Gestational age assessment by ultrasound cerebellar measurements in fetal and perinatal deaths

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BACKGROUND: Perinatal mortality remains high in low- and middle-income countries. Accurate assessment of fetal gestational age is crucial to distinguish between prematurity and intrauterine growth restriction, 2 conditions commonly associated with perinatal mortality that require different preventive strategies and management. Ultrasound measurements of the cerebellum have been shown to be accurate in assessing gestational age during pregnancy, but their postmortem performance has not yet been evaluated.

OBJECTIVE: We aimed to explore the feasibility and validity of gestational age estimation in fetal and perinatal deaths by ultrasound measurements of the cerebellum.

STUDY DESIGN: This is an observational cross-sectional study. Between August 2020 and November 2022 postmortem cerebellar ultrasound measurements were conducted in a tertiary referral hospital in Barcelona, Spain. Extrauterine assessment included transcerebellar diameter, cerebellar vermis height, and cerebellar vermis length. Moreover, intrauterine ultrasound and autopsy direct cerebellar assessments were undertaken in a subset of cases. A total of 137 fetal and perinatal deaths [63 (46.0%) fetal deaths, 69 (50.4%) stillbirths, and 5 (3.6%) neonates] were included. First, we correlated different types of transcerebellar diameter measurements between them (intrauterine, extrauterine, and autopsy-based). Then, we evaluated the relationship between the extrauterine cerebellar ultrasound measurements and gestational age, and their performance across trimesters of gestation and in different central nervous system abnormalities.

RESULTS: Gestational age ranged from 15.2 to 40.6 weeks. High correlation was observed between extrauterine, intrauterine, and autopsy transcerebellar measurements ($P<.001$) and between all extrauterine cerebellar measurements and gestational age ($P<.001$). Extrauterine transcerebellar diameter was identified as the strongest predictor of gestational age (coefficient of determination=0.88; $P<.001$), and its accuracy was not affected by the trimester of gestation, intrauterine growth restriction, or central nervous system alterations.

CONCLUSION: This study shows the feasibility and accuracy of postmortem gestational age evaluation by extrauterine ultrasound measurements of the cerebellum, especially of transcerebellar diameter. Implementation of this method as part of postmortem assessment could improve cause of death attribution, especially in resource-constrained settings.

Key words: fetal, gestational age, perinatal, stillbirth, transcerebellar diameter, ultrasonography

Introduction

Despite a global decrease in child mortality rates over the last 3 decades, progress in reducing perinatal mortality has been slow, particularly in low- and middle-income countries (LMICs).[1,2] Understanding the causes of perinatal death is critical to offer better-quality care, identify trends, and develop preventive practices.[3,4] Complete autopsies, the gold standard for cause of death attribution, are difficult to perform in most LMICs.[5] Minimally invasive autopsy, also known as minimally invasive tissue sampling (MITS), based on performing a series of percutaneous punctures to obtain core tissue biopsies, has emerged as a helpful tool to assess the cause of death and improve mortality surveillance in LMICs, where information on causes of death is very poor.[6,7]

Accurate data on gestational age (GA) and fetal growth are crucial when interpreting postmortem findings and assigning the cause of death in perinatal MITS.[4,8] Two prevalent conditions commonly associated with perinatal death are prematurity and intrauterine growth restriction (IUGR),[9–12] which share clinicopathological findings and low birth weight. Accurate GA estimation enables differential diagnosis between the 2 conditions, which have different underlying etiological factors and require different preventive measures and management.

The standard method to determine GA in high-income settings has classically been the clinical history (days since the beginning of the last menstrual period). Moreover, due to the frequent uncertainty of the data provided by the women and the variability of menstrual cycles, other measures that accurately correlate with GA are used, such as ultrasound measurement of crown-rump length during the first trimester of pregnancy, which is the gold standard method for assessing GA.[13] Unfortunately, in LMICs, recall of the last menstrual period is frequently imprecise[14] and first trimester ultrasound biometry is hardly ever performed due to the limited capacity of healthcare services and societal issues.[15] Thus, there is an urgent need for innovative approaches allowing accurate evaluation of GA in a cost-effective and feasible manner.

Recently, several intrauterine ultrasound measurements of the cerebellum,

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AJOG at a Glance

Why was this study conducted?

Gestational age (GA) data are crucial in perinatal deaths but are often missing in the deaths occurring in low-resource settings. The question remains: can we reliably estimate the GA in fetal and perinatal deaths using a postmortem cranial ultrasound?

Key findings

The results of this study show that this method is valid to estimate GA.
We showed a high concordance between postmortem cerebellar ultrasound measurements and GA in a series of fetal deaths and stillbirths, particularly when transcerebellar diameter (TCD) was measured.
The performance of TCD to assess GA was not affected by the trimester of gestation, intrauterine growth restriction, or central nervous system abnormalities.

What does this add to what is known?

