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Small Estimated Placental Volume Predicts Low Birthweight

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Small Estimated Placental Volume Predicts Low Birthweight

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Abstract

Small Estimated Placental Volume Predicts Low Birthweight

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The objective of the study was to validate Estimated Placental Volume (EPV) in a population of patients at Yale-New Haven Hospital (YNHH) across a range of gestational ages, and to evaluate the association between small EPV and low birthweight (BW). From 2009 to 2011, 366 patients at YNHH received ultrasound scans between 11+0 to 38+6 weeks gestational age (GA) to measure placental dimensions from 2009 to 2011. EPVs were calculated using a previously validated convex-concave shell equation. An EPV vs GA best fit curve was generated. The relationship between EPV and BW was analyzed. Subgroup analyses were performed to evaluate differences between study participants who delivered at YNHH, and those who did not. Analysis of EPV versus gestational age revealed a parabolic curve with the following best fit equation: $EPV = (0.372\ GA - 0.00364GA^2)^3$. One hundred seventy four of the 366 women who underwent EPV measurement delivered at Yale-New

Haven Hospital (YNHH) and had their infants' BWs recorded. The remaining patients delivered at outlying hospitals, where BWs were not available to the investigators. However, parabolic EPV GA curves generated from these two patient populations were superimposable. YNHH patients with an EPV in the bottom 50th percentile had 2.42 times the odds of having a newborn with a BW in the bottom 50th percentile (95% CI 1.27 – 4.68). Microscopic evaluation of two placentas corresponding to the smallest EPV outliers revealed significant placental pathology. We conclude that placental volume increases throughout gestation and follows a predictable parabolic curve. Very low EPV measurements are associated with low BWs. Therefore, EPV may be useful as a screen to identify women who are carrying fetuses who are at risk for intrauterine growth restriction.

For Mom and Dad.

Acknowledgements

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To my family – despite my years of medical training, I can't quantify the effect your love and support have had on my wellbeing (perhaps I will know when I am an attending). **Mom** and **Dad**, thank you for your unconditional support of my dreams from the very start, from when I decided I wanted to be a red firetruck (age 3) to a Radiologist (age 26). This means more to me than you can ever know. Dad, I am so sorry you are no longer with us, but your love is undying (and I know you would be proud). **Tanya L.**, thank you for being the best big sister anyone could hope for. Thank you for teaching me the importance of having a gentle bedside manner when explaining imaging studies to patients – I will carry this with me always. **Aunt Darlene**, **Uncle Kevin**, and **Uncle Butch**, thank you for the years of love and support (and fun-filled Thanksgiving celebrations). **Grandma**, I am sorry you could not see me complete this milestone, but your quest for scholarship continues to inspire and motivate me. To **Dr. Leah I., Gregory I.**, **Anna I.**, and **Michael I.** – thank you for warmly accepting me into your family, and for providing a peaceful retreat to rest, think, and work. And finally, to my dearest **Dr. Alex I.** – we did this together! You listened patiently while I talked *at* you for hours about MRI and ultrasound physics. You insisted on keeping my ever-

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"  *[placenta] is the bundle of life."*
-Ancient Egyptian text (3000 B.C.)

1

Introduction

Role of the Placenta in Pregnancy

It is well established that the placenta plays several vital roles during pregnancy, and is essential for nutrient and oxygen transfer between mother and fetus.[1] Placentas that are small for gestational age are associated with intrauterine growth restriction (IUGR), intrauterine fetal demise (IUFD), and other complications.[2-7] The relationship between small placental size and fetal complications was explored by Wolf *et al* in 1989[8]. The authors performed a longitudinal study on 18 pregnant patients between 16 and 20 weeks gestation, and estimated placental volume and fetal weight by ultrasound at regular intervals. In normal fetuses, a sigmoid relationship was observed between placental volume and gestational age, as well as fetal growth and gestational age. Eleven patients experienced adverse fetal outcomes, including fetal distress requiring Cesarean-section, fetal death, or birthweight below the 10th percentile. For all of these patients, the placental vs gestational age growth curve demonstrated restricted placental growth. The authors concluded that placental growth restriction preceded fetal growth restriction and adverse events.

