Comparison of the Morphology and Morphometry of Placenta from Normal and Assisted Reproduction in Port Harcourt, Rivers State, Nigeria.

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ABSTRACT

The placenta is a fetomaternal organ which connects the developing fetus to the mother. This connection is both structural and functional. This study is aimed at examining the relationship between placental morphology and morphometry in normal and assisted reproduction (ART) in Port Harcourt, Rivers State, Nigeria. A case-controlled descriptive, prospective cross-sectional study was done using placentas from normal and assisted reproduction, with a sample size (n=96. The placenta was obtained immediately after delivery, and the membrane was trimmed off to expose the chorionic plate. Morphologic parameters were recorded, while measurements were taken for morphometric parameters. The means of placental morphometry were determined in normal and assisted reproduction. A statistically significant relationship ($p< 0.05$) was found to exist between morphometric parameters of the placenta in normal and assisted reproduction. At the confidence level ($p< 0.05$), all the morphometric parameters of the placenta, except the number of cotyledons and volume of placenta, showed a significant difference in normal and assisted reproduction. There was no statistically significant difference in the morphology of the placenta in normal and assisted reproduction, except for umbilical cord insertion. Assisted reproduction caused a significant effect on the morphometry of the placenta, umbilical cord insertion and feto-placental ratio. Our study showed that assisted reproduction increases the thickness and diameter of the placenta, but causes a reduction in placental weight and feto-placental ratio. ART increased the incidence of central and velamentous insertion but decreased eccentric and marginal insertion of the umbilical cord. Our study provides baseline data on morphometric parameters of the placenta in normal (spontaneous) and assisted production in Port Harcourt, Rivers State, Nigeria. These findings contribute to the global understanding of the dynamics of the effect of hormonal drugs used in ART.

Key words: Morphology and Morphometry of Placenta, Normal (Spontaneous) Reproduction, Assisted Reproduction (ART).

INTRODUCTION

The placenta is a fetomaternal organ that establishes both the structural and functional connection between the developing fetus and the mother. Arising from the trophoectoderm at the point of implantation, it plays a central role in regulating intrauterine development and shaping individual susceptibility to chronic diseases in adulthood (1). The pre-implantation period represents a critical window during which epigenetic regulatory changes may occur, particularly in response to environmental influences associated with assisted reproduction technology. Such influences include controlled ovarian stimulation, in vitro fertilisation, embryo culture, selective embryo transfer, and hormonal priming, all of which differ significantly from the in vivo environment in oxygen concentration, temperature, cytokines, growth factors, and hormonal levels (2, 3). These differences may introduce stress to the gametes and early embryos, potentially resulting in alterations in placental cellular proliferation and fetal development.

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The pre-implantation embryo is susceptible to environmental changes, making it prone to abnormalities in gene expression and developmental programming that can manifest as prenatal complications, postnatal disorders, or long-term diseases. Evidence indicates that the incidence of placenta previa and other placental abnormalities is significantly higher in pregnancies achieved through assisted reproduction compared to spontaneous conception (4, 5). Further studies have reported metabolic alterations, particularly within lipid pathways, in assisted reproduction pregnancies. Assisted reproduction has also been associated with increased placental growth dynamics mediated by alterations in the imprinting gene network, resulting in modified placental gene expression and signalling pathways (6). These molecular and structural changes contribute to variations in placental phenotype, morphology, and morphometry, particularly through effects on cell differentiation and growth of trophoblastic subpopulations such as glycogen cells, thereby influencing nutrient transport capacity and fetal well-being.

