

MORPHOMETRIC CHARACTERIZATION OF UMBILICAL CORD VESSELS AND NEONATAL OUTCOME

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ABSTRACT

Morphologic and morphometric characterization of the umbilical cord and vessel components could greatly assist in improving adverse maternal and neonatal outcomes. The aim of this study was to evaluate the relationship between morphometry of umbilical cord vessel components and neonatal outcome. A descriptive cross sectional study was conducted on 207 umbilical cords attached to placentae obtained from Victory Maternity Home and Clinic in Kumasi (Ghana) between November, 2013 and October, 2014. Umbilical cord length, diameter, and vessels' diameter were measured with the umbilical cord still attached to the placenta. Neonatal anthropometries were recorded within 24 hours after delivery. The mean \pm SD of vein diameter between neonates of normotensive 3.36 (\pm 0.88) and hypertensive mothers 3.82 (\pm 0.50) showed a significant difference. The body length of neonates with short umbilical cord length was significantly lower ($p < 0.05$) than that of those with long cord lengths. Quantitative analysis indicated a positive linear relationship in umbilical cord and its vessels components with neonatal anthropometry ($p < 0.05$). In conclusion, the morphometry of the umbilical cord and its vessels could predict maternal and neonatal outcome and therefore would be useful in early detection and management of neonatal abnormalities.

KEY WORDS: Neonatal, Umbilical cord, Wharton's jelly, Morphometry, Normotensive.

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INTRODUCTION

Several studies have reported on significant variations in the morphology and morphometry of the placenta and umbilical cord vessels between normal and preeclamptic pregnancies.

Among such studies is Di Naro et al [1], who demonstrated that growth retarded fetuses have smaller umbilical cord cross-sectional area at delivery as compared to normal healthy babies. Pathak et al [2] also observed varied

umbilical cord diameters and areas during gestation, which were attributed to reduced Wharton's jelly rather than the umbilical cord vessels themselves. It has been reported that in preeclamptic pregnancies, the umbilical cord arteries were comparatively thicker than uncomplicated pregnancies [3]. Structurally, the walls of umbilical cord arteries and the vein are identical; the intima has a thin layer of endothelial cells, collagen, elastin and a matrix [4]. Koech et al [3] reported that in preeclamptic cords, there was an increase in thickness of the tunica media and intima in the arteries and higher rate of internal elastic lamina duplication. However, a reduced vessel diameter and wall thickness in both cord artery and the vein in preeclampsia as against normal pregnancies and pregnancies affected by chronic hypertension have been observed [5,6]. These variations were as a result of the umbilical cord vessels adapting to the altered haemodynamic conditions. The differences could also arise due to reduction in vasodilator or increase in vasoconstrictor substances as a result of a pathophysiologic state [6]. The knowledge about the umbilical cord is important because the vessels in the cord are essential components of the foetal circulation. This study was conducted to investigate relationship between foetal indices and umbilical cord vessel morphometry and also to assess the perinatal outcome of these indices among normotensive and hypertensive mothers.

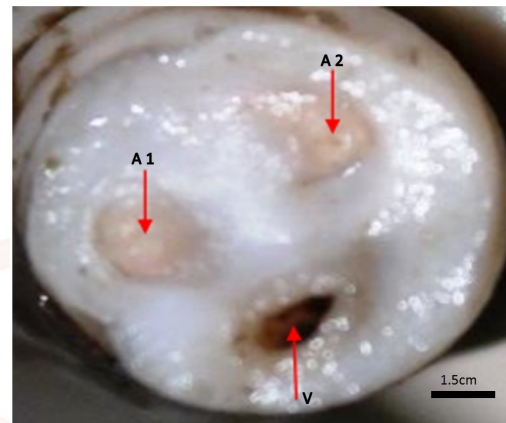
MATERIALS AND METHODS

The study was conducted between November 2013 and October 2014 on delivered placentae, foetal anthropometry and maternal demographic characteristics.

