Original Article

Excessive Placental Calcification Observed in PIH Patients and its Relation to Fetal Outcome

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ABSTRACT

OBJECTIVE: To see the morphological changes in excessive placental calcification in pregnancy induced hypertension (PIH) and its relation with fetal outcome.
STUDY DESIGN: Prospective comparative study.
PLACE AND DURATION: This study was conducted from June 2008 to July 2009 at the department of Anatomy of Liaquat University of Medical & Health Sciences Jamshoro and placentae were collected from department of Gynecology & Obstetrics of Liaquat University Hospital.
MATERIAL AND METHODS: Eighty freshly delivered placentae were collected from labor room and gynecology operation theatre of Liaquat University Hospital. Forty placentae from parturient that had pregnancy induced hypertension (PIH) & forty placentae from parturient belonged to uncomplicated pregnancy (control group). Ages of all parturient were between 17 to 32 years. Fetal outcome and data was recorded. Placentae were measured on a weighing machine graduated in grams and diameter was measured with the help of a measuring tape in centimeters. Approximately five mm piece of placenta was taken and processed for histological examination.
RESULTS: The weight of placenta in control group ranges from 450 to 650 gm with a mean weight of 526.25±8.414 gm having diameter from 19 to 24 cm with a mean of 21.225±0.2148cm. In PIH group weight of placenta ranges from 200 to 550gm with a mean weight of 432.25 ± 11.889gm with diameter ranges from 10 to 16cms with a mean14.208 ± 0.1914cm. The difference in weight and diameter of placenta in PIH was found statistically significant when compared with weight and diameter of normal placentae.
The birth weight of new born babies in control group was 1.8 kg to 3.6 kg with mean of 2.790± 0.0689kg. In PIH group the fetal weight was 1.4 kg to 3.0 kg with mean weight of 2.195 ±0.0703kg.
CONCLUSION: Fetal outcome in terms of birth weight of newborn to mother having PIH and calcification of placentae (grossly and microscopically) was poor as compared to control group.

KEY WORDS: Placenta, PIH, calcification.

INTRODUCTION

The placenta is unique organ. Short lived by design, its brief existence ensures survival of human embryo/ fetus in the intrauterine environment. The placenta performs diversity of functions, ranging from anchoring the fertilized ovum, preventing its rejection by the maternal immune system to enabling the transport of nutrients and wastes between the mother and the embryo/fetus [1].

The placenta was first recognized by early Egyptian, while named by Realdus Columbus in 1559. The word placenta comes from the Latin word plakos for cake or from Greek plakóenta mean “flat, slab-like", referring to its round and flat appearance in humans [2,3] .

Calcification is common in human placentae and recognized as a normal feature of maturation and aging of this organ. The placental calcification is a constant process occurring at a time in different parts of placenta to variable degree. The incidence of fetal distress in excessively calcified placenta is four times more than in the uncalcified group. During pregnancy placental growth must correspond with fetal growth because increased calcification of placenta has serious negative consequences. Pathological maturation of placenta leads to fetal growth restriction. During pregnancy minor placental calcification tend to be found at late stages of gestation which is associated with tissue aging due to the induction of some age related dystrophic changes in the placenta [4, 5].

Deposition of calcium salts is more on the maternal surface, in the basal plate, along the septa and basement membrane of placental villi, perivillous space, and subchorionic space, degenerative villi also show calcification [6-8].

Morphologically calcium deposits are seen as white or pale colored fine granules or clumps often felt as gritty deposits. Microscopically calcification appears intracellular as well as extracellular basophilic deposits when stained with haemotoxylin and eosin staining [9].

Excessive placental calcification may be associated with Pregnancy induced hypertension (PIH), Placental

abruption, intra uterine growth restriction (IUGR), cigarette smoking [10].

Pregnancy-induced hypertension (PIH), also known as toxemia of pregnancy, is a life-threatening disorder that usually develops late in the second trimester or in the third trimester. The ratio of this complication is approximately 1:1500 pregnancies. It is a major cause of maternal and perinatal morbidity and mortality worldwide. It is an etiologically heterogeneous disorder that occurs in at least two subsets, one with normal or enhanced placental function, and another involving placental dysfunction and fetal growth restriction, often with asymmetric fetal body proportion, reduced fetal length, and preterm delivery [11, 12].

PIH significantly increases the risk of low birth weight, both by increasing preterm birth as well as decreasing fetal growth because of hypoxia consequent upon utero-placental insufficiency; which when severe can cause intrauterine death. Mothers with moderate to severe PIH have smaller, irregular placentae, marginal insertion of umbilical cord. Also calcification and histological features of vascular insufficiency like thrombosis, infarctions etc were observed. These changes are directly proportional to the severity of the disease and perinatal outcome becomes worse with advancing grades of pregnancy induced hypertension.[13,14,15]

This study was conducted to examine excessive calcification in the placentae and its relation to fetal outcome in pregnancies complicated by PIH.