Beyond its role in nutrient and gas exchange, the placenta functions as both an endocrine and immunological organ. It secretes a wide range of hormones that regulate maternal metabolism, physiology, and behaviour throughout pregnancy (7), and it plays a central role in coordinating the remodelling of maternal spiral arteries to ensure adequate uteroplacental perfusion (8, 9). Impairment of this remodelling process has been implicated in major obstetric complications collectively described as the “Great Obstetrical Syndromes,” including pre-eclampsia and fetal growth restriction (10). Additionally, maternal conditions such as diabetes mellitus and hypertension can significantly alter placental morphology and morphometry, thereby affecting fetal and maternal outcomes. At term, the typical human placenta is discoid in shape and measures approximately 20 centimetres in diameter, 2–3 centimetres in thickness, and about 500 grams in weight (11).

Despite the importance of placental structure in clinical outcomes, limited research exists on the effects of assisted reproduction on placental morphology and morphometry in Port Harcourt, Rivers State. This study, therefore, aimed to evaluate the morphology and morphometric characteristics of placentas from normal and assisted reproduction pregnancies, and to investigate the relationships and comparative differences between these groups.

Objectives

1. To determine the morphologic and morphometric parameters of the placenta from spontaneous pregnancies.
2. To determine the morphologic and morphometric parameters of the placenta in pregnancy from assisted reproductive techniques. (ART).
3. To determine the effect of ART on the morphology and morphometry of the placenta.

Justification

The findings of the study will provide essential information on how normal and assisted reproductive pregnancies differ in placental morphology and morphometry. These insights will support developmental and reproductive anatomical scientists, clinical embryologists, forensic experts, and obstetricians and gynaecologists in improving fetal assessment, guiding clinical decision-making, and enhancing overall pregnancy management.

LITERATURE REVIEW

The development of the human placenta begins at the blastocyst stage with the formation of the trophectoderm, which differentiates into distinct trophoblast lineages through interactions with the maternal endometrium (12, 13). The syncytiotrophoblast invades the maternal decidua and erodes maternal sinusoids, allowing maternal blood to fill the lacunae and establish early maternofetal circulation. Cytotrophoblasts proliferate to form primary, secondary, and tertiary villi, while extra-embryonic mesoderm and fetal blood vessels develop within the villous cores. Branching of tertiary villi produces free villi, and anchoring villi arise from cytotrophoblast contact with the decidua, forming the cytotrophoblastic shell (13, 14). As the chorionic villi expand to form the chorion frondosum, multiple villous structures merge into cotyledons, through which approximately 150 millilitres of maternal blood per minute circulate. This arrangement supports efficient oxygen, nutrient, and

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waste exchange, and the placenta, typically attached to the posterior uterine wall near the fundus, displays a granular maternal surface organised into 15–20 lobes called cotyledons (15).

Placentation in vertebrates is understood to have evolved from pre-existing tissues such as the uterus and chorioallantoic membrane, despite vertebrates having a generally conserved body plan (16). As one of the most recently evolved organs, the placenta has arisen independently across various lineages, coinciding with multiple independent occurrences of viviparity, where embryos develop within the reproductive tract and are born live, rather than via egg-laying (17). In mammals, placentas are categorised based on fetal membranes into the choriovitelline and chorioallantoic types (18). The choriovitelline placenta, a vascularized trilaminar yolk sac, functions transiently post-implantation before regressing in most species except rodents and rabbits. The chorioallantoic placenta, derived from the trophectoderm and uterine endometrium, is the primary placental type in mid- to late gestation, showing diverse forms such as diffuse, multicotyledonary, zonary, and discoid or bidiscoid types depending on species morphology (19, 20).

Microscopically, human placentation involves repeated branching of chorionic villi, producing a highly complex arborization pattern (21). This structural complexity is so extensive that even expert pediatric pathologists cannot reliably quantify it. As arborization mirrors the underlying fertility of the maternal environment, placental morphology directly reflects maternal health and influences fetal well-being (22). Possessing distinct maternal and fetal surfaces, the placenta serves as the primary interface for nutrient and gas exchange. Morphologic and morphometric indices correlate strongly with gestational age and birth weight, with placental morphometry and newborn sex demonstrating predictive value for birth outcomes (23). Aberrations in placental morphology accompany pregnancy complications; for instance, pre-eclampsia reduces placental weight, thickness, and diameter, all of which correlate with fetal growth and neonatal outcomes. Moreover, assisted reproduction has been associated with increased placental weight and feto-placental ratio compared to spontaneous pregnancies (24).

