

Microscopic Changes of the Placental Components in Maternal Anaemia

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Abstract:

Background: The placenta is vital for the maintenance of a normal pregnancy. Foetal growth and well-being depend on the functional and structural component of the placenta. The architecture of the placenta has been claimed to be changed in maternal diseases like anaemia.

Study design: Descriptive type of study.

Place and period of study: Department of Anatomy, Dhaka Medical College, Dhaka, from July 2005 to June 2006.

Materials and Methods: Sixty (60) placentae of Bangladeshi women were collected from Department of Obstetrics & Gynaecology, Dhaka Medical College Hospital and Maternal and Child Health Training Institute (widely known as Azimpur Maternity), Dhaka within 6 hours of delivery. Out of 60 placentae, anaemic and control group were 40 and 20 respectively. The study was designed to determine the histological changes of placental components, which is influenced by maternal anaemia. Microscopic study of placenta was done by light compound microscope with Harris' Haematoxylin & Eosin stain. The samples were divided into group A (control), group B₁ (mild anaemia), group B₂ (moderate anaemia) and group B₃ (severe anaemia).

Result: Pathological areas were significantly increased and villous area was reduced in anaemic groups.

Conclusion: A comprehensive work considering the physiological, biochemical, genetic, electron microscopic and morphometric placental studies are needed to support the present study.

Keywords: Maternal anaemia, placenta, villous area, pathological area.

Introduction:

Pregnancy is a healthy and welcoming process and children are God's blessing to a couple as well as to that family. The placenta is vital for the maintenance of a normal pregnancy¹. Foetal growth and well-being depends on the functional and structural component of the placenta. The architecture of the placenta has been claimed to be changed in maternal diseases like anaemia². The

placenta is a fetomaternal organ, which begins developing at implantation of the blastocyst and is delivered with the foetus at birth. During that 9 months period, it provides nutrition, gas exchange, waste removal, endocrine and immune support for the developing foetus³. Placenta is the structure where the foetal and maternal tissues come in direct contact without rejection. Maternal blood bathes the surfaces of the chorion, which fill the intervillous space. In early development, the blood vessels of the villi become connected with vessels from the embryo. The placenta is the site of exchange between maternal and foetal circulation⁴.

The basic parenchyma of the placenta is the trophoblast. The trophoblast lies facing to the endometrium and produces elongated projection known as the primary villi. With the progressive development primary, secondary and tertiary villi are

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formed. Villi are the functional unit of the placenta⁵. Placenta is an organ that is essential to the survival of the foetus of the mammals. When pregnancy is complicated by maternal anaemia, many pathological changes, such as infraction, intervillous thrombosis, foetal vessel haemolysis etc., occur which reduces the functional villous mass. Placental development influenced by anaemia and hypoxia, e.g. causes abnormal trophoblast invasion and release of hypoxia inducible factor (HIF) in anaemia⁶. The present study was aimed to see the extent of histological changes of the placental components in maternal anaemia.

Materials and Methods:

This study was carried out on 60 placentas from Bangladeshi mothers. The mothers were selected as who were suffering from antenatal anaemia i.e. having haemoglobin level below 10gm/dl at any time before delivery and those who were control i.e. their haemoglobin levels were ≥ 10 gm/dl and their gestational age were from 36 to 40 weeks⁷. All placentae were collected after 35 weeks of pregnancy. The placentae were collected from Department of Obstetrics & Gynaecology, Dhaka Medical College Hospital and Maternal and Child Health Training Institute (widely known as Azimpur Maternity), Dhaka within 6 (six) hours of delivery. After collection of placentae, grouping was done according to mother's haemoglobin concentration, which was done during primary screening. Out of 60 placentae, 20 were from control mothers (A), who were not anaemic, 27 from mothers having mild anaemic (B₁) and 13 from mothers having moderately anaemic (B₂). Severe anaemic (B₃) mother was not found during the period of collection of placenta for this study. Grouping was done according to Singla et al¹ (Table-I). The mean haemoglobin level in control group was 10.5gm/dl, mean haemoglobin levels in mild and moderate anaemic groups were as 9.2gm/dl and 7.2gm/dl respectively⁷.

Procedure:

Each collected placenta was placed on a metallic tray and umbilical cord was cut at the insertion site. The placenta was cleaned with tap water until appearance of clear washout water and then the placenta was fixed in 10% formal saline solution.

A. Selection of the tissue:

Two Placental tissue blocks were taken from two widely separated cotyledon of each placenta. One block was taken from the cotyledon lying just opposite the insertion of the umbilical cord and other block from the periphery. The center of the cotyledon was identified by the typical central subchorionic blood lake⁸. Each block was measuring approximately 1cm x 1cm x 0.5cm. Tissue blocks were taken from areas, which appear, normal or least pathological⁹.