MATERIAL AND METHODS

This case control (prospective) study was conducted from June 2008 to July 2009 at the department of Anatomy of Liaquat University of Medical & Health Sciences Jamshoro. Placentae were collected from department of Gynecology & Obstetrics of Liaquat University Hospital.

Eighty placentae were collected from labor room and gynecology operation theatre of Liaquat University Hospital. Forty placentae from parturient that had pregnancy complicated by pregnancy induced hypertension & forty cases belonged to uncomplicated pregnancy (control group). All samples collected randomly from singleton pregnancy. In control group, women with normal blood pressure were included. In pregnancy-induced hypertension, women having blood pressure ranged 140/90mm of Hg or above were included. The patients having hypertension or diabetes before pregnancy were excluded. Age of all women was between 17 to 32 years.

The data was collected on the prescribed proforma. Fetal outcome was also recorded.

Method of sample collection

Placenta with umbilical cord and membranes was collected after delivery. In all cases, the amnion and chorion were removed from the placenta. The umbilical cord was cut at a distance of 5 centimeters from the site of attachment. Placenta was washed with water, dried with the help of blotting paper and weighed. The placentae along with the umbilical cord identified by code numbers and were preserved in 10 % formalin solution in plastic jar for 48 hours for fixation. [16]

The study of gross morphology of placenta was conducted in the Department of Anatomy at Liaquat University of Medical & Health Sciences Jamshoro. Placentae were weighted on a weighing machine graduated in grams (gm) and diameter was measured with the help of a measuring tape in centimeters (cm). All placentae were cut in two equal halves and then were further cut in small pieces. The cut pieces were labeled and randomly selected for further processing. Slides were stained with H & E for routine histological study and Von Kossa stain for calcium.[17,18]

RESULTS

Morphological changes (gross and histological) were recorded on the proforma. The results of the study are depicted in text and tables.

The excessive calcification of placenta in women of different parity and age group from PIH and control group is shown in **Table I**. In both groups 80% cases were between 17-28 years age. The primiparous in control group were 20%, while it was 50% in PIH group. Gravida 1-2 were equal in both groups and representing 30% for each group. Gravida 3-7 were 50% for control group while it was 20% for PIH group.

The variables of interest related to placentae are shown in **Table II** and **III**. The weight and diameter for both groups were compared using t test. The results show statistically significant ($p=<0.001$) difference in weight and diameter of placenta in PIH group as compared to control group. The weight of placenta in control group ranges from 450 to 650 gm with a mean weight of 526.25± 8.414 gm (mean ± SEM). In PIH group weight of placenta ranges from 200 to 550gm with a mean weight of 432.25 ± 11.889gm (mean ± SEM). The diameter of placenta in control group ranges from 19 to 24 cm with a mean of 21.225±0.2148cm (mean ± SEM). In PIH group diameter of placenta ranges from 10 to 16cms with a mean14.208 ± 0.1914cm (mean ± SEM).

Fetal outcome was considered with respect to the birth weight of the newborn. The birth weight of newborn in control group was 2.790 Kg (ranges from 1.8 to 3.6 Kg) as compare to mean birth weight of 2.195 Kg (ranges from 1.4 to 3.0 Kg). The difference in mean birth weight was statistically significant as shown in **Table IV**.

Microscopic examination was done by identifying

number of calcified areas per low power field. Numbers of classifications were counted by moving slide from right to left then above downwards. There was a tendency of more placental calcification in PIH placentae than that of control group. In control group the number of calcified areas per/lpf ranges from 01 to 05 with a mean of 2.93±0.210 (mean ± SEM) while it was 10 to 20 with a mean of 14.97 ±0.445 (mean ± SEM) in placentae from PIH group. The difference was statistically highly significant with p= <0.001 as shown in table V. In general fetal outcome found poor in PIH group than control when compared with respect to still birth, intrauterine death, alive with Apgar score 03 or more than 05.

When comparing still birth , it was 5% in control group while it was 12.5% for PIH group, only 2.5% intrauterine death recorded in control as compared to 12.5% in PIH group. Alive with Apgar score 03 was seen in 05(12.5%) in control, in PIH it was 10(25%). Apgar score more than 05was seen high in control 32 (80%) as compared with 20 (50%) in PIH shown in table VI.