Epidemiological evidence further suggests that events in intrauterine life influence long-term susceptibility to chronic disease (1, 25), as early development represents a critical biological window of heightened plasticity. Studies show that placental parameters correlate positively with birth weight, with male and female newborns exhibiting distinct mean values for placental weight, surface area, volume, and thickness. Additional findings reveal reduced placental thickness in pregnancy-induced hypertension and significantly diminished dimensions, including thickness and weight, in ART pregnancies compared with natural conception (15). Other reports document variations in normal placental weight ranges, alongside evidence that vascular abnormalities influence placental thickness and fetal size. Collectively, these findings underscore that ART pregnancies are consistently associated with reduced placental dimensions and altered morphology (15, 26).

METHODOLOGY

Study Design

For the purpose of this study, the cross-sectional descriptive research design was employed to analyse and compare the morphological and morphometric characteristics of the placenta from normal and assisted reproduction in Port Harcourt, Rivers State, Nigeria.

Study Area

The study was conducted in Port Harcourt, Rivers State, Nigeria. Placental samples were obtained from spontaneous (natural) pregnancies and from pregnancies conceived through Assisted Reproductive Technology, provided they met the study's inclusion criteria. The clinical facilities involved were the University of Port Harcourt Teaching Hospital and the Rivers State University Teaching Hospital.

Sample Size Determination

The sample size (n) was calculated using the formula:

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$$n = \frac{Z^2 P(1-P)}{d^2}$$

Where:

Z= Z-score of 1.96 for a 95% confidence interval.

d= Precision or margin of error corresponding to the effect size of 0.1

p= Expected population proportion taken as 0.5

n= Sample size of 96

METHODS OF DATA COLLECTION

The study materials include; placenta collected following delivery for normal and assisted reproduction, measuring tape, weight scale, calibrated plastic bow, calibrated knitting needle and vernier calipers.

The placenta was collected immediately after delivery, washed under running tap water and the fetal membrane trimmed off to expose the chorionic plate. Placenta was examined for completeness, and the morphologic parameters noted, including sharp consistency, colour and insertion of the umbilical cord. The morphometric parameters were measured as follows:

(i) Weight: Then freshly collected placenta was placed on the plastic bow and weighed together with the plastic bow; the weight of both the placenta and the bow was recorded. The weight of the placenta was calculated by subtracting the weight of the plastic bow only from the weight of both the plastic bow and the placenta.

(ii) Volume of Placenta: This was estimated by the water displacement method. A graduated plastic bucket was filled with water to the volume of two litres (2L). The placenta was then immersed into the bowl of water. The increase in volume which represented the volume of water displaced by the placenta was recorded as the volume of the placenta.

(iii) Thickness of the Placenta: This was measured using a long-graduated knitting needle. The placenta was placed on a flat surface, and the needle was used to pierce through the placenta at three points; first at the centre, then at the margin and finally midway between the margin and the centre. The average of the three readings was calculated and recorded as the thickness of the placenta.

(iv) Diameter of the Placenta: The diameter of the placenta was measured using a measuring tape. The maximum diameter representing the longest diameter was measured and recorded. The minimum diameter was also measured and recorded. The average of the two was taken as the diameter of the placenta.