Thereby, postmortem cerebellar ultrasound measurements can be used to estimate GA in high burden, resource-limited settings.

including transcerebellar diameter (TCD) and cerebellar vermis dimensions (height and length), have been proposed as an alternative to reliably evaluate GA.[16−22] The cerebellum can be visualized by ultrasound as early as 10 to 11 weeks and its growth is not altered by extrinsic pressures, as it is surrounded by dense bones. Increases in cerebellar size have been shown to correlate linearly with GA, even during the third trimester,[16,23] and to remain consistent in cases of IUGR.[24,25] Ultrasound cerebellar measurements have been shown to accurately predict GA when conducted in utero during pregnancy[16,26,27] and in neonates after birth.[17,21,22] Additionally, some studies have shown the usefulness of TCD in determining GA when measured postmortem on formalin-fixed brain specimens obtained in complete autopsies.[28,29] However, the value of these measurements in predicting GA when measured in fetal and perinatal deaths through postmortem extrauterine ultrasound has not yet been evaluated.

In this study, we aimed to explore the feasibility and validity of GA estimation in fetal and perinatal deaths by extrauterine percutaneous transfontanelle ultrasound measurements of the cerebellum, including TCD and the height and length of the cerebellar vermis. As part of this investigation, we first validated the accuracy of postmortem extrauterine cerebellar ultrasound by comparing its performance with well-characterized methods (ie, postmortem intrauterine ultrasound measurements and complete autopsy measurements in formalin-fixed brain specimens) in a subset of cases. Then, we determined the accuracy of these cerebellar measurements to ascertain the GA. The analysis was carried out in a series of fetal and perinatal deaths occurring at second and third trimesters of gestation and included a subset of cases with central nervous system (CNS) abnormalities and/or IUGR, which are conditions that might potentially affect the development of the cerebellum.[30]

Methods

Study setting and design

This observational cross-sectional study was conducted in the tertiary care Departments of Obstetrics and Gynaecology and of Anatomic Pathology of the Hospital Clínic de Barcelona (HCB), Spain, between August 2020 and November 2022 and included: fetal deaths, stillbirths, and neonatal deaths. Those deaths occurring between 15 and 21.9 weeks of gestation were defined as fetal deaths, while deaths at or beyond 22 weeks of gestation were classified as stillbirths, which aligns with the standard criteria used in our institution.

The following inclusion criteria were defined: 1) intrauterine fetal death due to natural causes or legal abortion, occurring later than 15 weeks of GA; 2) neonatal death occurring in the first 48 hours of life; 3) regular ultrasound scan controls of pregnancy, with determination of GA by the gold standard method (intrauterine ultrasound measurement of crown-rump length conducted during the first trimester); and 4) written informed consent provided by the mother and/or the relatives of the deceased to perform the ultrasound measurements or complete autopsy. Only 2 cases of multiple pregnancies were identified in our cohort, and given the small number, they were excluded to maintain homogeneity in the study population.

The investigation encompassed 2 substudies: a) validation of the extrauterine postmortem ultrasound cerebellar measurements (ie, TCD) by comparing its performance with the intrauterine ultrasound postmortem measurements (first set of 36 cases) and with the autopsy measurements of the cerebellum in formalin-fixed CNS specimens (second set of 35 cases, with 12 overlapping cases between these 2 sets); and b) to determine the accuracy of these cerebellar measurements to ascertain GA (entire cohort). The sample size was calculated based on the average number of perinatal deaths occurring at our institution in a period of 2 years. A sample size of 137 achieves 100% power to detect a change in slope from 0.00 under the null hypothesis to 0.88 under the alternative hypothesis when the standard deviation of the X's is 1.00, the standard deviation of Y is 1.00, and the two-sided significance level is 0.05.

The study was approved by the Clinical Research Ethics Committee of the HCB (Spain; approved, File HCB/2020/0289). Clinical data on pregnancy (including GA determined by intrauterine ultrasound measurement of crown-rump length conducted during the first trimester) were retrieved from institutional electronic records.

First trimester ultrasound: gold standard determination of GA

Intrauterine ultrasound scan was performed in all cases in the first trimester of gestation as part of the routine evaluation of the pregnancy at the outpatient obstetric clinics. In this evaluation, potential fetal abnormalities were identified, and GA was determined based on the measurement of crown-rump length.[31]

Postmortem intrauterine ultrasound cerebellar measurements

In case of suspected intrauterine fetal death, abdominal ultrasound scan was performed to confirm fetal death and, upon confirmation of fetal decease, measurement of fetal TCD was performed in a subset of cases. This procedure was performed at the maternity ward by an obstetrician expert in the fetal ultrasound. Ultrasound examination was carried out using Voluson E10 Expert and Voluson S8 US devices (GE Medical Systems, Australia). The TCD was measured as previously described.[23,32,33]

