



Original Research Article

Morphometric study of human placenta and an insight into its vascular pattern by corrosion cast technique

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ABSTRACT

Introduction: The placenta is a chorioallantoic, deciduate, haemochorial and villous organ. It is a vital organ which transfers vital nutrients from the mother to the foetus and removes waste products from the foetus to the mother. There has been paucity of literature regarding the morphometric study of human placenta and its vascular pattern (magisterial or dispersed) by corrosion cast technique. Therefore, the present study was conducted on morphometry and vascular pattern of the human placentae.

Aim: To do the morphological and morphometric study of placenta and to study the vascular pattern of placenta by corrosion cast technique.

Materials and Methods: Study was done on 43 placentae and its morphometric study and vascular pattern were studied by corrosion cast technique.

Results: Oval was the most common shape of placenta observed in the present study. Out of 43 placentae, 22(51.16%) had oval, 16(37.20%) placentae had round shape followed by triangular in 3(6.97%) placentae. Only 1 (2.32%) placenta each found to be irregular and multilobed. Most common shape recorded in complicated pregnancy was oval followed by round. Magisterial pattern was found in 13.95% of placenta and was associated with oval shape constituting 25.58% of placentae. Mixed pattern was seen in 18.60% placentae and was associated with oval placentae(23.25%). The disperse pattern was seen in 4.65% and was associated with oval and irregular placentae each constituting(2.32%).

Conclusion: The vascular cast preparation of placentae in our study will further help in contribution towards the understanding of the placental vasculature. An effort should be made for performing more studies on placentae of complicated pregnancies eg: hypertension and diabetes etc.

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1. Introduction

Placenta represents an extremely intimate parabolic union of maternal and foetal tissues for the inevitable requirement of the embryo. Human placenta is an important organ for maintaining the pregnancy and normal foetal progress. Foetal outcome is adversely affected by pathological changes observed in placenta. The placenta is a chorioallantoic, deciduate, haemochorial and villous organ. It is a vital organ which transfers vital nutrients from the mother to the foetus and removes waste products from the foetus to the mother.¹ In intrauterine life, foetus

depends upon the placenta for nutrition which function as its lungs, liver & kidney.² The wellbeing of the foetus depends on many factors, but healthy placenta is the single most common factor in producing a healthy baby.³ The placenta acts as a unique organ in higher mammals which is attached to the uterus and the foetus via umbilical cord. The observation of the placenta in utero or postpartum, gives an important information regarding condition of the foetal wellbeing.⁴⁻⁶ An abnormal development had adverse impact on the foetus and results in various types of complications such as maternal hypertension, gestational diabetes and intrauterine growth retardation etc. Vascular casting technique is most common way for investigating the various diseases.^{7,8} To examine the three-dimensional

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structure of blood vessels, ductal system, and cavities of organ, corrosion cast is found to be a well-established technique. Well-prepared cast provides a unique tool to have an in-depth knowledge of the organ and tissue as they replicate the vasculature and ductal system anatomy.⁹ The vascular corrosion casting technique produces a replica of the vascular beds by injecting a plastic polymer that fills the blood vessels and rapidly polymerizes followed by corrosion of adjoining tissues with alkali solutions. After washing and drying, the hardened cast represents the geometry of the original vascular system.¹⁰ There has been paucity of literature regarding the morphometric study of human placenta and its vascular pattern (magisterial or dispersed) by corrosion cast technique. Therefore, the present study was conducted on morphometry and vascular pattern of the human placenta.

The aim & objectives of the present study were as follows:

1. Study of the morphology, morphometry of placenta and site of umbilical cord in normal pregnancy and in complicated pregnancy.
2. To study the vascular pattern of placenta by corrosion cast technique.

