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Original Article

Fetal Cerebellar Growth Curves Based on Biomathematics in Normally Developing Japanese Fetuses and Fetuses with Trisomy 18

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We used biomathematics to describe and compare cerebellar growth in normally developing and trisomy 18 Japanese fetuses. This retrospective study included 407 singleton pregnancies with fetuses at 14-39 weeks of gestation and 33 fetuses with trisomy 18 at 17-35 weeks. We used ultrasonography to measure fetal transverse cerebellar diameter (TCD) and anteroposterior cerebellar diameter (APCD). We hypothesized that cerebellar growth is proportional to cerebellar length at any given time point. We determined the formula $L(t) \fallingdotseq Ke^{at} + r$, where e is Napier's number, t is time, L is cerebellar length, and a, K, and r are constants. We then obtained regression functions for each TCD and APCD in all fetuses. The regression equations for TCD and APCD values in normal fetuses, expressed as exponential functions, were $TCD(t) = 27.85e^{0.02788t} - 28.62$ (mm) (adjusted $R^2 = 0.997$), and $APCD(t) = 324.29e^{0.00286t} - 322.62$ (mm) (adjusted $R^2 = 0.995$). These functions indicated that TCD and APCD grew at constant rates of 2.788%/week and 0.286%/week, respectively, throughout gestation. TCD (0.0153%/week) and APCD (0.000430%/week) grew more slowly in trisomy 18 fetuses. This study demonstrates the potential of biomathematics in clinical research and may aid in biological understanding of fetal cerebellar growth.

Key words: biomathematics, cerebellum, fetus, trisomy 18 syndrome, ultrasonography

Ultrasound assessment of fetal cerebellar growth has been conducted to estimate gestational age [1,2] and to assess central nervous system abnormalities, especially trisomy 18 [3-5]. Many reference ranges for fetal transverse cerebellar diameter (TCD) acquired by ultrasonography throughout gestation have been reported [1-5], and published TCD nomograms do not substantially differ in recent reports using large sample sizes [1,2]. In terms of interpreting the growth curve, however, to the best of our knowledge, no studies have biologically evaluated growth throughout gestation, though linear and polynomial functions have been used for growth curve regression [1-5].

Two approaches have been used to describe the growth of organisms: empirical models and mechanistic models based on growth theories [6]. The growth curves obtained using empirical models including polynomial regression have no biological meaning; they are merely fitted to the data. Mechanistic models, however, describe growth processes, and these models are usually derived from a differential equation explaining

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natural phenomena [6].

Biomathematics is a branch of biology concerned with the construction of mathematical models to describe and solve key problems in biology, including growth [7]. However, it has not yet been adequately applied in medical research [8]. We consider that the synergetic interplay between biomathematics and medical research could yield tremendous results. Accordingly, in previous studies we have developed differential equations of fetal body weight change during pregnancy [9] and of the relation between maternal body mass index and breastmilk volume [10].

The present study aimed primarily to analyze TCD and anteroposterior cerebellar diameter (APCD) growth throughout gestation in normally developing Japanese fetuses, based on biomathematics. From the standpoint of a prenatal diagnosis of trisomy 18, the usefulness of APCD measurement [4,5] and the APCD/TCD ratio [5] were recently reported, in addition to that of TCD measurement [3]. Therefore, our secondary aim was to examine which among several parameters based on TCD and/or APCD most efficiently distinguishes trisomy 18, making use of the potential of biomathematics in clinical medicine.

Materials and Methods

Data for participants who met all of the following inclusion criteria were used for the creation of TCD and APCD nomograms in this retrospective study: (1) outpatient pregnant women visiting Okayama Medical Center (Okayama, Japan), with a gestational age ranging from 14 to 39 weeks, and for whom gestational age was confirmed by crown-rump length in the first trimester; (2) pregnancies with singleton fetuses in which the cerebellum was clearly visualized using the method described below; and (3) an ultrasound examination of each fetus for the study done only once in this series. The exclusion criteria were multiple pregnancies and pregnancies with maternal or fetal complications. To compare the data of trisomy 18 fetuses and normal fetuses, we also measured TCD and APCD in trisomy 18 fetuses born at the Center between January 2005 and December 2020. In all cases, trisomy 18 was diagnosed by karyotyping amniotic fluid. An ARIETTA 60 or Prosound α6 was used in normal cases, and a Prosound SSD-5000, Prosound SSD-α10 or Prosound F75 (all: Hitachi, Tokyo) with a convex transducer with a frequency range of 1-6 MHz for all machines was used in trisomy 18 cases.