A total of 207 pregnant women who attended prenatal care and delivered at Victory maternity home and clinic were enrolled in the study. Ethical approval was sought from the Committee on Human Research Publications and Ethics, School of Medical Sciences; Kwame Nkrumah University of Science and Technology and the Komfo Anokye Teaching Hospital, Kumasi, Ghana. Also, permission was sought from the facility as well as Informed Consent from prospective participants before sample collection. The placentae

at the maternity unit were collected and washed under running tap water to remove all blood traces. The placentae with their attached umbilical cords were tagged with numbers that corresponded to the numbers indicated in the register for foetal indices. They were then kept in sealed plastic containers filled with 10% formalin at room temperature. The samples were examined and measurements made.

Fig. 1: A photograph of umbilical cord showing three vessels.



(V = Umbilical cord vein, A1 = Umbilical cord artery 1, A2 = Umbilical cord artery 2)

Cross – sectional area and Volume: The cross-sectional areas of the umbilical cord, umbilical arteries, and umbilical vein in a free loop of the umbilical cord were computed using the formula for calculating the surface area of a cylinder with the assumption that the umbilical cord takes the shape of a cylinder. That is: $A = 2\pi r^2L$, where r is the radius and L , the length of cord vessel. Volume of umbilical cord, umbilical arteries and umbilical vein in a free loop of the umbilical cord were computed using the formula for calculating the volume of a cylinder: $V = \pi r^2L$, where r is the radius and L length of cord vessel.

The surface cross-sectional area of the Wharton jelly was computed by subtraction of the total vessels area from the cross-sectional area of the umbilical cord. $AWJ = UCA - (UCVA + A1A + A2A)$, where AWJ is the area of Wharton's jelly, UCA is umbilical cord area, UCV is umbilical cord vein area, $A1A$ is area of artery designated A1 and $A2A$ the area of artery designated A2.

The volume of the Wharton jelly was computed by subtraction of the total vessels volume from volume of the umbilical cord. $VWJ = UCV - (UCV + A1V + A2V)$, where VWJ is the volume of Wharton's jelly, UCV is umbilical cord volume,

UCVV is umbilical cord vein volume, **A1V** is volume of artery designated **A1** and **A2V** the volume of artery designated as **A2**.

Statistical Analysis: Data were analyzed using MS Excel and Graph Pad Prism 5 (GraphPad Software, Inc, San Diego, CA). Student's **t** test was used to compare the quantitative variable means. The adopted level of statistical significance was $p < 0.05$. Spearman correlation matrix and coefficients were used to determine correlations among various umbilical cord measurements with the infant anthropometric measures. Multiple linear regression was used to assess the effect of correlations observed between umbilical cord morphology, vessels morphometry and the infant anthropometric parameters.

RESULTS

Perinatal outcome of normotensive and hypertensive mothers: The morphometric characteristics of the umbilical cord vessel components of the neonates of normotensive and pregnancy induced hypertensive mothers are presented in Table 1. There was a significant difference ($p = 0.018$) in the mean vein diameter between neonates of normotensive (3.36 ± 0.88) and hypertensive mothers (3.82 ± 0.50).

There were no significant differences ($p > 0.05$) in umbilical cord morphometry and foetal indices between Normotensive and PIH mothers with the exception of mean umbilical cord vein diameter which recorded a mean of 3.36 ± 0.88 mm for normotensive and 3.82 ± 0.50 mm for PIH mothers ($p < 0.05$) (Table 1).

Table 1: Perinatal outcome of foetuses of normotensive and PIH mothers.

Variable	Normotensive	PIH	P-value
UC Length (cm)	42.86 \pm 9.08	43.27 \pm 10.13	0.843
UC Diameter (cm)	1.32 \pm 0.23	1.27 \pm 0.20	0.389
UC Vein Diameter (mm)	3.36 \pm 0.88	3.82 \pm 0.50	0.018
A1 Diameter (mm)	2.05 \pm 0.60	2.23 \pm 0.61	0.201
A2 Diameter (mm)	1.97 \pm 0.58	1.91 \pm 0.68	0.635
Area of WJ (cm ²)	80.51 \pm 38.69	67.46 \pm 37.95	0.135

Data are expressed in Mean \pm Standard Deviation with significant difference at p -value < 0.05 .