Physiologically, these small placentas were unable to meet the metabolic needs of the growing fetuses. [8]

Just as small placentas are linked to adverse fetal outcomes, Eskild *et al* showed that a placenta that are significantly larger than average are also associated with poor transition to extrauterine life.[9] The authors performed a population study of 522,360 singleton pregnancies in Norway between 1998-2008. They calculated the ratio of placental weight to birthweight, and divided this data set into quartiles. Higher placental weights, as well as high placental weight:birthweight ratios were associated with Apgar scores ≤7. When they compared the highest and lowest placental weight: birthweight quartiles, they found that infants with a higher ratio had 1.65 times the odds of having an Apgar score ≤7 (95% CI 1.57–1.74). Eskild *et al* hypothesized that in these cases, chronic intrauterine hypoxia causes compensatory increases in placental size, which in turn contributes to lower Apgar scores.[9]

Shehata *et al* further examined the relationship between the placental weight:birthweight ratio and fetal outcomes.[10] In a retrospective study, they divided their data set into three groups: high, normal, and low placental weight:birthweight ratios. In addition to reproducing the Eskild *et al* result that high ratios were associated with Apgar scores <7 [9], they also found that high ratios were associated with breech presentation, need for Cesarean-section, and NICU admission. Conversely, low placental weight: birthweight ratios were associated with lower rates of these adverse outcomes.[10]

Of note, with the exception of placental volume, many placental parameters do not impact placental weight or birthweight. Haeussner *et al* examined whether various physical characteristics of the placenta – diameter, thickness, roundness, shape, and cord insertion – were associated with placental weight. After analyzing these parameters in 418 placentas

from uncomplicated pregnancies, they concluded that variations in shape did not impact placental weight or birthweight.[11]

Ultrasound Methods for Assessing Placental Volume

3D Ultrasound

Given the association between extremes in placental size and fetal complications, physicians developed methods to use ultrasound to measure placental volume. Wolf *et al.* first described 3D placental ultrasound in 1989.[12] They imaged *n* parallel slices of the placenta separated by a given distance (d_n), and measured the corresponding area of each slice (A_n). They also recorded the angle of incidence of the ultrasound beam onto the slices (q). Using these parameters, they used the following rectangular model to calculate the placental volume (PV):

$$PV = \{1/3(A_1) + 1/2\,(d_1)(A_1) + ½\,(d_1 + d_2)(A_2)) + \ldots + 1/2\,(d_{n-2} + d_{n-1})(A_{n-1}) + 1/2\,(d_{n-1})(A_n) + 1/3(A_n)\}\sin(q)$$

Their equation could be applied to placentas up to 26 weeks gestation. It was challenging to assess the volume of posterior placentas after 26 weeks because the relatively large fetus cast an acoustic shadow on its placenta.[12]

As an alternative to the multiplanar method, several authors described a method for using Virtual Organ Computer-aided Analysis (VOCAL) and eXtended Imaging VOCAL (XI VOCAL) to measure placental volume.[13-15] Nowak *et al* compared multiplanar and VOCAL methods for measuring the placentas of fetuses between 7 and 10 weeks gestation. They found a strong relationship between placental volume and gestational age, and reported

that there was a strong agreement between multiplanar and VOCAL methods of placental ultrasound.[14] Despite the strengths of 3D ultrasound, there are several complications associated with this modality: many 3D ultrasound machines require specialized software for image processing, 3D reconstruction of the placenta, and calculation of the placental volume.

2D Ultrasound

In an effort to overcome the challenges associated with 3D ultrasound, Costantini *et al* studied whether they could use 2D ultrasound to predict low placental weight in utero.[16] In 95 patients with high-risk pregnancies between 19 and 23 weeks gestation, they measured the maximum placental dimension. High-risk pregnancies were defined as having significant IUGR or preeclampsia. Using a cut-off of 10 cm for maximal dimension, they found that small placentas were not associated with a placental weight less than the 10th percentile. Since the false-positive rate was 25.5%, the authors concluded that placental length was not an effective screening tool for low placental weight. [16]

Although placental length was not a good predictor of placental weight in high-risk populations, in 2012, McGinty *et al* aimed to determine whether measuring both placental length and thickness could be used to predict whether a pregnancy would result in an SGA or AGA infant among low-risk patients.[17] They measured placental length and thickness in 520 low-risk pregnancies, and correlated these with birthweight percentiles upon delivery. Neither the placental thickness, nor the ratio between placental thickness and length, were associated with birthweight. However, placental length less than the 10th percentile was associated with 2.8 times the odds of an SGA infant (95% CI 1.1 – 6.9).[17] In an analysis of 1909 pregnancies, Schwartz *et al* also found that placental thickness and diameter were much

smaller in pregnancies resulting in SGA infants. Therefore, both groups of authors recommended placental ultrasound as a means to evaluate risk of SGA infants.[18]

Comparison of 2D and 3D Ultrasound

While 2D ultrasound requires fewer computational and physical resources than 2D ultrasound, according to Riccabona *et al*, using 2D images to estimate volumes of irregular and ellipsoid structures is associated with an error of 10-20%.[19] In light of this, they evaluated whether 3D ultrasound could more accurately measure volumes than 2D ultrasound. Using a 3D ultrasound scanner that utilized the multiplanar method, they measured the volume of balloon phantoms of various sizes and shapes by placing the probe on the phantoms directly, and then by submerging them in a bath of water and cornstarch (to mimic the echogenicity of tissue). They demonstrated that 3D ultrasound had similar accuracy to 2D ultrasound when measuring elliptical objects, but was markedly more accurately when measuring irregularly-shaped objects. Since the placenta is irregularly shaped, 3D ultrasound was a promising tool for measuring placental volume.[19]