2. Materials and Methods

The present study was conducted in the Department of Anatomy. 43 placentae were procured from Department of Obstetrics and Gynaecology of Medical College & Associated Hospital. Approval was taken from the institutional ethical committee before the start of the study and the informed consent was taken for procurement of placenta from patients or their husband. This study was done for one year i.e. from November 2018 to November 2019. Out of 43 placentae which were taken for this study, 28 were from uncomplicated pregnancy and 13 were from complicated pregnancy having gestational hypertension and diabetes.

2.1. Morphological study

Placentae were examined for placental completeness, placental shape, umbilical cord insertion.

2.2. Morphometric study

1. Placental thickness was noted by Vernier callipers.
2. Diameter of the placenta was measured with a measuring tape.
3. Placental weight was recorded using a weighing scale.
4. Foetal surface was examined for vascular pattern—disperse, magisterial and mixed pattern and was recorded.
5. Type of insertion of the umbilical cord was noted.

6. Number of umbilical arteries and umbilical vein were counted.

2.3. Processing of placenta

Trimming of membrane was done. Specimens were washed under running tap water to remove the clot and debris on the surface and all the blood in the blood vessels was squeezed out. The vessels were then infused with 5000IU Heparin to avoid clots formation in the umbilical vessels.

2.4. Corrosion cast model formation

With the help of disposable syringes attached to plastic cannulas, the casting material was injected into the vessels according to the colour code (red for arteries and blue for veins). The injection was continued until the resistance was felt. The cannulas were removed, and the blood vessels were clamped and ligated. After that the placentae were kept in a refrigerator at a temperature of <20 degree Celsius for 3 to 4 days to allow the casting material to solidify. The specimen was immersed in KOH (corrosive agent) to dissolve the placental tissue, leaving behind the cast model of the placental vasculature. After the formation of placental vascular cast model, three type of placental vascular pattern were observed that is Disperse (Dichotomous), Magisterial (Monopodial), and Mixed. The diameter of umbilical vessels was also measured one at the entrance, second at their 1st bifurcation and also at the end with the help of vernier calliper. Besides that, the number of bifurcations were also counted.

2.5. Statistical analysis

At the end of the study, the data was collected and analysed statistically by using SPSS v. 17.0. A p value <0.05 was considered as significant.

3. Results

Maximum number of placentae collected for the study were taken from the age group ranged between 26-30 years i.e. 20 cases (50%), followed by 13(32.5%) whose age was < 25 years. Only 1 case (2.5%) was of age > 35 years. Mean age of the study population was 27.22 ± 3.61 years with the age range of 21-36 years.

Gestational Diabetes Mellitus(GDM)cases ranged between age 23 to 31 years & Hypertension(HTN) cases ranged between 24 years to 29 years

Gestational age of patients ranged between 30 to 41 weeks. Mean period of gestation of the present study was 38.14±2.15 weeks with a range of 30.5 - 44.1 weeks. POG for GDM pregnancy ranged from 36 to 40 weeks of gestation and for gestational pregnancy ranged from 30 to 41 weeks of gestation.

In the present study, range of placental weight was between 320 to 850 g. Mean placental weight of the present study was 549.5 ± 137.43 . Out of 12 complicated pregnancies, maximum placental weight recorded was 800 gms (GDM mother) and minimum recorded was 380 gm (HTN mother).

Oval was the most common shape observed in the present study. Out of 43 placentae, 22(51.16%) had oval shape followed by round in 16(37.20%) placentae, followed by triangular in 3(6.97%). Only 1 (2.32%) placenta each was found to be irregular and multilobed. Most common shape recorded in complicated pregnancy was oval followed by round. As shown in Figure 1.