Spearman correlation matrix of foetal indices and umbilical cord and vessel morphometry: Gestational age showed a significant correlation with the artery designated as **A2** volume ($r = 0.153$, $p < 0.05$) and its area ($r = 0.152$, $p < 0.05$). Birth weight had significant positive correlation with the following; umbilical cord diameter ($r = 0.178$, $p < 0.05$), umbilical cord volume ($r = 0.162$, $p < 0.05$), and the volume of Wharton's jelly ($r = 0.174$, $p < 0.05$).

Table 2: Spearman correlation matrix between foetal indices and umbilical cord vessels morphometry.

	GA	BW	BL	HC	AC	UCL	UCD	UCVD	A1D	A2D	UCV	UCA	UCVV	UCVA	A1V	A1A	A2V	A2A	VWJ	AWJ
GA	1																			
BW	0.041	1																		
BL	0.079	0.360†	1																	
HC	-0.019	0.415†	0.111	1																
AC	0.122	0.463†	0.134	0.460†	1															
UCL	0.063	0.045	0.221†	-0.035	0.084	1														
UCD	0.049	0.178*	0.077	0.077	0.168*	0.07	1													
UCVD	0.074	0.103	0.011	0.048	0.018	0.075	0.275†	1												
A1D	0.099	-0.025	-0.031	-0.114	0.013	0.115	0.191†	0.657†	1											
A2D	0.135	0.014	-0.012	-0.005	0.023	0.095	0.164*	0.619†	0.830†	1										
UCV	0.051	0.162*	0.169*	0.056	0.176*	0.573†	0.834†	0.269†	0.211†	0.196†	1									
UCA	0.057	0.132	0.194†	0.033	0.158*	0.782	0.641†	0.228†	0.200†	0.187†	0.953†	1								
UCVV	0.088	0.099	0.095	0.026	0.045	0.467†	0.275†	0.906†	0.622†	0.584†	0.476†	0.522†	1							
UCVA	0.093	0.103	0.137*	0.022	0.061	0.652†	0.257†	0.774†	0.559†	0.528†	0.564†	0.655†	0.965†	1						
A1V	0.109	0.004	0.094	-0.118	0.034	0.595†	0.184†	0.553†	0.833†	0.698†	0.454†	0.552†	0.741†	0.786†	1					
A1A	0.109	0.018	0.108	-0.099	0.05	0.656†	0.183†	0.528†	0.791†	0.665†	0.487†	0.600†	0.744†	0.809†	0.994†	1				
A2V	0.153*	0.051	0.137*	-0.016	0.049	0.588†	0.155*	0.502†	0.649†	0.809†	0.438†	0.542†	0.694†	0.749†	0.860†	0.866†	1			
A2A	0.152*	0.067	0.145*	-0.006	0.069	0.641†	0.154*	0.478†	0.615†	0.768†	0.466†	0.582†	0.694†	0.765†	0.856†	0.872†	0.994†	1		
VWJ	0.048	0.174*	0.176*	0.069	0.183†	0.530†	0.846†	0.13	0.081	0.062	0.980†	0.921†	0.333†	0.429†	0.329†	0.367†	0.315†	0.348†	1	
AWJ	-0.008	0.13	0.161*	0.061	0.140*	0.415†	0.649†	-0.305†	-0.370†	-0.397†	0.743†	0.701†	-0.098	0.03	-0.081	-0.031	-0.09	-0.044	0.843	1