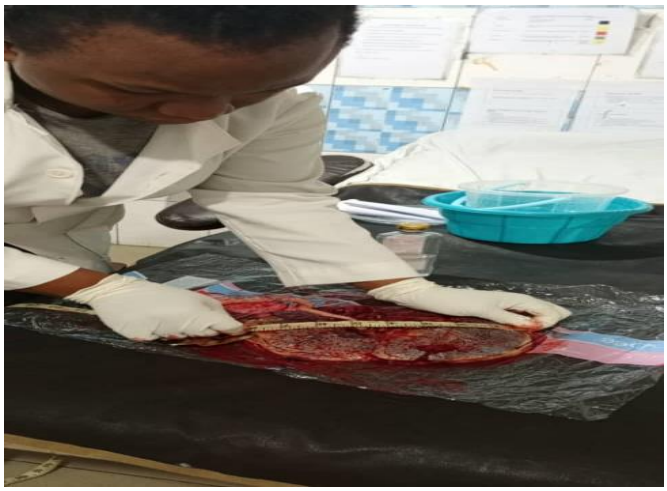

Figure 1: Photograph demonstrating the measurement of placental diameter.

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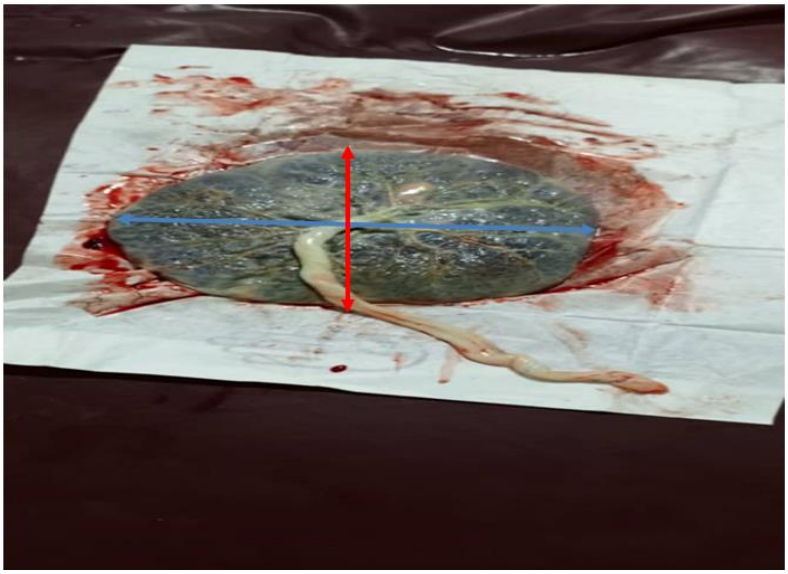

Figure. 2: Photograph illustrating the measurements of Placental Diameter (Red Arrow Minimum Diameter, Blue Arrow Maximum Diameter)

(v) Length of Umbilical Cord: This was measured with a measuring tape and recorded to the nearest centimetre.

(vi)Thickness of the Umbilical Cord: This was measured with the aid of Vainer callipers and recorded to the nearest centimetres.

Morphometric Indices

These were calculated from the morphometric parameters obtained in this study.

The indices include;

(i) Surface Area of the placenta was calculated using the formula.

surface area = $\pi \times \frac{\text{maximum diameter}}{2} \times \frac{\text{minimum diameter}}{2}$

$$= \frac{\pi \times R1 \times R2}{2}$$

Where $\pi = 3.14$ (constant)

R1= = maximum diameter

R2 = minimum diameter

(ii) Feto-placental Ratio: This was determined by dividing the fetal weight by the placental weight.

(iii) Circumference of the placenta: This was determined using the formula;

$$placental\ circumference = \pi \times \frac{maximum\ diameter + minimum\ diameter}{2}$$

Ethical Consideration

This study received approval from the Research Ethics Committee of the University of Port Harcourt before its commencement.

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RESULTS

A paired sample (t-test) for comparison of mean morphometric parameters in normal and assisted reproduction in female and male fetuses, showed that all morphometric parameter except number of cotyledon and volume of placenta had statistical significant difference see table 1 and 2.

Table. 1: paired sample statistics for females.