Complete autopsy and conventional TCD measurement on brain specimens

From the time of delivery until autopsy, deceased bodies were kept at 4°C. The date and the time of the death in all feticides, the date of delivery, and the date and time of autopsy were recorded for each case. The autopsy was performed following a standard protocol. Histological samples from the main organs including the liver, brain, and lungs were analyzed at the Pathology Laboratory of the HCB. The presence of IUGR and/or CNS abnormalities possibly affecting cerebellar growth was carefully evaluated at autopsy. IUGR cases were defined as cases below the 10th percentile of weight expected for the GA.[34] The causes of death were assigned and coded following the International Classification of Diseases, 10th revision for deaths occurring in the fetal and perinatal period. The brain specimen was fixed in formalin for a minimum of 3 weeks, and the TCD was measured as the widest transverse diameter of the cerebellum in a subset of cases.[28]

Postmortem extrauterine cerebellar ultrasound measurements

Extrauterine postmortem cerebellar ultrasound measurements were performed in the autopsy room of the HCB. In the cases in which a complete autopsy was requested and authorized, the measurements were obtained immediately before autopsy. Three cerebellar measurements were obtained: 1) the TCD, 2) the cranio-caudal length (height) of the cerebellar vermis, and 3) the anteroposterior length (length) of the cerebellar vermis. All the measurements were performed by a pathologist or a pathology technician who had received previous training in the procedure using a Siemens Acuson S2000 pediatric ultrasound scanner (Siemens Medical Solutions, Mountain View, CA) with a 4 to 9 MHz transducer. First, the ultrasound transducer was placed on the anterior fontanelle with ultrasound transmission gel. Then, the transcerebellar plane was identified and the TCD was measured in an outer-to-outer method following the same method used for intrauterine ultrasound evaluation. Then, the direction of the transducer was changed until obtaining a midsagittal plane, which allowed visualization of the cerebellar vermis as a hyperechoic structure delimited anteriorly by the fourth ventricle and posteriorly by the cisterna magna. To obtain a precise midsagittal plane, we ensured that the corpus callosum was clearly visualized anteriorly.

All postmortem measurements and estimations were blinded to the estimation of GA performed in the first trimester of pregnancy.

Figure 1 shows the representative images of the postmortem extrauterine ultrasound measurements conducted in the autopsy room.

Statistical methods

All the statistics were performed using R software (version 4.0). Postmortem interval (PMI) time was calculated for the legal abortion cases as time between administration of feticide and autopsy. In addition, interval between diagnosis of fetal death (feticide) and fetal delivery was calculated in the same subset of cases.

Normal distribution of the data was assessed using the Shapiro-Wilk normality test. Mann-Whitney *U* tests were used to assess differences in cerebellar measurements between sexes.

Scatter plots were used to illustrate the relationship between pairs of cerebellar measurements and between them and GA. Spearman's rank correlation and Bland-Altman plots were used to assess the correlation and agreement between the different methods of TCD measurement (intrauterine ultrasound, extrauterine ultrasound, complete autopsy), respectively. Univariate linear regression analysis was used to assess the relationship between the postmortem extrauterine cerebellar ultrasound measurements and GA, and multivariate analysis to evaluate interactions between cerebellar measurements and various conditions (trimester of gestation, IUGR, CNS abnormalities, PMI, or interval between diagnosis of fetal death and delivery). For the univariate and multivariate models, the equation of the model, the 95% confidence interval of the intercept and slope, the coefficient of determination (R^2) and the *P* value were reported.