B. Processing of tissue:

Placental tissues were fixed in 10% formal saline in a plastic container for another 24 hours. Sections were prepared following a standard histological procedure and stained with Harris' Haematoxylin & Eosin (H & E) stain. Two paraffin blocks were made from each of the placentae. Sections were cut from each paraffin block at 5 μ m thickness.

C. Procedure of the measurement of the microscopic variables:

i) Measurement of the villous area:

Measurement of the total area occupied by the chorionic villi was done by using the integrating eye piece, which simply replaced the usual eye pieces of light microscope. These special eyepiece permits more rapid measurement and are accurate enough to measure the component parts of placental tissue⁹. Integrating eyepiece is a replica of zeiss integrating eyepiece which was prepared with transparent plastic sheet. The eyepiece contains a point network of 25 points, arranged within a circle, which delimits the counting field (Fig.-1). The test point graduation serves for determining the quantity, in volume percentage of individual constituents in a heterogeneous material according to the point counting procedure. Every point of the lattice, which accidentally lies above a particular component, is counted as 'hit' for the component. After completing the count, the hits for each component are separately added up and the ratio of the sum to the total count determined. The quota of hits for each individual component is equal to its volume i.e. the number of hits is proportional to the volume percentage. In the present study, this integrating eyepiece was used

to measure the volume percentage of vilolus area (chorionic villi).

ii) Measurement of volume of pathological area:

Histologically, the early infarct is characterized by aggregation of the villi in the affected side with marked narrowing, often obliteration of the intervillous space (Fig.2). The villous foetal vessels are dilated and congested, while the syncytial nuclei show early necrotic change such as pyknosis and there is progressive coagulative necrosis of the villi. The foetal erythrocytes trapped in the vessel of the infarcted villi undergo haemolysis¹⁰ (Fig.3). Intervillous thrombosis is a villous free nodular thrombosis with in the intervillous space (Fig.4). This measurement was done by same point counting method as mentioned above⁹ (Fig.5).

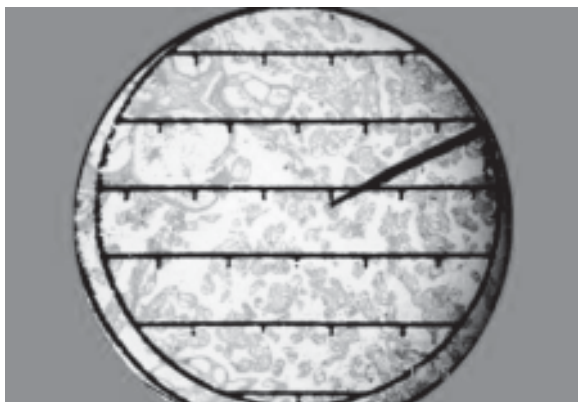


Fig.-1: Photomicrograph of placenta under integrating eyepiece to measure the villous area (H & E stain x 400).



Fig.-2: Photomicrograph of placenta of moderate group of anaemia showing infarction marked by indicator (H & E stain x 400).

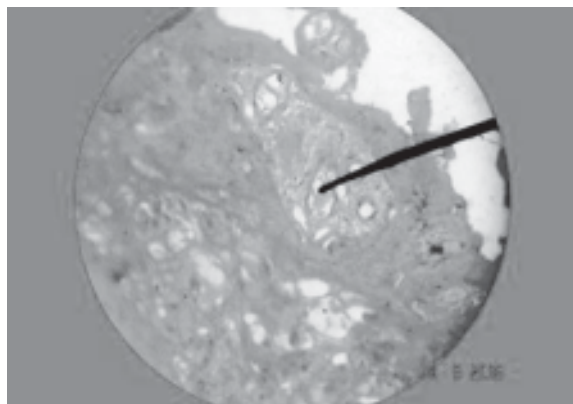


Fig.-3: Indicator of Photomicrograph indicates haemolysis of foetal capillary of moderate anaemic group (H & E stain x 400).

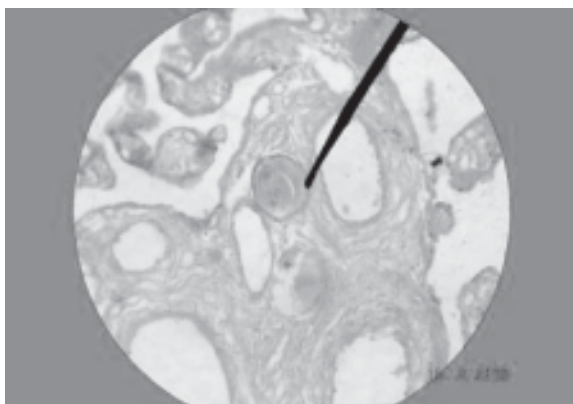


Fig.-4: Photomicrograph of placenta showing the intervillous thrombus, as marked by the indicator (H & E stain x 400).