We used standard procedures to measure TCD and APCD, as follows. The transverse cerebellar plane was obtained on a biparietal diameter plane with slight posterior tilting [11]. On this plane, the fetal cerebellum appears as a butterfly-shaped structure formed by the round cerebellar hemispheres [2,11]. Following identification of the cerebellar configuration, TCD and APCD were measured at the outer margins (Fig. 1). APCD was defined as the hemisphere's anteroposterior diameter, and the APCD value was expressed as the mean value of the APCD diameters of both hemispheres. One author (K.T.) performed all measurements in normal fetuses. Several different examiners measured TCD and APCD in trisomy 18 cases using the same procedure.

All data were anonymized at the Center and then transferred to Medical Data Labo (Okayama, Japan), where data analysis was conducted as follows. To analyze the relationship between cerebellar growth and time, we assumed that the increase rate of the number of cerebellar cells was proportional to the number of cerebellar cells at any given time point in accordance with the concept of organism growth [6,9]. If we accept this hypothesis, the differential equation can be written as:

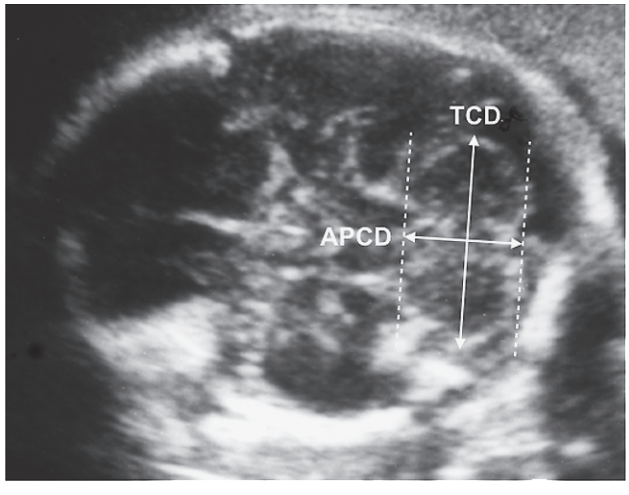

Fig. 1 Axial ultrasound image at 30 weeks of gestation during measurement of the transverse cerebellar diameter (TCD) and anteroposterior cerebellar diameter (APCD). The APCD value was expressed as the mean value of the APCD diameters of the two hemispheres.

$$\frac{dx}{dt} = kx(t), \quad (1)$$

where k is a constant, t is time (gestational days), and x is the number of cerebellar cells. The solution of Equation (1) is as follows:

$$x(t) = e^{kt} + q, \quad (2)$$

where e is Napier's number and q is the constant. The number of cerebellar cells (*i.e.*, cerebellar volume) is expressed as an exponential function. This number is considered to be proportional to the cubed length of an arbitrary site of the cerebellum because it is proportional to the cerebellar volume. The following formula is then obtained:

$$e^{kt} + q = jL(t)^3 \quad (3)$$

where j is a constant and L is the length of the cerebellum site. Thus, cerebellar length can be expressed as a function of time, as in Equation (4) below; that is, cerebellar length is also an exponential function of time:

$$L(t) \fallingdotseq Ke^{at} + r. \quad (4)$$

Here, a, K, and r are the constants. We then assumed that TCD and APCD grow independently, and obtained the regression functions for each TCD and APCD value in normal and trisomy 18 fetuses using Equation (4).

We also sought to validate the clinical significance of the regression formula (4). To do so, we obtained the intersection of the mean regression curve of the trisomy 18 fetuses and the lower limit of the 95% confidence interval (CI) curve of the nomogram for the following seven functions: fTCD, fAPCD, fAPCD/fTCD ratio, fTCD × fAPCD, ln (fTCD × fAPCD), $\sqrt{\text{fTCD} \times \text{fAPCD}}$, and ln ($\sqrt{\text{fTCD} \times \text{fAPCD}}$), where fTCD and fAPCD are the regression functions of TCD and APCD, respectively. We assumed that the parameter with the lowest x-coordinate (*i.e.*, gestational age) value of the intersection estimates trisomy 18 most efficiently.