Higgins *et al* compared the accuracy of 2D and 3D ultrasound during third trimester pregnancies in predicting placental volume (measured after delivery).[20] They used the ellipse and shell technique for assessing placental volume with 2D ultrasound, and the multiplanar approach for assessing placental volume with 3D ultrasound. They found good agreement between the two methods, and concluded that they both correlated well with placental volume ex vivo (with 3D ultrasound being more accurate).[20]

Several mathematical models that have been shown to improve the accuracy of 2D ultrasound. In 2015, Kozinszky *et al* compared the 3D VOCAL to two separate 2D

ultrasound models for calculating volume: the *shell of a spherical sector* model and the *spherical cap* model. In this group of 346 uncomplicated pregnancies, they found a correlation of 0.86 between the 2D method and VOCAL. Additionally, the two methods demonstrated good agreement with the measured placental volume *ex vivo*.[21]

Low Placental Volume Estimates Associated with Low Birthweight

Several studies explored the relationship between placental volume (estimated from imaging *in utero*) and low birthweight. Some of the earliest of these studies utilized 2D ultrasound. In 2000, Kinare *et al* examined the relationship between mid-pregnancy placental volume and birthweight in a population of rural Indian women.[22] They used 2D ultrasound to measure placental volume, and recorded data about the patients' height, weight, and biochemical parameters. Of note, most of the women were of short stature and had low BMI (average height 1.52 m; average BMI 18 kg/m^2). They found that low pre-pregnancy maternal weight was associated with low placental volume. Furthermore, small placental volume was independently associated with low birthweight.[22] In 2001, Thame *et al* reevaluated whether 2D ultrasound estimates of placental volume could predict the size of the infant at birth.[1] They assessed placental volume, biparietal diameter, femoral length, and head and abdominal circumference, at 14, 17, and 20 weeks gestation. At the time of delivery, neonate weight and length, as well as length, chest, head, and abdominal circumference were assessed. The authors also measured placental weight at birth. Compared to fetal anthropometry, placental volume was the most predictive of birthweight at the three points of gestation.[1]

Studies using 3D ultrasound also showed a positive relationship between *in utero* placental volume estimates and birthweight. In 2011, Antsaklis *et al* performed a study on 156 patients to evaluate the relationship between placental, gestational sac, and embryo volumes (measured by 3D ultrasound) and birthweight at delivery.[23] They concluded that placental, gestational sac, and embryo volumes increased as the gestation progressed. Fetal volume had the strongest association with birthweight ($r^2 = 0.24$, $p = 0.003$). Although placental volume had a weakly positive association with birthweight ($r^2 = 0.16$, $p = 0.047$), the authors concluded that placental volume was still weakly predictive of birthweight.[23]

The relationship between small placental volume and low birthweight was also observed during MRI studies of placenta. Derwig *et al* obtained placental MRI images during the second trimester of 83 singleton pregnancies.[24] Of these patients, 46 were considered small for gestational age (SGA), defined as having a birthweight below the 10^{th} percentile. Placental volumes were significantly lower in pregnancies leading to SGA infants than in pregnancies leading to average for gestational age (AGA) infants. The authors also used transvaginal ultrasound to image the uterine arteries, and assessed the pulsatility index (PI). The PI provides an estimate of placental blood flow; higher PI values represent higher impedance to blood flow, and therefore reduced flow. In addition to being associated with SGA infants, low placental volumes were associated with high PI. Therefore, the authors concluded that low uterine perfusion likely contributed small placental volumes (likely due to failure of the spiral arteries to convert to low-resistance blood vessels).[24]

Pomorski *et al* used 3D power Doppler to further explore these relationships between low placental volume, reduced placental perfusion, and birthweight.[25] In a study of 120 patients (20 IUGR, 100 normal) during the second and third trimesters, they used 3D power Doppler to measure three vascular indices: vascularization index (VI), flow index (FI), and

vascularization flow index (VFI). The VI, which represents the fraction of organ perfusion, is obtained by calculating the ratio of color voxels to total voxels on the Doppler image. The FI represents the flow velocity, and is obtained by averaging the colors of the Doppler image. The VFI represents the total perfusion and blood flow, and is calculated by averaging the voxels with and without color. The authors found that the placentas in IUGR pregnancies were, on average, 92.42 cm^3 smaller than those in normal pregnancies, and had lower values for VI, FI, and VFI.[25] Similarly, during a study of 388 women, Odibo *et al* found that low values for three vascular indices in the first trimester are associated with preeclampsia.[26]