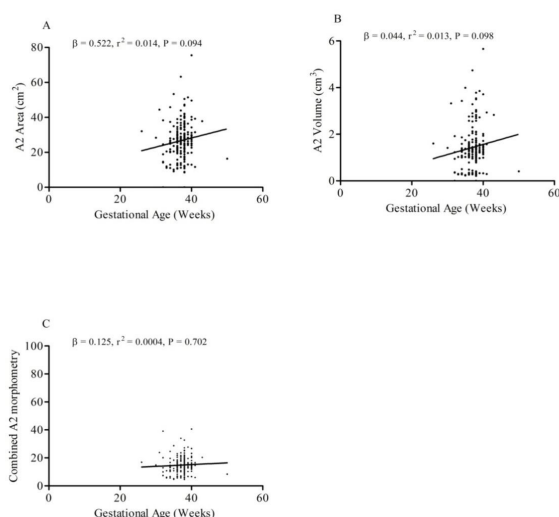
† = $p < 0.0001$; † = $p < 0.001$; * = $p < 0.05$

Body length also showed significant positive correlation with the umbilical cord length ($r = 0.221$, $p < 0.01$), umbilical cord area ($r = 0.194$, $p < 0.01$), umbilical cord volume ($r = 0.169$, $p < 0.05$), the umbilical cord vein area ($r = 0.137$, $p < 0.05$), A2 area ($r = 0.145$, $p < 0.05$) and its volume ($r = 0.137$, $p < 0.05$), the area and volume of Wharton's jelly with $r = 0.161$ and $r = 0.176$, $p < 0.05$ respectively. Significant correlation was observed between abdominal circumference and umbilical cord diameter ($r = 0.168$, $p < 0.05$) as well as umbilical cord area ($r = 0.158$, $p < 0.05$) and volume ($r = 0.176$, $p < 0.05$). It also correlated with the Wharton's jelly area ($r = 0.140$, $p < 0.05$) and volume ($r = 0.183$, $p < 0.01$). The area of Wharton's jelly showed strong significant negative correlation with the umbilical cord vein diameter ($r = -0.305$, $p < 0.001$), the A1 diameter ($r = -0.370$, $p < 0.001$) and A2 diameter ($r = -0.0397$, $p < 0.001$).

Linear regression analysis of foetal indices with umbilical cord and vessel morphometry:

The correlation observed between A2 area and the volume with GA in the Spearman correlation matrix (Table 2) was analyzed using linear regression. A2 area showed no relation with GA ($\beta = 0.522$, $r^2 = 0.014$, $p = 0.094$) (Fig.2A). Similarly A2 volume did not show significant linear relation with the GA ($\beta = 0.044$, $r^2 = 0.013$, $p = 0.098$) (Fig.2B). The combined result of these artery morphometry also showed no significant relation with GA ($\beta = 0.125$, $r^2 = 0.0004$, $p = 0.702$) (Fig.2C).

Fig. 2: Linear regression graph of A2 area, volume and the combined morphometry against gestational age.



Umbilical cord diameter, volume and Wharton's jelly volume correlated significantly with birth weight. Linear regression analysis of these umbilical cord measurements yielded the following results; the UCD showed significant linear relation with BW ($\beta = 0.071$, $r^2 = 0.022$, $p = 0.033$) (Fig. 3A). UCV exhibited significant linear relation with BW ($\beta = 9.351$, $r^2 = 0.028$, $p = 0.016$) (Fig. 3B). Significant linear relation was also observed between VWJ and BW ($\beta = 9.165$, $r^2 = 0.030$, $p = 0.013$) (Fig. 3C). The cumulative effect of these UC quantitative indices on birth weight was statistically significant ($\beta = 6.196$, $r^2 = 0.008$, $p = 0.010$) (Fig. 3D).

Fig. 3: Linear regression graph of UCD, UCV, VWJ and the combined umbilical cord morphometry against BW.