In the present study, we observed eccentric cord insertion in 29 placentae (67.44%), central in 8 Placentae (18.60%), marginal in 5 placentae (11.62%) and velamentous cord insertion found only in 01 Placenta (2.32%), as shown in Figure 2 & Figure 3. Out of complicated pregnancy most common site of umbilical cord insertion found to be eccentric followed by central

In the present study the number of blood vessels observed in umbilical cord was three in 39 placenta (97%) and only two in one placenta (2.3%). In complicated cases one placental umbilical cord showed one artery and one vein. (Figures 3 and 4)

In an uncomplicated case, umbilical cord initially showed 2 arteries going towards placenta but after entering the placenta, one umbilical artery ends into the lumen of 2nd umbilical artery (Figure 5). Before it enters another umbilical artery, there is an anastomosis between both the umbilical arteries known as Hyrtl's Anastomosis (Figure 6). Hyrtl's anastomosis was seen in all the placentae except one which only had one umbilical artery.

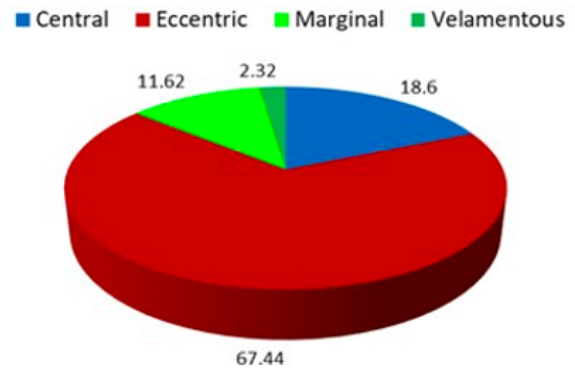


Fig. 2: Distribution of cases according to site of attachment of umbilical cord(n=43)