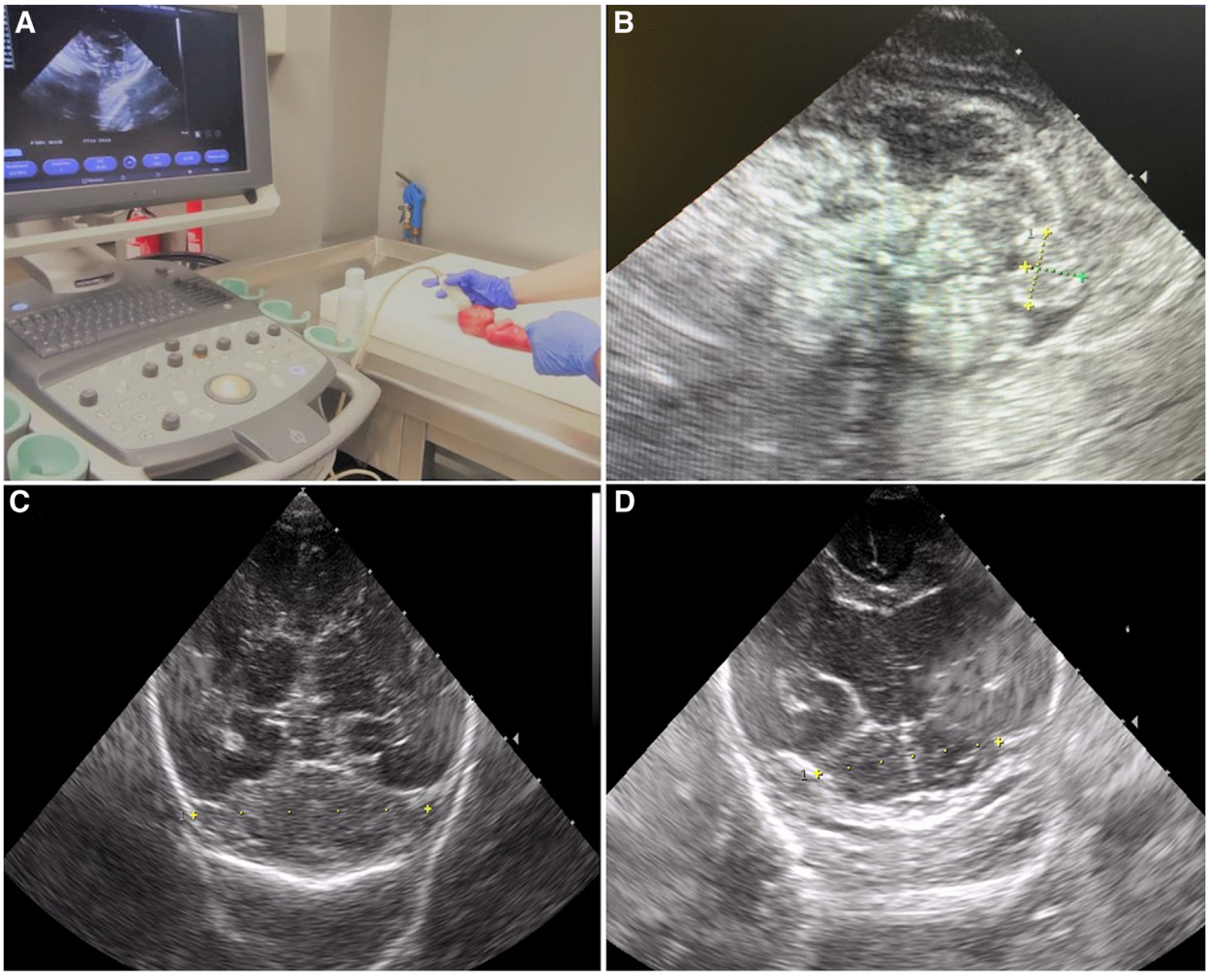
Results

General characteristics of the enrolled cases

A total of 137 cases (59.9% males and 40.1% females) were enrolled: 63 (46.0%) fetal deaths, 69 (50.4%) stillbirths, and 5 (3.6%) neonates. The median GA determined by the gold standard (intrauterine ultrasound measurement of crown-rump length during the first trimester) was 20.5 weeks (interquartile range [IQR]=19.5−21.3) for fetal deaths, 23.0 weeks (IQR=22.5−30.6) for stillbirths, and 28.5 weeks (IQR=23.2−35.3) for neonates. The median life of the neonates was 12 hours (IQR=12.0−28.0). A complete autopsy was performed in 116 out of 137 cases (84.7%). Mean PMI was 94 hours (range 23−293 hours). Mean time from feticide to delivery was 35 hours.

Table 1 shows the basic demographic and clinical features of the pregnant

FIGURE 1
Images of the extrauterine postmortem ultrasound measurements



Representative images of the extrauterine postmortem ultrasound measurements of the cerebellum conducted on a stillbirth in the autopsy room. **A,** percutaneous transfontanelle visualization and measurement of the cerebellum using the pediatric ultrasound equipment; **B,** cerebellar vermis height and length measured midsagitally from the anterior fontanelle (*yellow and green line*, respectively; a characteristic triangular shape of the cerebellar vermis, as well as the indentation on its anterior border are clearly recognized; **C** and **D,** transverse cerebellar diameter postmortem measurements in the premature neonate of 35 weeks of gestation and a few hours of life and in the fetus of 20 weeks of gestational age.

women and the fetuses, stillbirths, and neonates enrolled in the study. Of all 137 decedents, 104 (75.9%) were legal abortions, whereas 33 deaths (24.1%) were due to natural causes. The causes of deaths (clinical or autopsy-based diagnosis) are listed in Supplemental Table 1.

Validation of the postmortem extrauterine measurement of the TCD

TCD was measured by intrauterine ultrasound and, subsequently, by extrauterine ultrasound in 36 cases (26.3%), obtaining median values of 20.8 mm (IQR=19.0−24.4) and 23.7 mm (IQR=21.5−29.1), respectively. Additionally, TCD was measured by extrauterine ultrasound and, subsequently, directly on formalin-fixed cerebellums in 35 out of 137 cases (25.5%), obtaining median of 25.1 mm (IQR=22.8−36.6) and 25.0 mm (IQR=23.0−33.5), respectively. No differences in these TCD measurements were noted between males and females (Supplemental Table 2).

Strong correlation was noted between extrauterine ultrasound TCD, intrauterine ultrasound TCD, and autopsy TCD ($P<.001$ for all the correlations) (Figure 2). The correlation coefficient ranged from 0.81 (extrauterine ultrasound vs autopsy) to 0.87 (extrauterine vs intrauterine ultrasound). Intrauterine measurements generally yielded lower TCD values compared to those obtained by extrauterine ultrasound (mean bias=2.67 mm) and autopsy (mean bias=3.61 mm), while the latter 2 measurements were more similar (mean bias=0.55 mm) (Supplemental Figure 1).