Current Practice

Despite the association between small placental size and adverse fetal outcomes, evaluation of placental area or volume is not part of current prenatal care guidelines.[2, 27] A number of technical challenges could explain why measuring placental volume is not common practice: historically, methods for measuring placental volume relied on MRI or 3D ultrasound, and were time-consuming, expensive, and required extensive training.[4, 28-30] In an effort to promote placental evaluation during routine prenatal examinations, Abramowicz *et al* described a systematic approach for placental ultrasound.[28] They recommended measuring placental size in two dimensions, and measuring thickness if the placenta seems abnormally small. In addition to measuring placental size, they suggested imaging placental location, implantation, anatomy, and morphology.[28] However, despite these recommendations and advances in techniques to measure placental volume, it is still not commonly performed in prenatal evaluation.

Estimated Placental Volume (EPV) Technique

Given the potential importance of measuring placental volumes on gestational outcomes, a simpler approach was needed to encourage physicians to incorporate this screening tool into their practice. In 2010, Azpurua *et al* described a technique that utilizes 2-dimensional (2D) and mathematical modeling to estimate placental volume.[27] Unlike previous techniques, their estimated placental volume (EPV) technique could be performed quickly at the bedside by a healthcare provider with minimal training. This approach for estimating placental volume correlated well with actual placental weights.

In 2013, Arleo *et al* validated this method and developed normative EPV growth curves based on data from 423 patients at Weill Cornell Medical Center.[2] In a small subset of four patients, they demonstrated that EPV may be a useful tool for detecting abnormally small placentas. Aye *et al* compared EPV to a semi-automated method for measuring placental volume.[31] This technique, which was developed by Grady *et al*,[32] utilizes a random walk algorithm to rapidly assess the volume of placentas of varying shapes. They found that the semi-automated method was more accurate than EPV at estimating placental between 11 and 13 weeks gestation, when placentas are more heterogeneous in shape. Nonetheless, EPV was still an accurate measure of assessing placental volume, and is less expensive than the technique presented by Aye *et al*.[32]

Our Study: EPV in the YNHH Population

In our study, we aimed to validate EPV compared to birthweight in a large cohort at Yale, and to evaluate the odds that small EPV is associated with low BW. Our goal was to contribute to the growing EPV literature, in an effort to promote adoption of simple screening tool with potential clinical benefit.

"…neither is ther eoccasion for returning and refining this blood [of the fetus] in the lungs of the mother, because that office is sufficiently performed in the placenta until the foetus is delivered, when its own lungs are put to sufficient use"
-William Smellie, 1752

2

Statement of Purpose

Specific Aims

- To evaluate the relationship between EPV and BW in a population of patients presenting for obstetrical care at YNHH
- To examine placental pathology of select patients whose infants had low EPVs or adverse fetal outcomes

Hypothesis

- Small EPV is associated with a statistically significant increased risk of low BW. Therefore, EPV can be used as a screening tool to detect low BW.
- The placentas associated with low EPV or adverse fetal outcomes demonstrate significant pathology.

"The [umbilical] vessels join on the uterus like the roots of plants and through them the embryo receives its nourishment."

-Aristotle, *On the Generation of Animals* (340 B.C.)

3

Methods

Placental Ultrasound

This study was approved by the Yale University School of Medicine Human Investigation Committee (protocol number 0905005157). Informed written consent was obtained for each patient. Using a method for calculating EPV previously developed and validated [2, 27], 419 studies were performed on 366 patients (by trained ultrasonographers or physicians) to measure placental width, height and thickness dimensions. For each participant, an ultrasound image of the placenta with overlaid measurements was printed and saved for future quality control review. The patients' estimated gestational age (GA) at the time of the scan was recorded. For this analysis we only evaluated the first EPV collected from each patient, resulting in a set of 366 EPV data points. For participants who delivered their baby at YNHH, the infant's BW (BW) was recorded at the time of delivery. For the

remaining patients who did not deliver at YNHH, BW data was not available to the investigators.

Inclusion/Exclusion Criteria

Inclusion criteria were as follows: any pregnant woman between 8 and 42 weeks gestation; singleton gestation; and 18 years old or greater. Exclusion criteria included: rupture of membranes, intramural fibroid, placenta previa; and women in active labor.

Data Analysis

Using R version 3.3.2 (The R Foundation for Statistical Computing Platform) statistical software, an EPV vs GA best fit curve was generated. Subgroup analyses were performed to elucidate differences between the populations of participants who delivered at YNHH, and those who did not. In particular, the gestational age at which women presented for their first ultrasound scan was compared. Additionally, individual EPV vs GA curves were generated and compared for these two groups. For the 174 patients who we had BW data for, plots were generated comparing the standard residuals of EPV and BW. Small EPVs and small BWs (defined based on the plots of standard residuals of EPV and BW) were defined as a positive screening test and positive condition, respectively. R was also used to calculate the odds ratio that a small EPV is associated with a small BW. (Statistical regressions were performed by author JE; statistical analysis and interpretation performed by authors KMM, JE, HJK.)