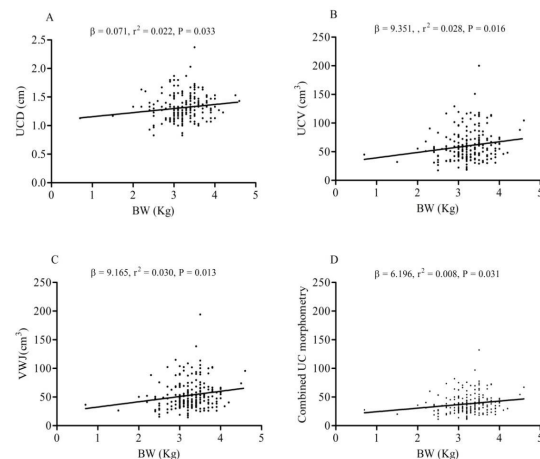
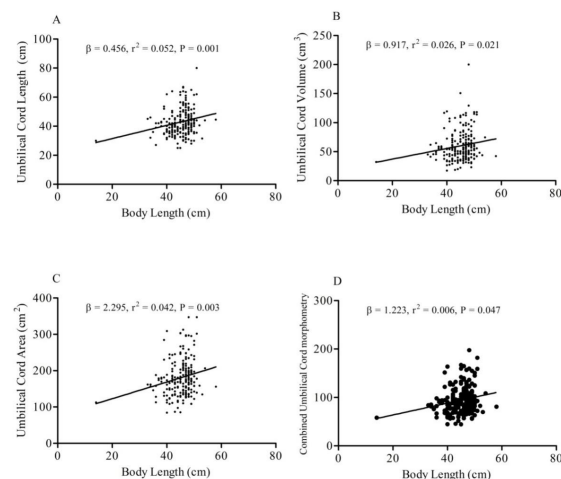


Fig. 4: Linear regression graph of UCL, UCV, UCA and combined morphometry against BL.



The correlations observed between UCL, UCV and UCA with BL were also investigated using linear regression analysis. UCL showed significant linear relation with BW ($\beta = 0.456$, $r^2 = 0.052$, $p = 0.001$) (Fig. 4A). There was a significant relation between UCV and BL ($\beta = 0.917$, $r^2 = 0.026$, $p = 0.021$) (Fig. 4B). Linear relation

observed between UCA and BL was significant ($\beta = 2.295$, $r^2 = 0.042$, $p = 0.003$). The combined effect of these umbilical cord measurements on body length showed significant linear relationship ($\beta = 1.223$, $r^2 = 0.006$, $p = 0.047$).

The volume and area of umbilical cord artery (A2) though the volume alone showed no significant relation with body length ($\beta = 0.017$, $r^2 = 0.008$, $p = 0.203$) (Fig. 5A). However, A2 area showed significant relation with body length ($\beta = 3.194$, $r^2 = 0.019$, $p = 0.046$) (Fig. 5B). The overall effect of these A2 measurements did not show significant relation with body length ($\beta = 0.168$, $r^2 = 0.003$, $p = 0.289$) (Fig. 5C).

Fig. 5: Linear regression graph of A2 volume, area and combined A2 morphometry against body length.

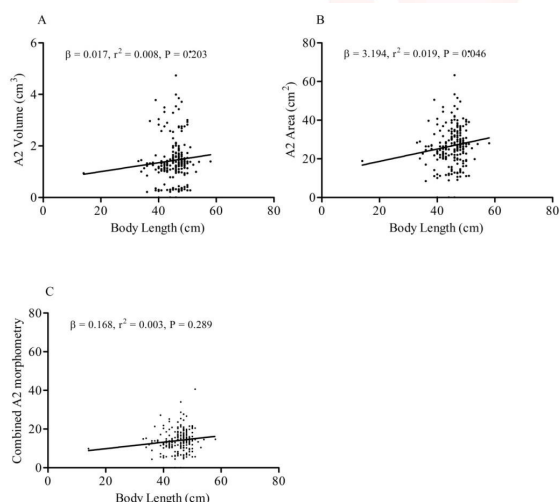
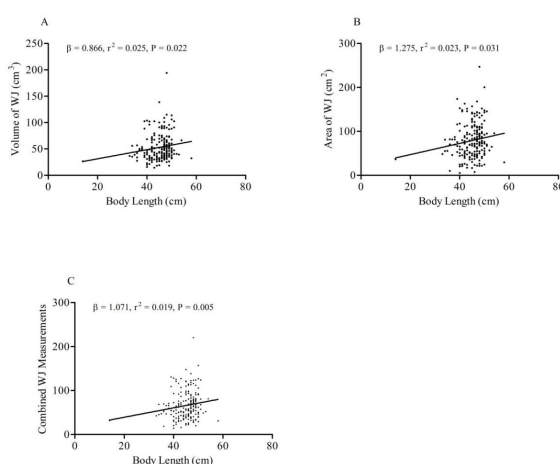


Fig. 6: Linear regression graph of volume, area and combined measurements of WJ against body length.