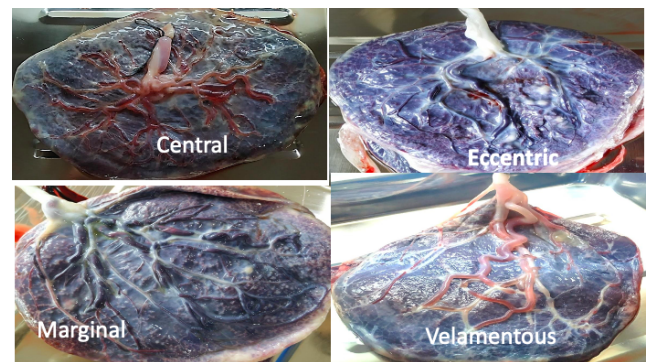


Fig. 3: Types of umbilical cord insertion

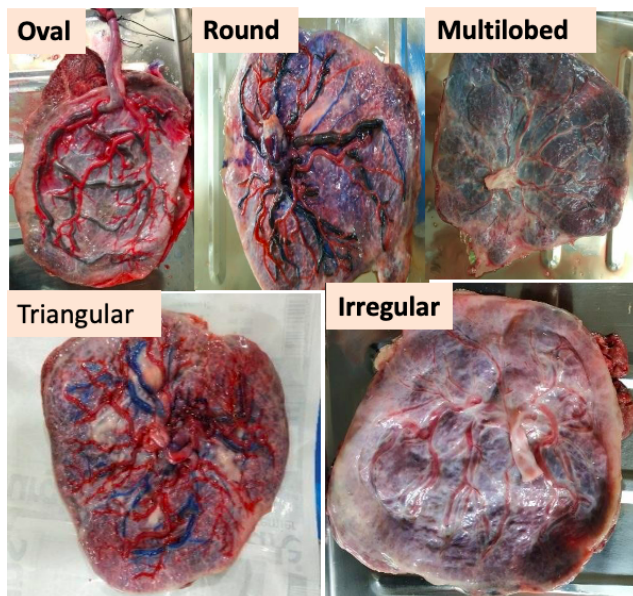


Fig. 1: Shapes of placenta

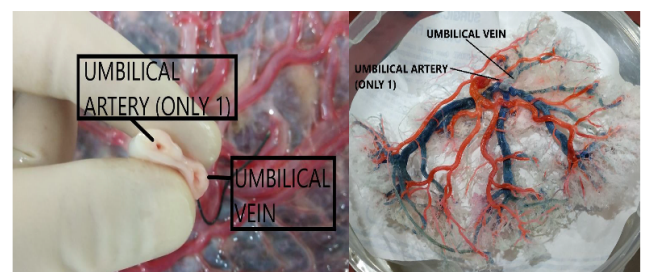


Fig. 4: Umbilical cord & cast showing 2 umbilical vessels

3.1. Vascular pattern

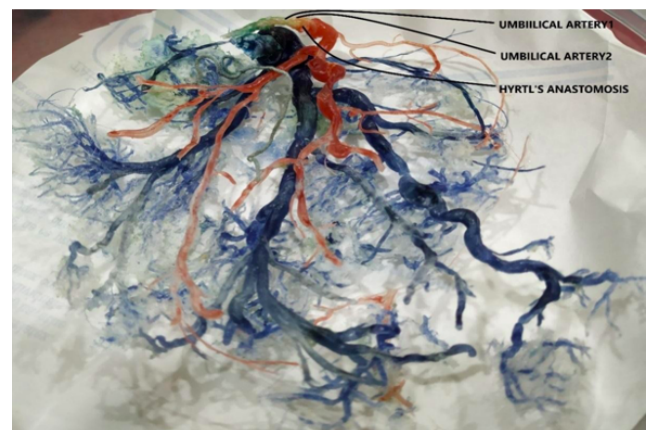
Vascular pattern of placental blood vessel is described as of three types magisterial (monopodial), disperse (dichotomous) and mixed. In the disperse pattern, umbilical vessels undergo successive divisions and rapidly diminished in caliber. In magisterial pattern umbilical vessels give side branches and the caliber of vessel is almost equal up to periphery. In mixed pattern, umbilical vessels show combination of dispersed and magisterial in which umbilical

Table 1: Relationship between shape and vascular pattern of placentae

S. No.	Pattern	Total	Round	%	Oval	%	Irregular	%	Triangular	%
1	Dispersed	4	2	4.65	1	2.32	1	2.32	0	
2	Magisterial	19	6	13.95	11	25.58	1	2.32	1	2.32
3	Mixed	20	8	18.60	10	23.25	0		2	4.65
	Total	43	16	37.20	22	51.16	2	4.65	3	6.97

Table 2: Relationship between shape, vascular pattern of placenta and U.C. insertion in abnormal cases

	Pattern	Total	Round n(%)	Shape of placenta			Eccentric n(%)	U.C. insertion		
				Oval n(%)	Irregular n(%)	Triangular n(%)		Central n(%)	Marginal	Velamentous
GD	Magisterial	2	0	2	0	0	2	0	0	0
M	Mixed	4	2	2	0	0	3	1	0	0
HTN	Magisterial	2	2	0	0	0	2	0	0	0
	Mixed	3	2	1	0	0	3	0	0	0

**Fig. 5:** Cast showing umbilical artery 1 draining into umbilical artery 2 after Hyrtl's anastomosis**Fig. 7:** Cast showing vascular pattern**Fig. 6:** Cast showing Hyrtl's anastomosis

vessel shows successive divisions along with side branches as shown in Figure 7.

3.2. Relationship between shape and vascular pattern of placentae (Table 1)

Relationship between shape and vascular pattern of placentae showed three different types of vascular pattern. A total of 4 placentas showed dispersed pattern, Out of which 2 were round, 1 oval and 1 irregular. Magisterial pattern was seen in 19 placentas out of which 6 were round, 11 oval and 1 each were irregular and triangular. In mixed pattern

category, Out of 20 placentae, 8 were round, 10 were oval and 2 were triangular.