TABLE 1
Demographic and clinical characteristics of 137 study participants (pregnant women and stillborn babies and neonates)

Characteristics	n (%)
Maternal age	35.0 (31.0–38.0)[a]
Maternal age, groups	
<20 y	3 (2)
20–30 y	30 (22)
31–40 y	90 (66)
41–50 y	14 (10)
Type of perinatal death	
Fetal	63 (46)
Stillbirth	69 (50)
Neonatal	5 (4)
Sex	
Male	82 (60)
Female	55 (40)
Gestational age (GA)	
Second trimester	104 (76)
Third trimester	33 (24)
Birth weight	
<500g	85 (62)
500–1000g	28 (20)
1000–2000g	12 (9)
2000–3000g	8 (6)
>3000g	4 (3)
Intrauterine growth restriction (IUGR)	
Yes	13 (9)
No	124 (91)
Central nervous system (CNS) abnormalities	
Yes	22 (16)
No	115 (84)
IUGR and/or CNS abnormalities	
Yes	36 (26)[b]
No	101 (74)

%, proportion of total number of cases; *n*, number of cases.

[a] Values shown are median (interquartile range); [b] Three cases had both IUGR and CNS abnormalities.

Prediction of GA by the different cerebellar measurements

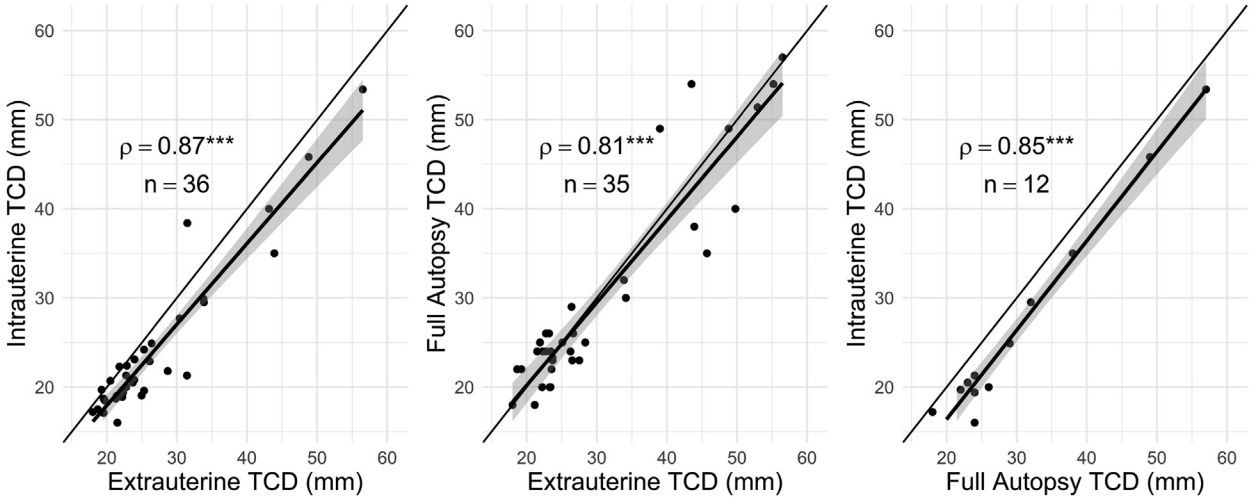
Of 137 cases, extrauterine postmortem TCD was successfully measured in all (100%) and the cerebellar vermis height and length in 135 cases (98.5%). Measurement of the vermis was not feasible in 2 cases; one with rhomboencephalosynapsis, a condition characterized by the absence of the vermis, and one with extreme maceration. Median values of cerebellar measurements were: TCD 25.9 mm (IQR=22.0–30.4), vermis height 12.5 mm (IQR=10.1–14.2), and vermis length 5.8 mm (IQR=4.7–7.6). No differences between sex assigned at birth and cerebellar measurements were identified (Supplemental Table 2).

Univariate linear regression confirmed an excellent prediction of GA via intrauterine ultrasound TCD (R^2=0.918, P<.001) and autopsy TCD (R^2=0.934, P<.001). Additionally, it revealed a significant (P<.001) positive linear relationship among the 3 extrauterine postmortem cerebellar ultrasound measurements and GA (TCD R^2=0.885; vermis height R^2=0.875; and vermis length R^2=0.633). The 3 linear models for the estimation of GA via postmortem cerebellar measurements were as follows: GA=9.75+0.49 (mm of TCD), GA=10.52+0.97 (mm of vermis height), GA=13.12+1.62 (mm of vermis length). Scatter plots and linear regression equations are shown in Figure 3 and linear regression parameters are outlined in Table 2. The complete list of parameters calculated for the univariate linear regression analysis is shown in Supplemental Table 3. A calibration graph showing the GA predicted by the goldstandard crown-rump length and the GA predicted by extrauterine cerebellar measurements is shown in Supplemental Figure 2.