An original program created at Medical Data Labo using the Wolfram Language (Wolfram Research, Champaign, IL, USA) was used for all statistical analyses and regression analyses, including those with 95% CIs. This study was approved by the Institutional Review Board of Okayama Medical Center (IRB No. 2020-264) and was conducted in accordance with the Declaration of Helsinki. This study was carried out with explanations provided to the patients and a website with additional information, including an opt-out option.

Results

We included the retrospective data from the cases of 407 pregnant women in this study and created TCD and APCD nomograms. The regression formulae for the TCD and APCD values were TCD $(t) = (27.85 \pm 3.74)$ $e^{(0.02788 \pm 0.0021)t} - (28.62 \pm 4.52)$ (mm) (adjusted $R^2 = 0.997$), and APCD $(t) = (324.29 \pm 307.66)\, e^{(0.00286 \pm 0.0026)t} - (322.62$ $\pm 308.47)$ (mm) (adjusted $R^2 = 0.995$); (estimated value $\pm$ standard error), respectively. The obtained regression curves fit well with the data obtained in this study (Figs. 2, 3). The regression formulae indicated a 2.788% and 0.286% weekly rate of increase in TCD and APCD, respectively, and the predicted TCD and APCD values with 95% CIs for each gestational age did not differ substantially from previously reported data [1, 2] (not shown). When the growth curve formula of the APCD/TCD ratio was represented as the ratio of each regression formula — that is, APCD/TCD $(t) = (324.29e^{0.00286t}$ $- 322.62)/(27.85e^{0.02788t} - 28.62)$ — the regression curve fit well with the APCD/TCD data obtained in this study (adjusted $R^2 = 0.529$) (Fig. 4). The APCD/TCD ratio increased rapidly between 14 and 22 weeks of gestation

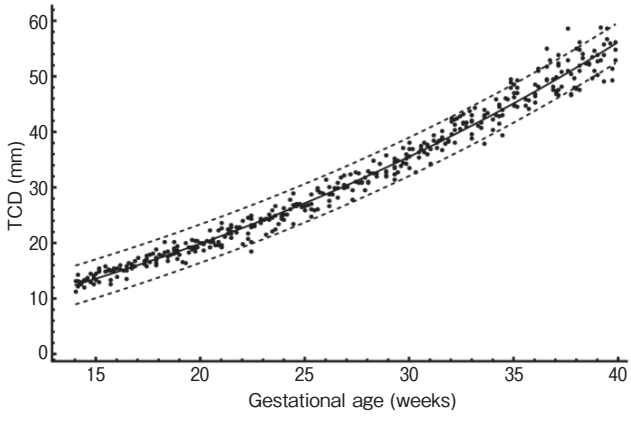

Fig. 2 Scatterplots and regression curve with 95% confidence interval for the transverse cerebellar diameter (TCD) in normal fetuses against gestational age. TCD $(t) = (27.85 \pm 3.74)\, e^{(0.02788 \pm 0.0021)t} - (28.62 \pm 4.52)$ (estimated value $\pm$ standard error), (adjusted $R^2 = 0.997$); t, gestational age (weeks).

and then gradually until 30 weeks; it gradually decreased from 30 weeks onward, yielding a convex growth pattern across gestation.

We obtained 45 TCD and APCD measurements from 33 fetuses with trisomy 18, with gestational ages ranging from 17 to 35 weeks. The average regression formulae for the TCD and APCD values of trisomy 18 fetuses were TCD $(t) = 7914.74e^{0.000153t} - 7920.90$ (mm) (adjusted $R^2 = 0.994$) and APCD $(t) = 126946.41e^{0.00000430t} - 126948.89$ (mm) (adjusted $R^2 = 0.979$), respectively.

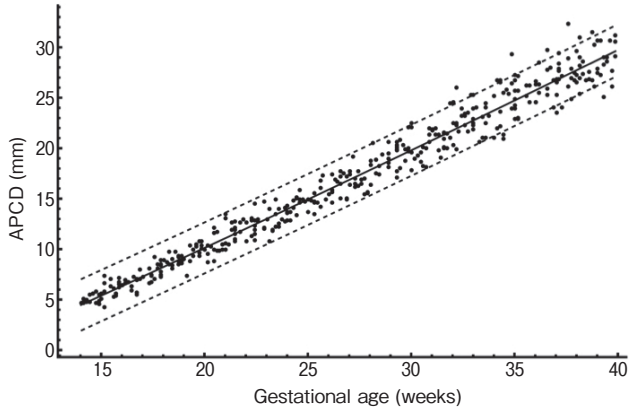

Fig. 3 Scatterplots and regression curve with 95% confidence interval for the anteroposterior cerebellar diameter (APCD) in normal fetuses against gestational age. APCD $(t) = (324.29 \pm 307.66)e^{(0.00286 \pm 0.0026)t} - (322.62 \pm 308.47)$ (estimated value ± standard error), (adjusted $R^2 = 0.995$); t, gestational age (weeks).