3.3. Relationship between type of U.C. insertion & vascular branching pattern in normal & abnormal placentae

Relationship between type of umbilical cord insertion and vascular branching pattern showed insignificant results in both complicated and uncomplicated placentae. A total of 4 placentae were dichotomous, 19 were monopodial and 20 were mixed. Out of 8 placentae with Central cord insertion, 3 were showing Monopodial vascular pattern & 5 were mixed. Out of 29 placentae with Eccentric cord insertion, 4 were showing Dichotomous vascular pattern, 12 Monopodial & 13 were mixed. Out of 5 Marginal, 3 were Monopodial & 2 were mixed. There was one Velamentous cord insertion which was Monopodial. Hypertension was observed in 2 patients of monopodial and 3 with mixed

branching pattern. Similarly, gestational diabetes mellitus was found in 2 patients of monopodial and 4 patients of mixed branching pattern.

Relationship between shape, vascular pattern of placenta and U.C. insertion in abnormal cases is shown in Table 2.

3.4. Placental blood vessel diameter: Diameter of main umbilical artery 01 & 02 and their first bifurcation

Mean \pm SD of diameter of main umbilical artery 1 and 2 was 0.420 ± 0.145 & 0.357 ± 0.133 respectively and a range between 0.2-0.7 & 0.025-0.68 respectively. On 1st bifurcation of umbilical artery 1, mean \pm SD of right branch was 0.252 ± 0.146 with a range of 0.01 to 0.65 and in left branch, it was 0.372 ± 0.150 with a range of 0.15 - 0.74.

Similarly, in artery 2 on 1st bifurcation, right branch mean \pm SD was 0.118 ± 0.100 (range 0.02 - 0.4) and in left branch it was 0.281 ± 0.090 (range 0.02 - 0.042). On statistical analysis, the difference among artery 1 and artery 2 was found to be statistically significant.

Diameter of main umbilical vein and its first bifurcation along with maximum and minimum end vein diameters.

The difference among right branch and left branch diameter of 1st umbilical vein bifurcation and maximum & minimum end umbilical vein diameter was statistically found to be highly significant ($p < 0.001$).

3.5. Number of bifurcations of placental blood vessels in uncomplicated pregnancies

In the present study, number of bifurcations of placental blood vessels counted in uncomplicated pregnancies were 4 in 3 placentae, 5 in 2 placentae, 6 in 6 placentae, 7 in 6 placentae, 8 in 2 placentae, 9 in 1 placenta, 10 in 1 placenta, and 14 in 3 placentae with a maximum number of bifurcations 17 (in 1 Placenta) and minimum number 3 (in 3 Placentae).

3.6. Number of bifurcations in complicated pregnancies

In the present study, number of bifurcations of placental blood vessels counted in complicated pregnancies were 6 in 3 placentae, 8 in 4 placentae, 9 in 2 placentae with a maximum number of bifurcations recorded were 10 (in 1 Placenta), observed in IUD Placenta and minimum number were 5 (in 3 Placentae), both placentae were procured from hypertensive patients.

4. Discussion

The present study was conducted to assess the morphometry of the placentae i.e. weight, shape, diameter and thickness (both central and peripheral). Besides that the type of umbilical cord insertion and number of blood vessels in the cord were also studied. Corrosion cast of placental blood vessels were prepared and their vascular branching

pattern, vascular bifurcations, vascular diameter of main umbilical artery 1 & 2, main umbilical vein along their first bifurcations and diameters of end blood vessels were studied.

Placenta varies in shape like oval, circular, triangular, irregular, multi-lobular, circumvallate. In the present study out of 40 cases, we observed oval as most common shape in 22(51.16%) followed by round in 16(37.20%) placentae. Only 1 placenta each (2.32%) found to be irregular and multilobed. Gupta Anshu et al¹¹ observed most common shape as round in 60 placentae (60%), followed by oval (24%) and least common was irregular (16%). Sarojamma et al. (1986)¹² observed most common shape as round in 57% followed by oval in 36% and triangular in 7%.