PARAMETERS		Mean	Std. Deviation	Std. Error Mean	t	df	Sig. (2-tailed)	Inference
Pair 1	**nGA**	38.71	1.03	0.16	14.088	40	0.000	Significant
	aGA	35.80	0.84	0.13				
Pair 2	**nFBW**	3.07	0.25	0.04	7.743	40	0.000	Significant
	aFBW	2.23	0.66	0.10				
Pair 3	**nNCP**	20.00	1.00	0.16	-0.594	40	0.556	Not Significant
	aNCP	20.54	5.71	0.89				
Pair 4	**nDPMAX**	8.49	0.86	0.13	-19.521	40	0.000	Significant
	aDPMAX	21.34	4.16	0.65				
Pair 5	**nDPMIN**	7.38	0.38	0.06	-21.062	40	0.000	Significant
	aDPMIN	15.20	2.36	0.37				
Pair 6	**nDPAV**	7.93	0.52	0.08	-22.182	40	0.000	Significant
	aDPAV	18.71	3.05	0.48				
Pair 7	**nLUC**	14.16	2.95	0.46	-16.252	40	0.000	Significant
	aLUC	39.88	9.93	1.55				
Pair 8	**nTUC**	0.33	0.10	0.01	-21.769	40	0.000	Significant
	aTUC	1.36	0.27	0.04				
Pair 9	**nWP**	0.59	0.06	0.01	4.562	40	0.000	Significant
	aWP	0.49	0.14	0.02				
Pair 10	**nTP**	0.61	0.11	0.02	-24.717	40	0.000	Significant
	aTP	1.90	0.28	0.04				
Pair 11	**nVP**	432.80	34.30	5.36	-0.04	40	0.968	Not Significant
	aVP	433.78	143.36	22.39				
Pair 12	**nSAP**	49.40	6.31	0.99	-16.083	40	0.000	Significant

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	aSAP	253.28	80.54	12.58				
Pair 13	**nFPR**	5.23	0.69	0.11	3.048	40	0.004	Significant
	aFPR	4.68	0.95	0.15				
N		**41**						

Table 2: Paired sample statistics for males.

PARAMETERS		Mean	Std. Deviation	Std. Error Mean	t	df	Sig. (2-tailed)	Inference
Pair 1	nGA	38.92	0.90	0.13	17.995	49	0.000	Significant
	aGA	35.88	0.82	0.12				
Pair 2	nFBW	3.09	0.29	0.04	12.676	49	0.000	Significant
	aFBW	2.19	0.46	0.06				
Pair 3	nNCP	19.88	1.02	0.14	-3.146	49	0.003	Not Significant
	aNCP	22.54	5.74	0.81				
Pair 4	nDPMAX	8.18	0.86	0.12	-22.997	49	0.000	Significant
	aDPMAX	23.67	4.72	0.67				
Pair 5	nDPMIN	7.33	0.48	0.07	-21.212	49	0.000	Significant
	aDPMIN	16.31	2.90	0.41				
Pair 6	nDPAV	7.76	0.54	0.08	-29.022	49	0.000	Significant
	aDPAV	20.19	2.95	0.42				
Pair 7	nLUC	14.20	2.40	0.34	-18.846	49	0.000	Significant
	aLUC	41.07	9.69	1.37				
Pair 8	nTUC	0.33	0.09	0.01	-28.543	49	0.000	Significant
	aTUC	1.44	0.25	0.03				
Pair 9	nWP	0.59	0.06	0.01	4.369	49	0.000	Significant
	aWP	0.51	0.12	0.02				
Pair 10	nTP	0.57	0.10	0.01	-42.266	49	0.000	Significant
	aTP	1.82	0.19	0.03				
Pair 11	nVP	426.30	35.29	4.99	2.268	49	0.028	Not Significant