Placental Pathology

Placental pathology samples from select cases were formalin-fixed and paraffin embedded, stained with hematoxylin and eosin, and examined with a Nikon Eclipse 80i microscope.(Placental pathology samples were collected, stained, and examined by author

HJK. Interpretation and discussion of placental pathologies were performed by KMM and HJK).

"Just as the fingers of the hand are interwoven…so the fleshy [placental] villi of these little sponges are interwoven."
- Leonardo da Vinci (15th century)

4

Results

Our first objective was to explore the relationship between EPV and GA within the Yale data, and compare this to a previously published set of EPV versus GA data from Weill Cornell Medicine.[2] Both datasets demonstrated a parabolic relationship between EPV and GA (Figure 1). The previous data were best fit with the following equation: $EPV = (0.384\ GA - 0.00366GA^2)^3$, while our data were best fit with the following equation: $EPV = (0.372\ GA - 0.00364GA^2)^3$. The virtual identity of the coefficients suggested that the intrinsic biology of the placentas were the same in both groups, that is, the placentas grew at similar rates in both populations as the gestations progressed.

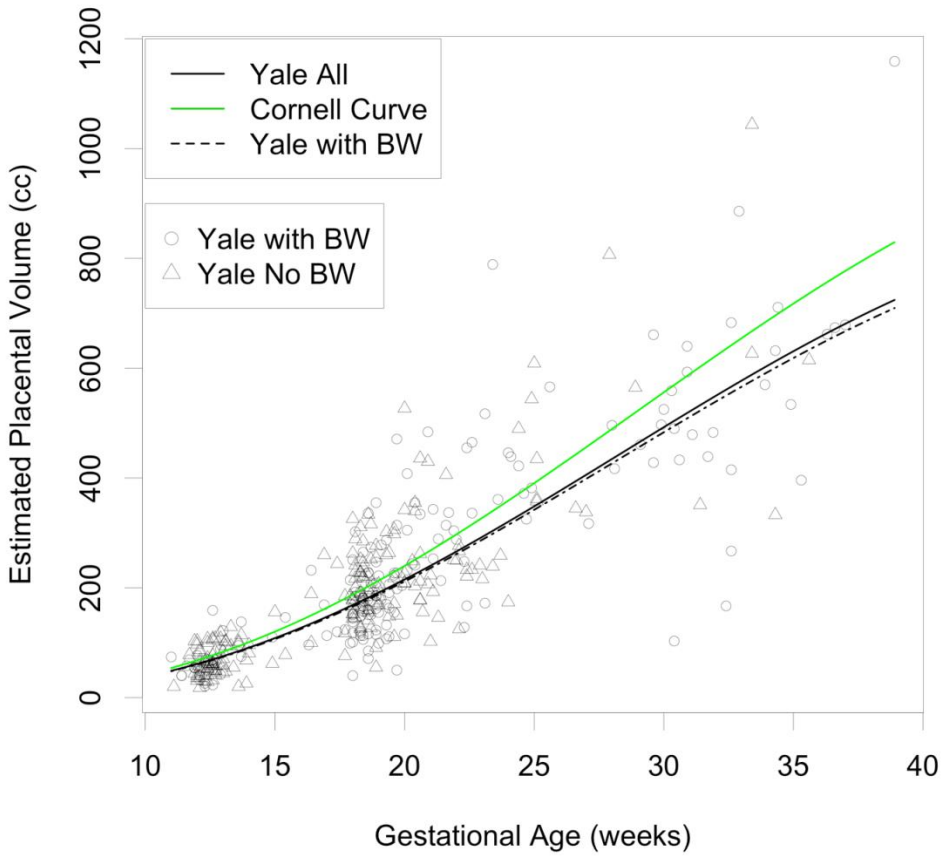

Fig. 1. Estimated Placental Volume (cc) versus Gestational Age (weeks) curves for the entire Yale data set (solid black curve), published Cornell coefficients (solid green curve), and the Yale data set with available BW (black dashed curve). The Yale raw data are plotted for the points that did have BWs recorded (circles) and did not (triangles).

We next explored whether there were systematic differences between the patients that delivered at YNHH and those that did not. If a patient delivered at YNHH, the BW was recorded. Unfortunately we did not have BW data for patients who had EPV scans performed at YNHH, but who delivered at outlying hospitals. However, the two data sets

were very similar, both in terms of GA at EPV accrual (Fig. 2A), and the best fit of EPV vs BW (Fig. 2B).