Out of 40 placental specimens, 08 specimens (18.60%) showed central type of cord insertion. Our study is similar to Eastman N.J. & Hellmann L.M. (1966)¹³ who quoted it as 18%. The percentage is higher than Hyrtl J (1870)¹⁴ who reported it as 16% and Gunapriya R. (2001)¹⁵ who reported it in 5%. Sarojamma (1986)¹² & Krone H.A. (1961)¹⁶ reported a higher percentage of 40% and 25% respectively. In 5 specimens (11.62%) we observed battledore/marginal type of cord insertion. Our study is similar to Krone H.A. (1961)¹⁶ who found it in 10% and less than Hyrtl J (1870)¹⁴ who reported it in (19%) cases. Eastman N.J. and Hellmann L.M (1966)¹³ reported a lower percentage of 7%. Velamentous type of cord insertion was observed in 1 specimen (2.3%). This finding is almost similar to Uyanwah et al. (1977)¹⁷ who observed it in 1.6% and less than Sarojamma (1986)¹² who found it in 4% cases. Eccentric type of cord insertion was observed in 29 specimens (67.44%). This finding is closely similar to Eastman N.J. & Hellmann L.M. (1966)¹³ who observed it in 73% specimens. Our finding is higher than the Hyrtl J (1870)¹⁴ and Sarojamma (1986)¹² who observed eccentric type in 54% and 53% specimens respectively Hence, it is concluded that eccentric cord insertion is the most common finding (29 placentae) while the least common is velamentous (01 placenta).

Number of blood vessels in all the umbilical cord specimens (including intrauterine death) was 2 umbilical arteries and 1 umbilical vein (97.68%) except for one placenta from GDM mother on insulin which showed only single umbilical artery & vein (2.32%) Edith. L. Potter (1952)¹⁸ and Keith. L. Moore & T.V.N. Persaud (1973)¹⁹ observed 2 umbilical arteries and 1 umbilical vein in all the specimens. Stephen A. Heifetz (1996)²⁰ quoted super numerary vessels are less common than single umbilical artery. H. Fox (1978)²¹ Hamilton Boyd,²² stated that only 1 artery is present in 1% of umbilical cord.

A large doubt is still present in the placental vascular pattern among different studies. So, we studied the vascular pattern by corrosion cast method for better understanding. The structural organization of the chorionic vasculature

was analyzed from cast models of placentas. The models were generated by the corrosion technique using a dental polymer. Fetoplacental vasculature is composed of large vessels that branches from the umbilical vessels at the umbilical insertion and traverse the chorionic plate. Smaller arteries branch from the chorionic vessels and enter the placenta to constitute the cotyledon vessels or the intra placental vessels that perfuses the cotyledons. The branching pattern of chorionic vessels was defined as disperse for a branching network that courses from a central cord insertion, and magisterial for branching vessels that course from a marginal cord insertion to the opposite edge. Classification of the chorionic arterial network was performed using the dichotomous and monopodial branching pattern. The dichotomous pattern defines a symmetric network of bifurcation in which each vessel branches into new fairly similar daughter vessels. The monopodial network defines a network for vessels in which a main, long mother tube courses for a long distance with an almost constant diameter, with small diameter daughter tubes branching off to the sides. The placental casts demonstrated that umbilical cord insertion was located anywhere between the center and the margins of the placenta. For the central insertion the dichotomous pattern was dominant, whereas for a marginal insertion the monopodial pattern was dominant. We observed the Hyrtl anastomosis in all of the casts which connects the umbilical arteries within the placenta at 5mm from the cord insertion. When one umbilical artery was injected with the red colored casting materials, the same was entering the 2nd umbilical artery via a connection between both the umbilical arteries known as Hyrtl anastomosis. The cast models of this study demonstrated very well the Hyrtl anastomosis between the umbilical arteries (Figure 6).

We have found three types of Vascular pattern in our study. Most common Vascular pattern observed was mixed in 20 placentae, followed by magisterial in 19 placentae and least common pattern was dispersed, in 4 placentae. Gupta Anshu et al²³ and Verma Ranjana et al.² have quoted only 2 types of vascular pattern (Disperse & Magisterial) in their study. While Lakshmi Devi C.K. et al.²⁴ found only disperse pattern in their study.