Linear regression analysis of Wharton's jelly volume and area against body length supported the significant correlations observed between these measurements in the Spearman correlation matrix. Wharton's jelly volume showed strong linear relation with the body length

($\beta = 0.866$, $r^2 = 0.025$, $p = 0.022$) (Fig. 6A), and of Wharton's jelly area with body length ($\beta = 1.275$, $r^2 = 0.023$, $p = 0.031$) (Fig. 6B). Similarly, a significant linear relation was observed between the combined effect of the Wharton's jelly volume and area with body length ($\beta = 1.071$, $r^2 = 0.019$, $p = 0.005$) (Fig. 6C).

When the correlations between UCD, UCV and UCA with AC were further analyzed, it was observed that UCD showed significant relationship with the AC ($\beta = 0.009$, $r^2 = 0.019$, $p = 0.048$) (Fig. 7A). The UCV significantly related with the AC ($\beta = 1.261$, $r^2 = 0.028$, $p = 0.017$) (Fig. 7B). The linear relationship between the UCA and AC was significant ($\beta = 2.623$, $r^2 = 0.031$, $p = 0.0116$) (Fig. 7C). The combined effect of the UC measurements, however, showed no significant linear relationship with abdominal circumference ($\beta = 1.298$, $r^2 = 0.003$, $p = 0.172$) (Fig. 7D).

Fig. 7: Linear regression graph of umbilical cord diameter, volume, area and combined umbilical cord morphometry against AC.

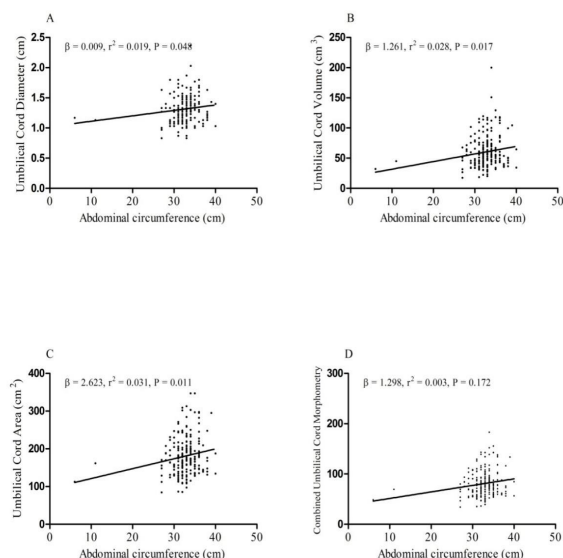
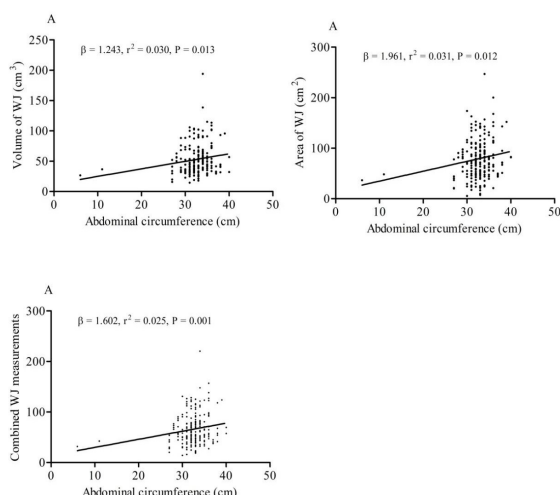


Figure 8 illustrates the linear relationships between Wharton's jelly volume and area with abdominal circumference. A significant linear relationship was observed between Wharton's jelly volume and abdominal circumference ($\beta = 1.243$, $r^2 = 0.030$, $p = 0.013$) (Fig. 8A), likewise between area of Wharton's jelly and abdominal circumference ($\beta = 1.961$, $r^2 = 0.031$, $p = 0.012$) (Fig. 8B). The cumulative effect of the Wharton's jelly parameters measured exhibited strong statistically significant relation with abdominal circumference ($\beta = 1.602$, $r^2 = 0.025$, $p = 0.001$) (Fig. 8C).