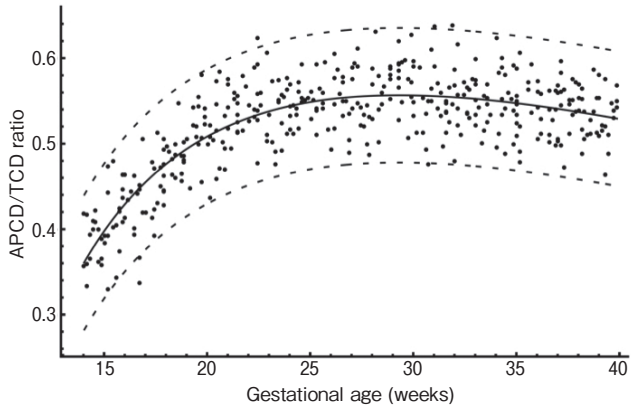

Fig. 4 Scatterplots and regression curve with 95% confidence interval for the ratio of the anteroposterior cerebellar diameter (APCD) to the transverse cerebellar diameter (TCD) in normal fetuses against gestational age. APCD/TCD $(t) = (324.29e^{0.00286t} - 322.62)/(27.85e^{0.02788t} - 28.62)$, (adjusted $R^2 = 0.529$); t, gestational age (weeks).

The x-coordinates of the intersection for TCD, APCD, TCD × APCD, ln (TCD × APCD), $\sqrt{\text{TCD} \times \text{APCD}}$ and ln $(\sqrt{\text{TCD} \times \text{APCD}})$ excluding the APCD/TCD ratio were 25.6, 21.9, 27.8, 20.1, 21.2 and 20.2 weeks of gestation, respectively. The value of ln (TCD × APCD) (20.1 weeks of gestation) was the lowest of these values (Fig. 5). The APCD/TCD ratio was not useful for predicting trisomy 18 because the incidence of trisomy 18 fetuses within the 95% CI of normal fetuses was 42.2%, which is too large to distinguish trisomy 18 fetuses sufficiently (Fig. 6).

Discussion

Our biomathematics-based study found that cerebellar length for both TCD and APCD showed exponential growth throughout gestation. The growth processes of the weight and volume of most organs in mammals demonstrate a sigmoid or S-shaped pattern, and some growth curves (*e.g.*, logistic and Gompertz curves) derived from differential equations have been used with human data [6,12]. Many studies using ultrasonography or magnetic resonance imaging have examined TCD growth [1,2] or cerebellar volume growth [13,14] before birth, showing an exponential pattern. Cerebellar volume growth after birth has been shown to increase rapidly until age 2 and peak at 12-14 years of age [15,16]. Cerebellar growth is thus consid-

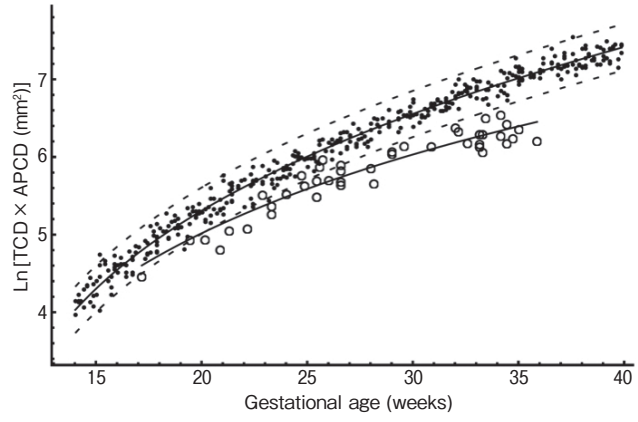

Fig. 5 Scatterplots and regression curve for ln (TCD × APCD) in trisomy 18 fetuses against gestational age. The x-coordinate of the intersection of the regression curve and the lower limit of the 95% confidence interval curve of the nomogram was 20.1 weeks of gestation. Closed circles and open circles represent normal and trisomy 18 fetuses, respectively. TCD, transverse cerebellar diameter; APCD, anteroposterior cerebellar diameter.