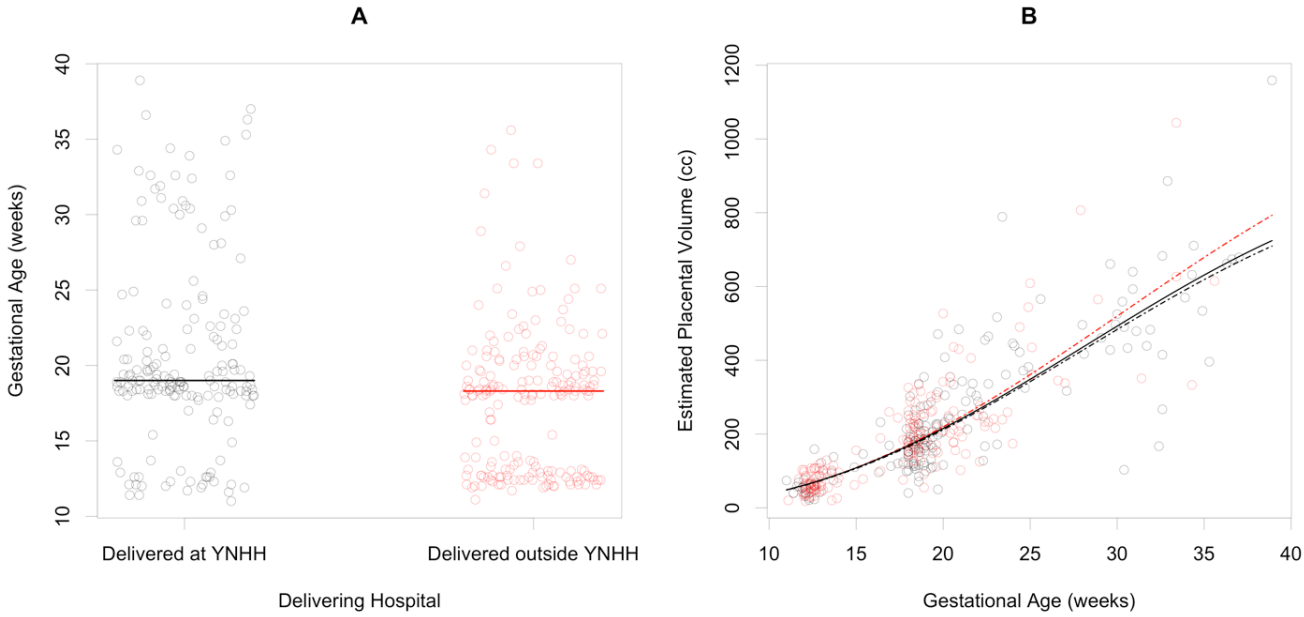

Fig. 2. Comparison of patients who delivered at YNHH and patients who delivered outside YNHH. **(A)** Gestational Age at time of EPV performance plotted for patients who delivered at YNHH (black) and outside of YNHH (red). The solid horizontal lines represent the medians for each group. **(B)** EPV versus GA for the entire Yale data set (black solid curve), patients who delivered at YNHH (black circles, fitted with black dashed curve), and outside of YNHH (red circles, fitted with red dashed curve).

We compared EPV values to BWs by plotting the standardized residuals of each parameter (Fig. 3). The majority of the data fell within ± 2.57 standard deviations for EPV and BW (red dots, corresponding to the 0.5 to 99.5th percentiles). Eight data points were well beyond 2.57 standard deviations (black dots). The r^2 value for the all the EPV vs BW data equaled 0.063 ($p<0.001$), black regression line, Fig. 3. When we eliminated the 8 outliers the r^2 equaled 0.054 ($p=0.003$), red regression line, Fig. 3. Since these regression lines

virtually overlapped, this suggested that the placentas corresponding to these extreme values of EPV and BW had the same intrinsic characteristics as the non-outlier points in the data set, and were therefore not biologically implausible.

Although dividing the data into four quadrants gives the data equal weight across the data set, one is able to perform a 2x2 analysis to evaluate the potential clinical utility of the EPV as a screening tool. The result of this analysis yielded an OR 2.42, 95% CI 1.27 – 4.68. This associates a small EPV with a small BW or conversely a large EPV with a large BW (Figure 3). Patients with an EPV in the bottom 50th percentile had 2.42 times the odds of having a newborn with a BW in the bottom 50th percentile.