Accuracy of extrauterine cerebellar ultrasound measurements to predict GA across trimesters of gestation and in the presence of IUGR or CNS abnormalities

IUGR, potentially affecting cerebellar growth, was detected and confirmed during autopsy in 13 out of 137 cases (9.5%). Twelve cases were classified as moderate IUGR (birth weight between 3rd–10th percentile) and one case as severe IUGR (birth weight <3rd percentile).[34] CNS abnormalities, potentially affecting cerebellar growth, were identified and confirmed at autopsy in 22 cases: 3 (13.6%) with neural tube defects (including Arnold-Chiari malformation); 6 (27.3%) with ventriculomegaly; 4 (18.2%) with midline abnormalities (including holoprosencephaly and agenesis of corpus callosum); 3 (13.6%) with disorders of neuronal proliferation (including

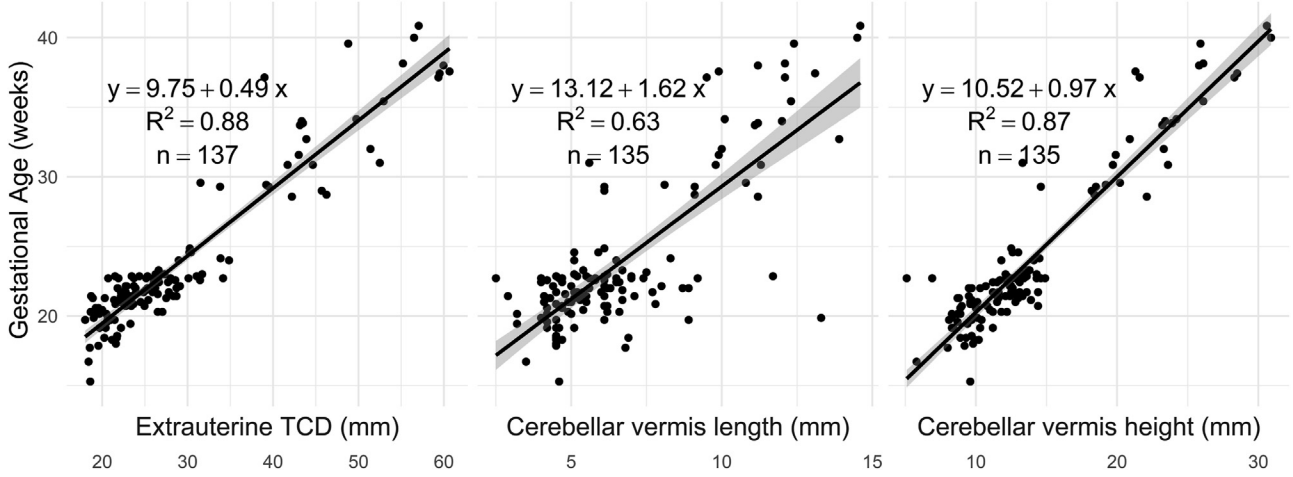
FIGURE 2
Correlation among different methods for measuring the transcerebellar diameter



Scatter plots showing the relationship between extrauterine ultrasound measurements of transcerebellar diameter (TCD), intrauterine ultrasound TCD, and measurement of the TCD at complete autopsy.
Spearman correlations (ρ) indicate the strength of the relationship between the 2 variables. The identity line (*thin line*), which indicates perfect correlation, is also shown for comparison. Each dot in the scatter plot represents an individual case, with one cerebellar measurement plotted on the x-axis and the other one on the y-axis. The regression line (thick line) illustrates the modeled relationship between the variables. The shaded area around this regression line represents the 95% confidence interval. ***P value $<.001$.

microcephaly); 1 (4.5%) with large cystic hygroma; 1 (4.5%) with enlarged cisterna magna; 1 (4.5%) with intracranial hemorrhage, 1 (4.5%) with hypoxic-ischemic encephalopathy; 1 (4.5%) with rhomboencephalosynapsis, and 1 (4.5%) case with a periventricular halo. Four out of 22 cases (18.2%) presented more than one abnormality.

FIGURE 3
Linear regression analysis of extrauterine ultrasound cerebellar measurements in relation to gestational age



Scatter plots showing the distribution of cerebellar measurements against gestational age (GA) for all cases together with the equations derived from univariate linear regression analysis, including the intercept of the regression line and the slope, which represents the change in y (weeks of GA) for a one-unit change in x (mm of the cerebellar measurement). R^2 indicates the goodness of fit of the model. All regression coefficients are significant (P value $<.001$). Each dot in the scatter plot represents an individual case, with the cerebellar measurement (mm) plotted on the x-axis and the GA (weeks) on the y-axis. The regression line (thick line) derived from univariate linear regression analysis models the relationship between these variables.The shaded area around this regression line represents the 95% confidence interval. R^2, coefficient of determination; *TCD*, transcerebellar diameter.