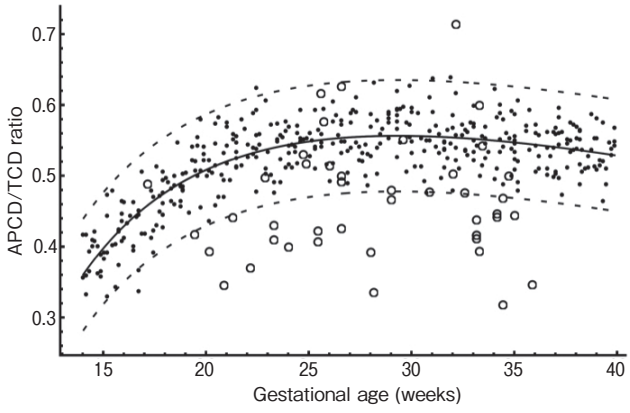

Fig. 6 Scatterplots for the ratio of the anteroposterior cerebellar diameter (APCD) to the transverse cerebellar diameter (TCD) in trisomy 18 fetuses against gestational age. Reference curves represent the regression curve with 95% confidence interval for the nomogram of the APCD/TCD ratio. Closed circles and open circles represent normal and trisomy 18 fetuses, respectively.

ered to show an S-shaped pattern from early pregnancy to puberty. The early stages of an S-shaped growth pattern biologically show an exponential function [6, 12]. As the fetal period is an early stage for cerebellar growth, the present finding that the TCD and APCD growth curves are expressed by an exponential function is biologically significant.

The advantage of using biomathematics for growth analysis is that the resulting equation itself has biological significance. The coefficients of a formula based on biomathematics have biological meaning, though those of a polynomial formula do not. Both TCD and APCD herein can be expressed as an exponential function of time, as in Equation (4), and the coefficient a in that formula represents the growth rate per time period (*i.e.*, per week). To the best of our knowledge, it is a novel finding that TCD and APCD in a normal fetus grow at a constant rate with increases of 2.788%/week and 0.286%/week, respectively. Two recent reports found that APCD growth is more restricted than TCD growth in fetuses with trisomy 18 [4, 5]. The present cerebellar growth analysis in trisomy 18 fetuses revealed low increase rates of 0.0153%/week and 0.000430%/week for TCD and APCD, respectively. Thus, our analysis also quantitatively revealed an extremely lower growth rate of APCD than TCD growth rate in trisomy 18 fetuses.

To our knowledge, there have been only two reports to date in which fetal APCD was measured [4, 5]. In the present study, the profile of the APCD/TCD ratio throughout gestation showed not a linear but rather a convex upward pattern (Fig. 4). There is, however, a single report in which the ratio remained constant at 0.52 throughout gestation by linear regression [5]. We found that the profiles of the ratio throughout gestation in other studies were similar to those in our present study, based on our calculations using the values in those studies [2, 4]. The finding in the present study that the fAPCD/fTCD ratio of the two regression functions seems to fit the data well suggests that these two functions derived from differential equations may better model the actual fetal growth than the previous use of a constant ratio throughout gestation [5]. Therefore, the hypothesis that the TCD and APCD grow independently may be statistically correct.

Our results indicate that the ln (TCD × APCD) function is the best for distinguishing trisomy 18 by this unique method, rather than the APCD [4, 5] or the APCD/TCD ratio [5]. The APCD/TCD ratio was not useful for distinguishing trisomy 18 in this study with 45 samples, though there is a report with only 15 samples in which the ratio was found to be useful [5]. The APCD/TCD ratio must be applied with care in clinical scenarios until a verifiable change in the ratio during gestation is established. Because the analysis based on the single formula (4) derived from the differential equation (1) for TCD and APCD demonstrated appropriate fit not only for TCD and APCD but also for the APCD/TCD ratio and TCD × APCD, our assumption concerning fetal cerebellar growth, that the increase rate of the number of cerebellar cells is proportional to the number of cerebellar cells at any given time point, seems to be correct.