TABLE I: SHOWING AGE AND PARITY

Age Groups	No. of Cases	
	Control	PIH
17-20 years	10(25%)	04(10%)
21-24 years	07(17.5%)	12(30%)
25-28 years	15(37.5%)	16(40%)
29-32 years	08(20%)	08(20%)
Parity		
Po	08(20%)	20(50%)
P1-2	12(30%)	12(30%)
P3-7	20(50%)	08(20%)

TABLE II: WEIGHT OF PLACENTAE

Placental weight in gms.	Min. wt	Max. wt	Mean	Sd	P value
In control	450	650	526.25	53.214	<0.0001
In PIH	200	550	432.25	75.192	

TABLE III: DIAMETER OF PLACENTAE

Placental diameter in cms	Min	Max	Mean	SD	P value
In control	19	24	21.225	1.3585	<0.0001
In PIH	10	16	14.208	1.2103	

TABLE IV: BIRTH WEIGHT OF NEWBORN (n = 40)

Weight of newborn in kg.	Min	Max	Mean	Std	P value
In control	1.8	3.6	2.790	.4361	<0.0001
In PIH	1.4	3.0	2.195	.4449	

TABLE V: CALCIFICATIONS SEEN ON LIGHT MICROSCOPY/LPF (10/20) (n = 40)

	Min	Max	Mean	std. Deviation	p value
control	1	5	2.93	1.328	<0.0001
PIH	10	20	14.97	2.815	

TABLE VI: SHOWING FETAL OUTCOME n=40 Cases

Fetal out come	Control	PIH
still birth (sb)	02 (5%)	05 (12.5%)
Intra uterine death (iud)	01 (2.5%)	05 (12.5%)
apgar score of 03 or less at 05 min	05 (12.5%)	10 (25%)
alive with apgar more than 05	32 (80%)	20 (50%)

DISCUSSION

The placenta is a highly specialized organ of pregnancy that supports the normal growth and development of the fetus. The fetus, placenta and mother form a composite triad of dynamic equilibrium, and dysfunction of any one of them can affect the others. Calcification is common in human placentae and recognized as a normal part of maturation and aging of this organ. The pathological maturation of placenta can lead to fetal growth restriction which is the second most common cause of perinatal death after prematurity.[19, 20] Excessive placental calcification is seen in Pregnancy induced hypertension (PIH) which is the major cause of perinatal morbidity and mortality globally, especially in the developing world. PIH is associated with uteroplacental disease causing reduced uteroplacental perfusion, dysfunction resulting in fetal growth restriction, reduced fetal length, and preterm delivery.[21]

Placental weight reduction is probably a consistent finding in PIH. Our study showed reduced placental

weight in 66% patient of PIH then in placentae from parturient of control group; this is reported to be 50% by Sultana S, et al[23] and Rahman[24] while Palaskar PA et al[25] find it to be 77%.

The mean diameter of placentae from PIH found about 66% less when compared to placentae of normal parturient (14.2 cm, 21.22 cm), a finding almost identical to the finding reported earlier[17].

In our study mean birth weight of babies born to PIH mother was significantly low when compare to newborn of normal parturient ($p < 0.0001$). This is attributed to uteroplacental insufficiency and same finding has previously been reported by Udaina[21] and Sarwan[25].

Mufti P et al [26] has stated the Perinatal Mortality Rate (PMR) in Pakistan is 50-60/1000 which is one of the highest in the world. They attribute this to low birth weight. PIH is associated with 3.4-fold increased in perinatal mortality as shown in studies conducted by Xiong X et al[27], and Abbasi RM et al[28]. PIH is also considered to be leading cause of still birth in 45% cases as reported by Patil Y et al. The perinatal mortality was 52.0% in hypertensive women compared with 29.8% for normotensive.. [29, 30]

Placental calcification is seen more frequently in primigravidas as reported by Russel J G B, Fielden P. Identical results were observed in this study as well. Similar results have previously been cited by Spirit B A,Cohen W N, Weinstein H M. [31,32]

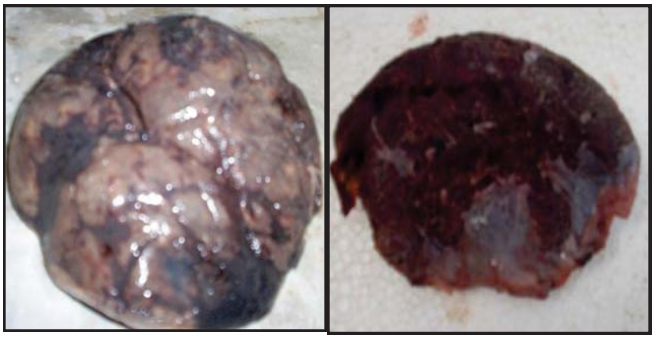
The calcium deposits are mostly observed in the villi and basement membrane of the villi which is strongly suggestive of uteroplacental insufficency because of narrow lumen. We see calcification from one millimeter to biggest one of about 5 millimeters. In placentae from PIH women, mean number of calcified areas on gross as well as on microscopy seen were more than in normal placenta. In the study conducted by Majumandar S form India observed similar findings [14] . The still birth in this study was 12.5%, however the fetal mortality and morbidity associated with PIH is found to be responsible for 28% of stillbirths. [33]