It was observed that in central cord insertion, the blood vessels bifurcate through 6 to 7 generations in dichotomous pattern with few monopodial ramifications to reach the margins of the placenta and these findings are similar to the findings of Gordon et al.²⁵ In contrast, in marginal cord insertion, 1st bifurcation was dichotomous followed by monopodial pattern and in the end it was dichotomous. Maximum number of bifurcations of blood vessels counted in uncomplicated placentae was 17, and minimum number of bifurcations was 03. Maximum number of bifurcations recorded by Bernad E.S.²⁶ in 2016 was 14. No-one in the available literature has quoted 17 number of bifurcations

which was seen in our study. In complicated pregnancies, maximum number of bifurcations observed was 10 (IUD) and minimum number of bifurcations observed was 05 (HTN).

The diameter of the placental blood vessel cast was measured by vernier caliper. Mean diameter of main umbilical artery 1 and 2 was 4.20 mm and the maximum and minimum diameter ranged between 2mm -7 mm & 0.25mm -6.8 mm, respectively.

Statistically the diameter of umbilical artery 1 & 2 is significant (p value = 0.04), and the diameter of their 1st bifurcations is also Significant (p value for UA1 & UA2 are 0.02 & 0.001). On 1st bifurcation of umbilical artery 1, mean of right branch was 2.52mm with a minimum diameter of 0.1 mm and maximum of 6.5 mm and in left branch, mean was 3.72 mm with a maximum and minimum range of 1.5 mm and 7.4 mm. Similarly, in artery 2 on 1st bifurcation, right branch mean was 1.88 mm (range 0.2 - 4 mm) and in left branch it was 2.81mm (range 0.2 – 4.2 mm). Gordon et al.²⁵ quoted the diameter of 1st arterial bifurcation ranged between 1.13 to 5.36 mm which coincides with our findings. On statistical analysis, the difference among artery 1 and artery 2 found to be statistically significant. In the present study, mean maximum sized end artery 1 was 2.30 mm with a range of 1mm – 5.5 mm and of minimum sized end artery was 1.06 mm with a range of 0.2 – 2.5 mm. Similarly, in end artery 2, maximum sized artery mean was 1.82 mm with a range of 0.5 - 3 mm and minimum sized end artery was 1.02 mm with a range of 0.4 – 1.8 mm. On Statistical analysis, the difference among maximum sized end artery 1 with maximum sized end artery 2 was found to be statistically significant (< 0.05), but minimum sized end artery difference was statistically insignificant (p > 0.05).

The umbilical vein diameter varies from 2mm to 12mm. On its 1st bifurcation, right branch diameter varies from 0.5 to 8.5 mm and left branch diameter varies from 2.2 to 10mm. End vein diameter of right branch ranges between 0.7mm to 6.5mm and of left branch ranges between 0.2mm to 3.5mm. The difference among right branch and left branch diameter of umbilical vein bifurcation and maximum & minimum end umbilical vein diameter was statistically found to be highly significant (p < 0.001).

5. Conclusion

Vascular cast study of the placenta showed three types of patterns. The most common pattern seen was mixed type followed by magisterial & dispersed. Mean Diameter of UA 1 & 2 was 0.42cm & 0.357cm with a range of (0.2-0.7) & (0.025-0.68) respectively. Mean diameter of umbilical vein was 0.661cm with a range of 0.2cm-1.2cm and the mean diameter of their 1st bifurcation, right is 0.385cm (range from 0.05- 0.85) and left is 0.550cm (0.22-1). Maximum number of bifurcations of the blood vessels seen was 17 & minimum number was 3. In most of the specimens, number

of bifurcations seen ranged between 5-7. Statistical analysis of diameter of main umbilical artery 1 and 2 (UA 1 & UA 2) was significant.

6. Source of Funding

None.

7. Conflict of Interest

The authors declare that there is no conflict of interest.

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