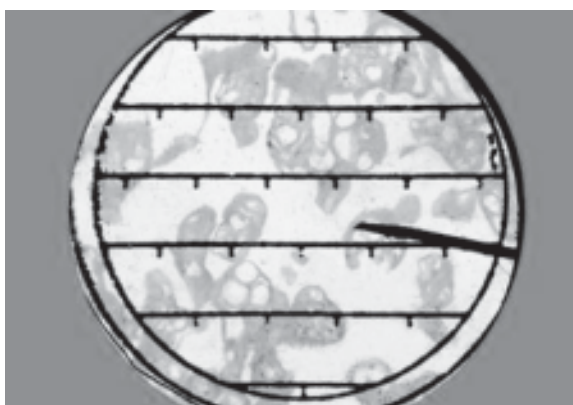


Fig.-5: Photomicrograph of placenta under integrating eyepiece to measure the pathological area (H & E stain x 400).

Results:*Volume of the villous area:*

In this present study, the mean±SD volume of the villous area of the placenta was 54.60±5.77, 52.01±3.40 and 51.01±5.57 in groups A, B₁ and B₂ respectively. Hence, it was evident that the difference in villous area of the placenta was statistically significant in between group A and B₂ (p<0.05) (Table-II).

Volume of the pathological area of the placenta:

It was found that the mean±SD volume of the pathological area of the placenta was 2.01±1.34, 3.00±2.05 and 4.28±1.94 in group A, B₁ and B₂ respectively. The mean difference in the volume of pathological area between group A and B₂ (p<0.001)

and B₁ and B₂ (p<0.05) were statistically significant (Table-II).

Table-I
Haemoglobin level in different groups

Group	No. of subjects (n)	Blood Haemoglobin Level(gm/dl)
A	20	10.50 (10-12)
B ₁	27	9.20 (8-9.2)
B ₂	13	7.20 (6.5-7.2)

Group A : Control

Group B₁ : Case (mild anaemic)

Group B₂ : Case (moderate anaemic)

Figures in parentheses indicate range.

Table-II
The volume of villous and pathological area of the placenta in different group

Group	No. of subjects (n)	Villous area (volume %)	Pathological area (volume %)
		Mean±SD	Mean±SD
A	20	54.60±5.77 (44.9 65.9)	2.01±1.34 (0.2 4.2)
B ₁	27	52.01±3.90 (45.4 60.1)	3.00±2.05 (0.3 8.0)
B ₂	13	51.01±5.57 (42.3 57.1)	4.28±1.94 (0.9 8.2)
		P value	P value
A vs B ₁		>0.05 ^{ns}	>0.050 ^{ns}
A vs B ₂		<0.05*	<0.001***
B ₁ vs B ₂		>0.50 ^{ns}	<0.05*

Group A : Control

Group B : Case (mildly anaemic)

Group C : Case (moderately anaemic)

Figures in parentheses indicate range. Statistical analysis done by ANOVA (multiple comparison), */***=significant, ns=not significant.

Discussion:

Placental tissues from different parts of the placenta may differ in their histometric characteristics¹⁰. Therefore, the some specific sites were chosen for taking tissues for histological examinations. Again, tissues were taken only from the intermediate part of the cotyledon half way between the maternal and foetal surfaces of the placenta for avoiding the structural difference in tissues from parabasal and subchorionic areas. Teasdale² demonstrated that the former areas had significantly more peripheral villi and villous space for necessary exchange between the mother and the foetus. Reshetnikova et al.¹¹ observed a considerable reduction in the total volume of terminal villi in anaemic group as compared to control group. They also observed a greater value of pathological area 32.1 ± 6.5 in anaemic group and 4.1 ± 0.6 in control group. Wong and Latour⁹ observed that the smaller villous area in anaemic group (53.6%) compared to control group (57.5%). Both observations are similar to the present study (51.01% villous area in moderately anaemic and 54.60% in control group). These are statistically highly significant ($p < 0.001$). Pathological area also increased significantly in anaemic group (3% in mild anaemia and 4% in moderate anaemia group as compared to 2 percent in control group).

Conclusion:

A comprehensive work considering the physiological, biochemical, genetic, electron microscopic and further morphometric placental studies with larger sample are needed to support the present study.

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