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	aVP	385.60	119.71	16.93				
Pair 12	nSAP	47.18	6.23	0.88	-21.211	49	0.000	Significant
	aSAP	303.15	84.61	11.97				
Pair 13	nFPR	5.25	0.68	0.10	4.509	49	0.000	Significant
	aFPR	4.41	1.07	0.15				
N		50						

Again, umbilical cord attachment showed a significant association with mode of conception, but no significant association was found with sex. See tables 3 and 4

Table 3: Nature of Cord Attachment by sex in mode of conception

Parameter		Nature of Cord Attachment				**df**	**□2**		Inference
Conception	Sex	Central	Eccentric	Marginal	Velamentous		**cal**	**0.05**	
Normal	female	1	30	17	0	3	3.46	7.81	Not Significant
	male	0	29	31	0				
Assisted	female	8	16	15	2	3	4.43	7.81	Not Significant
	male	11	26	9	4				

Table 4: Nature of Cord Attachment by Mode of Conception in males and females

Parameter		Nature of Cord Attachment				**df**	**□2**		Inference
Sex	female	central	eccentric	marginal	velamentous		**cal**	**0.05**	
Female	normal	1	30	17	0	3	11.4	7.81	Significant Association
	assisted	8	16	15	2				
Male	assisted	11	26	9	4	3	23.9	7.81	Significant Association
	normal	0	29	31	0				

The mean values for morphometric parameters in normal reproduction (spontaneous pregnancies) in female fetuses were observed as follows: number of cotyledons 20, maximum diameter 8.49cm, minimum diameter 7.38cm, Average diameter 7.93cm, Cord length 14.16 Cord thickness 0.33cm, Placental weight 0.59kg, Placenta thickness 0.16cm and Volume of placenta 432.80cm^3. The surface area of the placenta was recorded as 49.0cm^2 and the feto-placenta ratio 5.23. In a male fetus from normal reproduction, our findings for the means of morphometrics of placenta showed: number of cotyledons 19.88, maximum diameter 8.18cm, minimum diameter 7.33cm, Average diameter 7.76cm, Umbilical cord length 14.20cm, umbilical cord thickness 1.44cm, weight of placenta 0.59kg, Thickness of placenta 0.57cm, Volume of placenta 426.30cm. The mean surface area and feto-placental ratio were 47.18cm^2 and 5.25, respectively.

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For female fetuses in ART, the mean morphometric parameters were recorded as follows: number of cotyledons 20.54, Maximum diameter 21.34cm, Minimum diameter 15.20cm, Average diameter 18.71cm, Length of umbilical cord 39.88cm, thickness of umbilical cord 1.36cm, Weight of placenta 0.49kg, Thickness of placenta 1.90cm and Volume of placenta 433.78cm^3. The means of surface area and fetomaternal ratio were 253.28cm^2 and 4.68. For male fetuses from ART, the mean values of morphometric parameters were shown as: number of cotyledon 22.54cm, Maximum diameter of placenta 23.67cm, Minimum diameter 16.31, Average diameter 20.19cm, Length of umbilical cord 41.07cm, Thickness of umbilical cord 1.44cm, Weight of placenta 0.51kg, Thickness of placenta 1.82cm, Volume of placenta 385.60cm^3. The mean of surface area and feto-placenta ratio were noted as 303.15cm^2 and 4.41, respectively.

DISCUSSION

In the present study, the mean weight of the placenta in normal pregnancies for male and female fetuses was recorded at 590 grams (0.59 kg). This value is slightly higher than that reported by Begun et al. (27), which was 417 grams for males and 407 grams for females. Our mean values align with the range reported by Balihallimath et al. (23), which indicated a mean placental weight between 400-1000 grams.

Additionally, we measured the placental thickness and surface area, which were found to be 0.57 cm and 47.8 cm², respectively. These measurements are relatively smaller than those recorded by Begun et al. (27), who reported a thickness of 2.04 cm and a surface area of 226.5 cm² for male fetuses. These discrepancies may be attributed to various environmental, social, and lifestyle factors associated with different geographical locations as well as the study periods. Differences in the level of obstetric care, which can affect the gestational age at delivery, may also play a role.