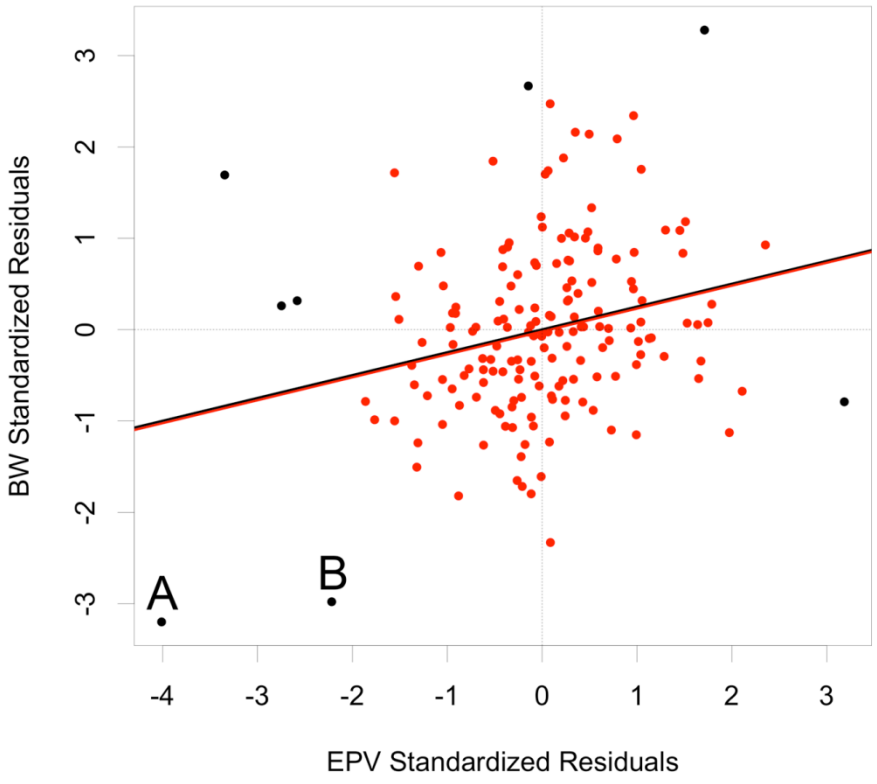

Fig. 3. Standardized residuals for EPV versus BW. The red points represent data with standardized residuals less than or equal to ±2.57, while the black points represent residuals greater than ±2.57 (less than the 0.5th percentile or greater than the 99.5th percentile). The red regression line represents the r^2 for the red-point data, while the black line represents the r^2 for all the data points. The outlier black data point (A) corresponds to Fig. 4A, while the black data point (B) corresponds to Fig. 4B.

Finally we analyzed the medical records of the 2 most extreme outliers with the lowest EPVs and BWs to determine if they could inform us about the pathogenesis of these very small placentas. For the first case (Figure 3, lower left corner black dot), the mother was a smoker who had previously delivered an infant with IUGR. In this study, her baby had an EPV of 103 cm3 at 30+3 weeks (more than 4 standard deviations below the mean). A male was delivered at 38+1 weeks with BW of 1,580 g (more than 3 standard deviations below the mean). Apgar scores at 1 and 5 minutes were 9 and 9, respectively. Microscopic analysis of the placenta revealed lymphocytic infiltrate of the chorionic villi, consistent with chorionic villitis (Fig. 4A). For the second case (Figure 3, lower left mid-quadrant black dot), the mother had a history of Crigler-Najjar syndrome (status-post liver transplant), and alcohol use disorder (in early remission during her pregnancy for this study). Her baby had an EPV of 269 cm3 at 32+4 weeks (more than 2 standard deviations below the mean). A female was born preterm at 34+1 weeks with a BW of 1,010 grams (almost 3 standard deviations below the mean). Apgar scores at 1 and 5 minutes were 4 and 6, respectively. Evaluation of the placenta revealed failure of conversion of the spiral arteries (Fig. 4B), which is associated with pre-eclampsia and IUGR. [33]