Fig. 8: Linear regression graph of volume, area and combined measurements of WJ against Abdominal circumference.



The umbilical cord length was grouped into short (< 40 cm), normal (40 ≤ 70 cm) and long (> 70cm). Out of the 207 umbilical cords studied, 39.62% (82) were short, 59.90% (124) normal and 0.48% (1) long cords. Table 3 compares the perinatal outcome characteristics of the study population stratified by umbilical cord length. Significant difference was observed in body length of fetuses with short and normal umbilical cord lengths (43.95 ± 5.15 and 45.82 ± 3.96, p < 0.05).

Table 3: Umbilical Cord Length and outcome characteristics of study population.

	Short UC (N = 82)	Normal UC (N =124)	Long UC (N = 1)
Maternal Age(yrs)	26.74 ± 5.36	27.35 ± 6.33	32.00 ± 0.00
Delivery Weight (kg)	69.39 ± 8.59	71.48 ± 10.14	68.00 ± 0.00
Parity	2.66 ± 1.76	2.68 ± 1.69	3.00 ± 0.00
Gestational Age (weeks)	36.98 ± 1.88	37.14 ± 2.60	40.00 ± 0.00
Birth Weight (kg)	3.20 ± 0.46	3.25 ± 0.48	3.30 ± 0.00
Body Length (cm)	43.95 ± 5.15*	45.82 ± 3.96	51.00 ± 0.00
Placental Weight (g)	586.20 ± 99.59*	620.60 ± 99.60	900.00 ± 0.00
Chorionic Plate Area(cm²)	92.97 ± 14.37	94.87 ± 15.81	103.70 ± 0.00
BW/PW Ratio	5.58 ± 1.11	5.34 ± 0.99	3.67 ± 0.00

Data are expressed in Mean ± Standard Deviation with p-value, * = P<0.05.

DISCUSSION

Morphologic and morphometric characterization of the umbilical cord components could greatly assist in improving perinatal outcome. With the exception of head circumference, this study found significant relationships between umbilical cord vessel morphometry and all other foetal measurements such as body length, abdominal circumference and birth weight.

Recent advancement in ultrasound technology has enhanced the study of morphometric variation of the umbilical cord vessels association with foetal outcome at birth [7]. For instance, evaluation of umbilical cord artery impedance to blood flow helps in identifying fetuses vulnerable to growth and developmental disorders [8]. The umbilical cords studied were all found to possess three cord vessels namely one vein and two arteries designated as A1 and A2 (Figure 1). A unique observation in this present study is that foetal head circumference and the first artery (designated A1) showed no correlation with any of the umbilical cord components' morphometry. This could possibly indicate that almost all the contribution of the umbilical cord components in respect of the rate of transport, diffusion, distribution and exchange of materials necessary for healthy growth and development are affected by reduction of the umbilical blood stream, with a foetal hypo-perfusion. The maintenance of these haemodynamic processes lead to the stabilization of the vascular and umbilical cord structural pathological modifications and to their constant association [9]. Notwithstanding, the cross-sectional area and the volume of the second artery (Labeled A2) significantly correlated with the gestational age. This is in line with the findings of Togni et al. [10] in which a statistically significant correlation was found between the cross – sectional area of the umbilical cord arteries and gestational age.

This study showed that umbilical cord diameter, volume and the volume of Wharton's jelly showed positive significant correlation with birth weight. Birth weight measures the nutritional status of neonate therefore the current finding explores the possibilities of the umbilical cord influencing birth, realizing the metabolically active role it plays when placental sufficiency is achieved. The Wharton's jelly is the supporting connective tissue of the vessels and facilitates diffusion through its interconnected cavities of water and growth metabolites to and from the umbilical cord vessels and the amniotic fluid [8,11-13].