Our findings indicate a statistically significant difference ($p < 0.05$) in all morphometric parameters of the placenta, except for the number of cotyledons and the placental volume in normal and assisted reproduction cases. Assisted reproductive technology (ART) led to an increase in placental thickness, diameter, and surface area. This is consistent with the report by Manna et al. (28), which showed that placentas from assisted reproduction have increased thickness and a higher incidence of hematoma compared to normal pregnancies. These changes may be influenced by environmental factors that cause epigenetic changes, thereby altering the imprinting gene network and impacting the phenotype. This could affect the development of trophoblastic cells, resulting in an increased number of cells and the deposition of metabolites and connective tissue in ART placentas. Furthermore, the hormonal drugs used during ART may contribute to these findings.

It was observed in the present study that ART resulted in a reduction in placental weight and the feto-placental ratio. This contradicts the findings of Burton et al. (29) and Zhang et al. (6), who reported that placentas conceived through ART had greater weight and feto-placental ratios. The discrepancy might be due to a higher incidence of preterm deliveries in many IVF pregnancies, associated with complications of ART (24), which can lead to low birth weight, especially in cases of pre-eclampsia. Cochrane et al. (30) also noted that ART may be linked to various obstetric complications that require early and premature delivery of the fetus.

Regarding umbilical cord characteristics, our findings indicated that ART increased both umbilical cord thickness and length. This is likely influenced by phenotypic changes caused by epigenetic environmental factors during the critical development period created by in vitro fertilisation and embryo transfer. However, no significant changes in the number of cotyledons or the volume of the placenta due to ART. This may be explained by the fact that the microscopic growth of the placenta involves repeated branching in the chorionic villi, which is dependent on the variable feto-placental environment, often described as "maternal soil" (31). The branching pattern of the chorionic villous tree determines the number and pattern of cotyledons because the cotyledons on the maternal surface of the placenta correspond to the position of the villous trees derived from the chorionic plate in the inter-villous space or blood chamber, as reported by Hupperts (32).

Additionally, a significant correlation was observed between placental volume and diameter, which provides further insight into why ART did not result in any significant changes in placental volume. Regarding the morphology of the placenta, our study showed significant associations, especially concerning umbilical cord insertion types between ART and normal pregnancies for both male and female fetuses. ART was found to

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increase the incidence of central and velamentous umbilical cord insertions while decreasing the occurrences of eccentric and marginal insertions. The increased incidence of central umbilical cord insertion aligns with findings from Yampolsky et al. (31). The abnormalities observed in our study may be explained by potential phenotypic changes arising from epigenetic environmental factors, which may have favoured different modes of umbilical cord insertion.

CONCLUSION

This study demonstrated a significant difference in all placental morphometric parameters, except for the number of cotyledons and placental volume ($p < 0.05$), between normal and assisted reproduction pregnancies, as well as a significant difference in umbilical cord attachment, although no significant variation was found in overall placental morphology. The findings contribute to global knowledge on how hormonal drug regimens used in assisted reproduction and the interval between in vitro fertilisation and embryo transfer influence embryo development and pregnancy outcomes, while also providing baseline data on placental morphology and morphometry in both groups.

RECOMMENDATIONS

Based on these findings, it is recommended that the interval between in vitro fertilisation and embryo transfer be reduced, hormonal treatment protocols for women undergoing assisted reproduction be optimised, and obstetric care for pregnancies achieved through assisted reproductive technology be strengthened to improve gestational age at delivery and overall pregnancy outcomes.

Conflict Of Interest

The authors declare that they have no conflict of interest regarding this publication.

Data Availability

The data supporting the findings of this study are available from the corresponding author upon reasonable request. All datasets have been anonymised to ensure confidentiality and comply with ethical requirements.

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