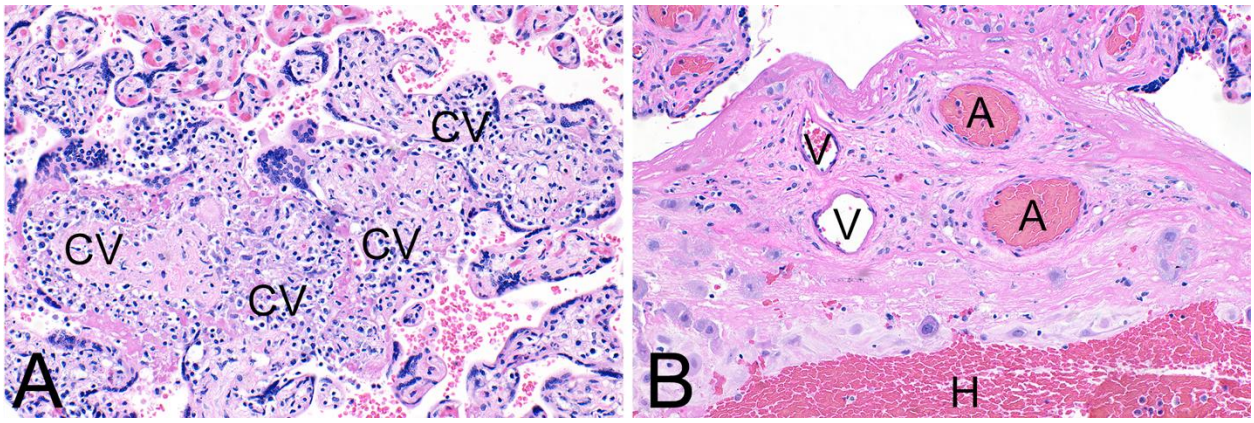

Fig. 4. Placental pathology of the two extreme EPV outlier cases highlighted in Fig. 3. **(A)** Multiple chorionic villi are agglutinated and enmeshed in fibrin and maternal T-cells, characteristic of chronic villitis (CV). **(B)** Junction of placenta and maternal decidua revealing normal maternal venules (V), but unconverted maternal spiral arterioles (A), a common finding in decreased maternal perfusion of the placenta. Hematoxylin and eosin staining.

"Surely, the placenta will deserve…increasing attention, for it is the essential structural basis…between mother and child."
-James Dixon Boyd, *Cambell oration to the Ulster Medical Society* (1959)

5

Discussion

This research study involved performing 2D ultrasound and EPV measurements on patients who presented to YNHH for prenatal care. The EPV vs GA data were plotted and fit with a validated mathematical model previously described by authors at Weill Cornell Medical.[2] Our EPV data were very similar to those collected at Cornell, suggesting that placental growth kinetics are an intrinsic characteristic of the placenta and not significantly influenced by patient population. Outliers should raise suspicion for intrinsic problems with the placenta (i.e. decreased maternal perfusion), or mismatch between the size of the placenta and the fetus.

Of the 366 patients where EPV studies were performed, only 174 patients eventually delivered at YNHH, where BW were recorded. The remaining patients delivered at an outlying hospital, where BW data was not available to the investigators. Because we were concerned that the YNHH delivered patients might represent a biased subgroup, we performed subgroup analyses on these two populations. We found that in fact they were

very similar, with almost superimposable EPV accrual GA, and EPV vs GA growth kinetics (Fig. 2).

When we compared EPV to BW we found a modest relationship between these two variables. An initial crude analysis by dividing the results into four quadrants of low and high EPV compared to low and high BW resulted in an OR 2.42, 95% CI 1.27 – 4.68 associating a low EPV with a low BW. When we used all the data in a continuous analysis, we found a weak correlation between EPV and BW ($r^2 = 0.063$). However, EPV was never intended to predict BW directly. Rather, we propose that EPV be used to identify extreme outliers of placental volume that may be associated with IUGR, IUFD, and other adverse fetal outcomes. Analysis of the standardized residuals (Fig. 3) suggested that extremely small EPVs are associated with extremely low BWs.

Examining the two extreme EPV outlier cases illustrated the importance of incorporating EPV into clinical practice. Both babies demonstrated IUGR. In both cases, there were several maternal variables that could contribute to adverse fetal outcomes (e.g. smoking, alcohol use, history of hepatobiliary pathology). Although these variables could potentially confound the relationship between placental size and fetal complications, it is noteworthy that EPV was extremely small in both cases. In practice, there may be important underlying maternal medical conditions that the mother and obstetrician might be unaware of. In such cases, EPV could serve as a red flag to follow the mother and fetus more closely, and to evaluate the placenta for underlying pathology.

Because EPV is so easy to perform, we recommend routine EPV measurements whenever the fetus is examined by ultrasound. A small EPV for gestational age could serve as an alert to the obstetrician to follow the mother and baby more closely.[2, 27] There are differences in the clinical utility between an early EPV versus a late gestational EPV

measurement. Prior to the GA of viability there is little direct action that can be proposed in the face of a very small EPV. In those cases following the patient and fetus, possibly with increased frequency, may be the only option. However, as the patient approaches GAs with increased probability of survival, the decision for more intense fetal evaluation, and possible delivery, becomes more advantageous.

This study has several limitations. First, having all of the BW data would have increased the number of patients analyzed and therefore would have increased the generalizability of the study. Second, as our patients were solicited in our routine prenatal care clinics, we had a low frequency of adverse pregnancy outcomes in the patients studied. Future studies could focus on high-risk patients where adverse outcomes are more common. Furthermore, EPV efficacy could be validated during labor and delivery triage to identify high-risk patients with very small placentas who might be inappropriately discharged due to reassuring fetal monitoring.

Unlike previous methods for determining placental volume,[27-30, 34] obtaining 2D ultrasound images of the placenta and calculating EPV is fast and requires minimal cost and training. It is a robust method with demonstrated validity across different populations. As such, it has the potential for clinical utility in a variety of settings.

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