Although placental abnormalities cannot be modified to improve the perinatal outcome, identification in the antenatal period, and early referral to hospitals, can decrease the associated maternal and perinatal morbidity and mortality.

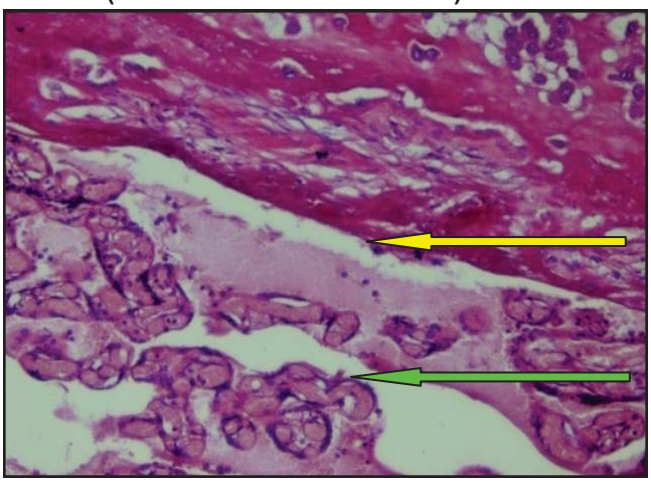
FIGURE I:

Placenta from normal parturient showing No gross morphological abnormality or calcification

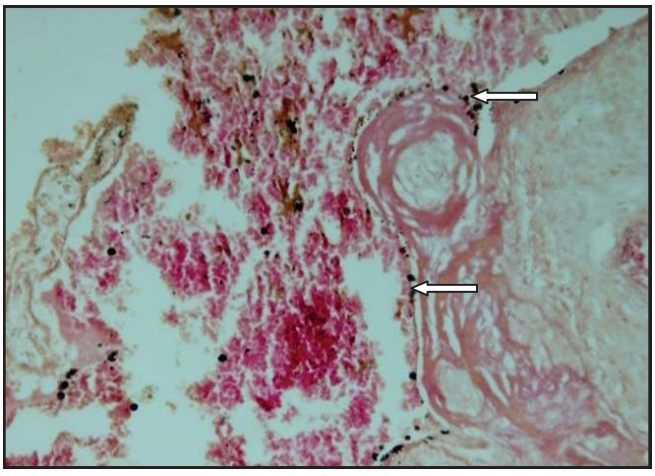
Placenta in PIH showing reduced diameter and visible calcification



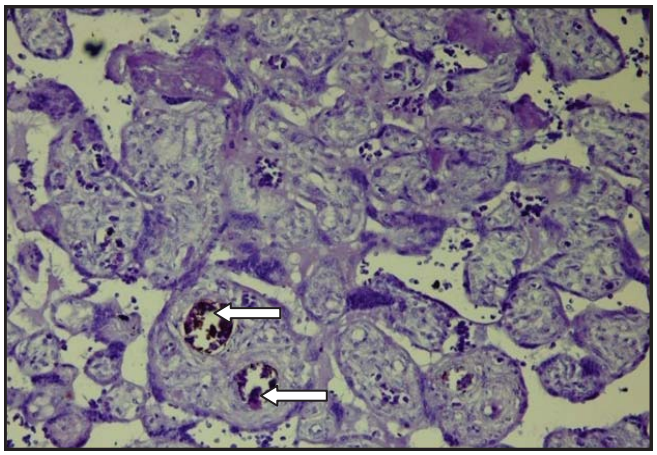
PHOTOMICROGRAPH I: AREAS OF CALCIFICATION IN NORMAL PLACENTA IN VILLI AND IN THE BASAL PLATE OF THE THIRD TRIMESTER PLACENTA (H & E×10 MAGNIFICATION)



PHOTOMICROGRAPH II OF PLACENTA IN PIH SHOWING CALCIFICATION BY ARROWS VON KOSSA STAIN X10 MAGNIFICATION



PHOTOMICROGRAPH III OF PLACENTA IN PIH SHOWING CALCIFICATION BY ARROWS IN VILLI ON H & E X 20



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