TABLE 2
Univariate linear regression analysis assessing the relationship between gestational age and cerebellar measurements

Cerebellar measurements	n	R^2	Regression model equation
Autopsy (conventional) TCD	35	0.934	8.891+0.546 (TCD)[a]
Intrauterine TCD	36	0.918	9.064+0.601 (TCD)[a]
Extrauterine TCD	137	0.885	9.753+0.486 (TCD)[a]
Cerebellar vermis height	135	0.875	10.524+0.974 (height)[a]
Cerebellar vermis length	135	0.633	13.117+1.619 (length)[a]

Each row corresponds to one model including an intercept and the explanatory variable indicated in the first column. Columns show the total sample (n), the coefficient of determination (R^2), and the model equation.

n, total number of cases per group analyzed; R^2, coefficient of determination; *TCD*, transcerebellar diameter.

[a] *P* value <.001.

Three cases out of 22 (13.6%) had both IUGR and CNS abnormalities. Supplemental Table 4 provides a detailed description of the CNS abnormalities for each case.

Multivariate models showed a robust performance of the TCD in predicting GA, with no interactions with trimester of gestation, IUGR or CNS abnormalities. In contrast, significant interactions were detected for cerebellar vermis length (with gestational trimester) and for cerebellar vermis height (with IUGR). Table 3 shows the multivariate analysis for the different postmortem cerebellar measurements considering trimester of gestation, IUGR, or CNS abnormalities. Additionally, no significant interactions were observed between most cerebellar measurements and PMI or the interval between the diagnosis of fetal death and delivery, with the exception of a mild interaction between vermis height and both intervals. Supplemental Tables 5 and 6 show the complete list of parameters calculated for the multivariate linear regression analysis for main covariates and PMIs, respectively.

Comment

Principal findings

Our study shows, for the first time, that GA can be feasibly and accurately estimated in fetal and perinatal deaths using post-mortem extrauterine ultrasound measurements of the cerebellum, particularly TCD. We have shown that extrauterine ultrasound measurement of TCD closely correlates with well-established methods of TCD assessment, including intrauterine ultrasound measurement.

Results in the context of what is known

The relationship between extrauterine ultrasound TCD and GA (R^2=0.885, P<.001) was slightly weaker than that between the well-established intrauterine ultrasound TCD and GA (R^2=0.918) and those found in other studies including only live fetuses (R^2=0.897, P<.001;[21] R^2=0.92, P=.0006;[26] R^2=0.997−0.982, P=.0002;[16] and R^2=0.96, P<.0001[27]). This slightly lower correlation could be attributed to postmortem changes in the cerebellum, such as cellular swelling.[35,36] The relationship between extrauterine TCD and GA was also weaker than that between the TCD measurement performed on formalin-fixed cerebellums from complete autopsies and GA shown in this study (R^2=0.934, P<.001) and in another previous report (R^2=0.96, P<.001[29]). These differences may be attributed to the higher accuracy of direct measurement compared with ultrasound-based. In spite of this slightly lower correlation, it is remarkable that, in the multivariate analysis, extrauterine TCD showed no interactions with trimester of gestation, IUGR or CNS abnormalities. These results highlight the robustness of extrauterine postmortem ultrasound TCD in predicting GA across different conditions.

Cerebellar vermis height and length measurements were slightly more difficult to obtain and were also less accurate in predicting GA compared to TCD, especially the measurement of vermis length. The association of vermis measurements with GA (vermis height: R^2=0.875; vermis length: R^2=0.633, P<.001) is similar to that reported in other studies focusing on live fetuses (vermis height: R^2=0.839, P<.001[18]; R^2=0.59, P<.001[21]; vermis length: R^2=0.757, P<.001[18]). In the multivariate analysis, the gestational trimester and IUGR showed a significant interaction with the dimensions of the cerebellar vermis when predicting GA. This interaction had not been reported in other studies analyzing vermis growth during cerebellar development in live fetuses. However, those studies focused solely on cases with adequate fetal growth for GA.[18,21,22] Considering the effect of gestational trimester and IUGR on the prediction of GA by vermis dimensions, these measurements, may not be as robust as TCD.

Clinical implications

Despite the slightly lower accuracy in predicting GA compared with well-known cerebellar measurements (intrauterine ultrasound or direct autopsy evaluation of cerebellum), our findings indicate that extrauterine postmortem ultrasound, especially TCD measurement, can be considered a useful tool for accurately estimating GA in fetal and perinatal deaths. This is particularly relevant in settings where conducting complete autopsies is challenging or not feasible.

Research implications

This relatively simple and noninvasive method might provide a more accurate diagnosis of the cause of death in MITS, and consequently, enhance the epidemiological surveillance of perinatal deaths in LMICs.