The present study used ultrasound imaging to describe fetal cerebellar growth in normally developing and trisomy 18 Japanese fetuses. Recently, many clinical practices have adopted a genetic test using maternal blood to determine the risk that the fetus has certain genetic abnormalities, including trisomy 18 [17]. In the future, new diagnostic methods, such as artificial intelligence that can integrate imaging and genetic testing information, might be developed to detect chromosomal anomalies and/or genetic diseases of the fetus. In conclusion, this study applied biomathematics in its methodology and clinically indicated the potential of biomathematics in the prenatal diagnosis of trisomy 18. The active introduction of biomathematics might be

useful to further our understanding of fetal growth.

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References

1. Chavez MR, Ananth CV, Smulian JC, Lashley S, Kontopoulos EV and Vintzileos AM: Fetal transcerebellar diameter nomogram in singleton gestations with special emphasis in the third trimester: A comparison with previously published nomograms. Am J Obstet Gynecol (2003) 189: 1021–1025.
2. Sherer DM, Sokolovski M, Dalloul M, Pezzullo JC, Osho JA and Abulafia O: Nomograms of the axial fetal cerebellar hemisphere circumference and area throughout gestation. Ultrasound Obstet Gynecol (2007) 29: 32–37.
3. Hill LM, Marchese S, Peterson C and Fries J: The effect of trisomy 18 on transverse cerebellar diameter. Am J Obstet Gynecol (1991) 165: 72–75.
4. Hayata K, Hiramatsu Y, Masuyama H, Etou E, Nobumoto E and Mitsui T: Creation of a cerebellar diameter reference standard and its clinical application to the detection of cerebellar hypoplasia unique to trisomy 18. J Obstet Gynaecol Res (2015) 41: 1899–1904.
5. Takano M, Nakata M, Oji A, Nagasaki S, Umemura N, Maemura T and Morita M: Utility of fetal anteroposterior to transverse cerebellar diameter ratio to exclude cerebellar hypoplasia in trisomy 18. J Obstet Gynaecol Res (2018) 44: 1031–1035.
6. Karkach AS: Trajectories and models of individual growth. Dem Res (2006) 15: 347–400.
7. Hoppensteadt F: Mathematical biology. Scholarpedia (2007) 2: 2877.
8. Jungck JR: Mathematical biology education: Modeling makes meaning. Math Model Nat Phenom (2011) 6: 1–21.
9. Miyagi Y, Tada K, Takayoshi R, Oguni N, Sato Y, Shibata M, Kiyokawa M, Hashimoto T, Takada T, Oda T and Miyake T: Formulae based on biomathematics to estimate the standard value of fetal growth of Japanese. Acta Med Okayama (2018) 72: 115–119.
10. Tada K, Miyagi Y, Nakamura K, Yorozu M, Fukushima E, Kumazawa K, Nakamura M and Kageyama M: The optimal prepregnancy body mass index for lactation in Japanese women with neonatal separation as analyzed by a differential equation. Acta Med Okayama (2021) 75: 63–69.
11. International Society of Ultrasound in Obstetrics & Gynecology Education Committee: Sonographic examination of the fetal central nervous system: guidelines for performing the 'basic examination' and the 'fetal neurosonogram'. Ultrasound Obstet Gynecol (2007) 29: 109–116.
12. Lampl M: Perspectives on modelling human growth: mathematical models and growth biology. Ann Hum Biol (2012) 39: 342–351.
13. Chang CH, Chang FM, Yu CH, Ko HC and Chen HY: Assessment of fetal cerebellar volume using three-dimensional ultrasound. Ultrasound Med Biol (2000) 26: 981–988.
14. Scott JA, Hamzelou KS, Rajagopalan V, Habas PA, Kim K, Barkovich AJ, Glenn OA and Studholme C: 3D morphometric analysis of human fetal cerebellar development. Cerebellum (2012) 11: 761–770.
15. Wu KH, Chen CY and Shen EY: The cerebellar development in Chinese children–a study by voxel-based volume measurement of reconstructed 3D MRI scan. Pediatr Res (2011) 69: 80–83.
16. Knickmeyer RC, Gouttard S, Kang C, Evans D, Wilber K, Smith JK, Hamer RM, Lin W, Gerig G and Gilmore JH: A structural MRI study of human brain development from birth to 2 years. J Neurosci (2008) 28: 12176–12182.
17. Benn P, Cuckle H and Pergament E: Non-invasive prenatal testing for aneuploidy: current status and future prospects. Ultrasound Obstet Gynecol (2013) 42: 15–33.