated that mother's blood sugar levels in the studied groups were not significantly different ( $p>0.05$ ). But the mother's blood sugar levels in diabetic cases were slightly higher than the control group. The mean blood sugar in the baby groups were significantly different between groups ( $p<0.05$ ), and in babies of diabetics the levels of blood sugar were lower than control babies.

The mean birth weight of babies and placenta weights were higher in diabetics but there were not statistically different between two groups.

The results showed that the average age of mothers in two groups were not statistically different, although diabetic mothers were older than control group mothers.

The effect of treatment on mean birth weight in the diabetic group, also were studied and the results indicated that the mean birth weight in insulin-treated group ( $3817.33\pm 103.42$ ) was higher very significantly ( $p<0.01$ ) than the diet treated group ( $3420.00\pm 99.16$ ).

Generally, placenta of diabetic mothers in comparison to non-diabetic mothers undergoes some changes. However, due to various contributing factors in gestational diabetes such as: type of diabetes, severity of diabetes, treatment method and quality of blood sugar control, in diabetic women still the exact form of placenta changes was not known [7, 9, 14, 23].

Although, some researchers believe that desirable control of the gestational diabetes, causes microscopic and macroscopic changes do not occurs in placenta [19, 20]. But others believe that the majority of histological changes in the placenta of mothers who have had diabetes for a longtime and even the well-controlled diabetics and also in those with diabetes is not well controlled, they can be seen, but none of the results have not demonstrated a pathological pattern [1, 8, 12, 25].

The results of this study showed that the mean placental weight were not significantly different between the two groups ( $p>0.05$ ). But the mean placental weight, in diabetic group higher than the control group.

Moreover, significant increase in placental weight, placental volume and volume of the inter-villous space in diabetic placenta were described [16].

Also it is demonstrated that the type of diet could also partly responsible for the increased size and weight of the placenta, since there is an inverse relationship exists between protein intake and placental weight [6]. It is indicated that the diabetic placenta with a protective mechanism experience some changes in structure and function in response to maternal diabetes [2].

Except increase in placenta weight there was not any precise macroscopic evaluations of pregnant diabetic women placenta with non-diabetics, thus most anomalies must be investigated from a microscopic point of view[21].

The results obtained from this study indicated glycogen deposition in the villous and collagen deposition in the placenta of pregnant women with diabetes. Our findings also confirmed by the Jones and Desoye [17]. Other researchers indicated Hyperglycemia causes accumulation of glycogen within the cells and also reduce the accumulation off at particles in the trophoblast and does not have any additional effect on the metabolism and function of trophoblast [22]. While phospholipids and triglyceride content increase in diabetic placenta have been described [10].

The other findings of this study, increasing syncytial knots in the diabetic group compared with the control group. The results of Ansari (2010) and Yang (1993) in relation with diabetics mothers placenta, indicated that the histological changes includes villous immaturity and increasing citotrophoblast syncytial knots [3, 26].

An increase in syncytial knots in diabetic placenta in comparison to normal placenta has been shown [13]. In gestational diabetes the thickness of the basal membrane of syncytio-trophoblast increases. This thickening can occur as a result of the accumulation of muco-polysaccharide [4].

## CONCLUSIONS

The results obtained from this study showed that histological and histochemical changes were seen in placenta of diabetic pregnant females. This study revealed higher PAS reactivity in diabetic placenta and collagen accumulation was stronger in diabetic group.

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