This observation is also in agreement with a study conducted by Raio et al [9,14] in which both umbilical cord diameter and area correlated

with foetal anthropometric parameters. Morphological studies into umbilical cord structure have observed that umbilical cord in the face of foetal intrauterine growth restriction and hypertensive disorders with normal umbilical artery Doppler parameters exhibited reduced total vessel area and Wharton's jelly area in comparison with normal fetuses [15-18]. However, significant difference in the vein diameter was observed between the neonates of normotensive mothers and pregnancy induced hypertensive mothers respectively, with the PIHs having larger vein diameter. This observation contradicts previous reports in which significant reduction in diameters of the umbilical cord and vessels were recorded [19]. This morphological variation of the umbilical cord vein could be an indication of some important postnatal and foetal haemodynamic deficiencies [20]. This is because Jarvis et al. [21] stated that altered endothelial cell function is a key factor associated with vascular disorders which is crucial in foetal growth and development. These authors also observed, particularly in pregnancies affected by gestational diabetes and preeclampsia, a dysfunction of the umbilical cord vein endothelia. Therefore the larger diameter of the cord vein could be as a result of reduced or atrophy of the vein endothelial cells. It has been reported that umbilical vein varix should be considered a risk factor for poor perinatal outcome, as such whenever there is evidence of umbilical cord varix, a careful search for evidence of other anomalies are necessary [3].

Abdominal circumference measurement which is among the four pillars of biometric measures in the evaluation of foetal size actually reflects abdominal visceral development. Abdominal circumference was found to have significant correlation with umbilical cord diameter, volume, area, and amount and volume of Wharton's jelly. This relationship suggests that abdominal circumference is a strong signal for foetal growth. Available evidence suggests that the size of abdominal circumference has some effect on clinical management decisions such as the need for series of ultrasonographic measurements, foetal monitoring and/or delivery [22].

Foetal body length was found to show a strong positive correlation with umbilical cord length,

area and volume as well as umbilical cord vein area, A2 area and volume and the Wharton's jelly area and volume. Furthermore, a significant difference in body length was observed between neonates with short and normal umbilical cord lengths (Table 3). This study is of the view that in addition to the tension theory as have been reported [22-24], genetic factors undoubtedly influence the determination of umbilical cord length. Indeed, it has been established that insulin-like growth factor I (IGF-I) and insulin-like growth factor binding protein 3 (IGFBP-3) play very important roles in foetal growth during pregnancy [25]. Various studies have reported a relationship between umbilical cord blood IGF-I and other measurements of foetal growth including birth length, crown-rump length, ponderal index and placental weight [26-29]. Various parameters to evaluate foetal development continue to be sought. Among them, vascular architecture of the umbilical cord which is not only fascinating in terms of morphology, but also from the functional interpretation perspective. This suggests a direct participation of the umbilical cord in foetal metabolism and further implies that its architecture relates to the nutritional status of the foetus [30].

The body length relation with the area and volume of the umbilical cord vessels manifests the haemodynamic state of umbilical cord blood flow velocity. It has been found that in cases of continuous diminution in the flow velocity of umbilical cord blood with increased foetoplacental obstruction, structural alteration in the umbilical cord vessel is induced. And as a compensatory mechanism for the insufficient transfer of nutrient, foetal growth velocity is significantly decreased [8].

CONCLUSION

In this study, umbilical cord vein diameter was larger in neonates of pregnancy induced hypertensive mothers. Therefore systematic prenatal monitoring of the haemodynamics of foetoplacental circulation could reduce the incidence of morphological alterations in the new-born babies of PIH mothers. Also neonates with short umbilical cords showed significantly lower placental weight and body length as compared to

their normal counterparts. Umbilical cord length, volume, area and Wharton's Jelly content correlated significantly with of body length, abdominal circumference and birth weight and may influence their development. These quantitative data on the umbilical cord and placenta provide baseline values for further studies.

Conflicts of Interests: None

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