Strengths and limitations

Several study limitations should be acknowledged. First, the sample size in this study was determined based on the availability of relevant cases within the selected time period at our institution. Thus, the small sample size and the fact

TABLE 3
Multivariate linear regression analysis assessing the interaction between gestational age determination using cerebellar measurements and the covariates trimester of gestation, intrauterine growth restriction, and central nervous system abnormalities

Covariates and measurement method	n	R^2	Regression model equation[a]
By trimester			
Complete Autopsy TCD	35	0.968	**14.287**+**0.308** (TCD)+4.512 (3rd trimester)+0.045 (TCD×3rd trimester)
Intrauterine TCD	36	0.941	**13.974**+**0.356** (TCD)−0.350 (3rd trimester)+0.151 (TCD×3rd trimester)
Extrauterine TCD	137	0.908	**13.779**+**0.312** (TCD)+3.615 (3rd trimester)+0.031 (TCD×3rd trimester)
Cerebellar vermis height	135	0.921	**15.466**+**0.517** (height)+1.897 (3rd trimester)+0.204 (height×3rd trimester)
Cerebellar vermis length	135	0.877	**20.004**+**0.243** (length)+1.814 (3rd trimester)+**0.875** (length×3rd trimester)
By IUGR			
Complete Autopsy TCD	35	0.941	**8.945**+**0.539** (TCD)−2.563 (IUGR)+0.133 (TCD×IUGR)
Intrauterine TCD	36	0.918	**9.001**+**0.604** (TCD)+0.718 (IUGR)−0.027 (TCD×IUGR)
Extrauterine TCD	137	0.887	**9.562**+**0.490** (TCD)+1.622 (IUGR)−0.035 (TCD×IUGR)
Cerebellar vermis height	135	0.881	**10.067**+**1.000** (height)+**3.633** (IUGR)−0.219 (TCD×IUGR)
Cerebellar vermis length	135	0.639	**12.684**+**1.679** (length)+3.024 (IUGR)−0.448 (TCD×IUGR)
By CNS abnormalities			
Complete Autopsy TCD	35	0.935	**8.704**+**0.553** (TCD)+1.034 (CNS abn.)−0.041 (TCD×CNS abn.)
Intrauterine TCD	36	0.925	**8.396**+**0.624** (TCD)+3.346 (CNS abn.)−0.121 (TCD×CNS abn.)
Extrauterine TCD	137	0.886	**9.582**+**0.489** (TCD)+1.038 (CNS abn.)−0.019 (TCD×CNS abn.)
Cerebellar vermis height	135	0.875	**10.424**+**0.983** (height)+0.840 (CNS abn.)−0.073 (height×CNS abn.)
Cerebellar vermis length	135	0.658	**12.588**+**1.747** (length)+1.703 (CNS abn.)−0.494 (length×CNS abn.)

Each row corresponds to one model including an intercept and the explanatory variable indicated in the first column. Columns report the total sample (n), coefficient of determination (R^2), and the regression model equation. Statistically significant coefficients (*P* value <.05) are shown in bold. Significant interactions between covariates and the explanatory variables indicate that these variables differentially explain the response.

CNS abn, central nervous system abnormalities; *IUGR*, intrauterine growth restriction; R^2, coefficient of determination; *TCD*, transcerebellar diameter.

[a] Significant (*P* value <.05) regression coefficients shown in bold.

that the study was conducted in a single tertiary hospital may limit the validity and generalizability of the findings, particularly given the variation in demographics and causes of fetal and perinatal death. Indeed, the study population comprised a high proportion of legal abortions attributed to various types of fetal abnormalities (76%) and a low proportion of intrauterine natural fetal deaths (24%). In contrast, in LMICs, intrauterine hypoxia due to hypertensive maternal disorders is the primary cause of death (6% to 24%).[37,38] Also, there was a relatively low number of late stillbirths (>28 weeks of gestation [27/137 cases, 19.7%]), whereas these cases are predominant in LMICs.[2] Future studies should address this limitation by incorporating data from multiple institutions, particularly from LMICs. Additionally, although no interaction between PMI and time between fetal death and delivery and TCD in predicting GA was observed in this study, PMI and preservation of the bodies after delivery should be strictly controlled, as suboptimal conditions could compromise the accuracy of these measurements. Finally, the design of the study, with only a set of cerebellar measurements per case performed by a single evaluator did not allow to assess interobserver agreement.

Furthermore, the use of ultrasound in LMICs might face important challenges, including limited access to the equipment, high costs, and need for training.[39] Although ultrasound is a promising tool, addressing these barriers is essential for maximizing its potential impact. In this regard, recent studies have shown that low-cost, portable, and easy-to-use ultrasound devices have been easily and successfully implemented in LMICs.[40–42] Training programs should be developed to enable healthcare personnel, who may not be experts in ultrasonography, to effectively perform and interpret postmortem ultrasound. Remarkably, the personnel performing the measurements were not experts in ultrasonography, highlighting the usefulness of the technique when performed by nonexpert personnel.

Conclusions

This study highlights the potential of postmortem extrauterine cerebellar ultrasound measurements, particularly of

TCD, for estimating GA. Future studies should validate our findings and feasibility in larger and more diverse cohorts, particularly those in LMICs.

Data availability

Data collected for the study including the data collection tool, the study protocol, search terms, and any code used for the analysis will be shared upon reasonable request to the corresponding author from the time of publication of the article. ■

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