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Sonographic Correlation between Distal Femoral Epiphysis Ossification Centre Diameter and Gestational Age in the Third Trimester of Pregnancy: A Prospective Cohort Study

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Abstract

Background: Accurately determining gestational age during the third trimester is crucial for appropriate prenatal care. As seen on sonograms, the ossification center of the distal femoral epiphysis could serve as a dependable marker for this purpose.

Objective: This study aimed to correlate the sonographic distal femoral epiphysis ossification center diameter (DFOC) and the gestational age calculated by crown rump length (CRL) in the period between 28 weeks and 40 weeks gestation

Methods: A prospective observational study included 74 women in their third trimester of pregnancy. The study was conducted at Badr University Hospital, Helwan University, from January 2024 to June 2024. Our Primary outcome was the correlation between DFOC and gestational age and its diagnostic accuracy in the prediction of Gestational age. In contrast, the Secondary Outcome was Ultrasound biometry of studied cases (BPD, AC, HC, and FL).

Results: The findings showed a significant correlation between the diameter of the distal femoral epiphysis ossification center and gestational age in the third trimester. At 32 weeks of gestation, the distal femoral epiphysis has a sensitivity of 68.2% and a specificity of 67.5%. Its positive predictive value (PPV) is 95.13%, and its negative predictive value (NPV) is 18.5%. The overall accuracy of the indicator at 32 weeks is 72.4%. At 36 weeks of gestation, the sensitivity increased to 98.3%, and the specificity increased to 82.7%. The positive predictive value was 97.6%, and the negative predictive value was 81.0%. The accuracy of the distal femoral epiphysis as an indicator of gestational age at 36 weeks was 96.9%.

Conclusion: As measured by ultrasound, the diameter of the ossification center in the distal femoral epiphysis seems to be a potential non-invasive approach for estimating gestational age during the third trimester.

Keywords: Pregnancy, Distal Femoral Epiphysis, gestational age

Introduction

Precise dating of pregnancy is crucial for effective management throughout all three trimesters. It helps determine fetal viability in cases of premature labor and postdate deliveries and is a vital predictor of perinatal mortality and morbidity. Accurate gestational age estimation is essential for antenatal care, obstetric decisions, and assessing fetal growth. If gestational age is miscalculated, misdiagnosis of abnormal growth patterns, like macrosomia or fetal growth restriction, can occur. Reliability in determining gestational age is also essential for calculating the rate of preterm deliveries in a population (1).

In numerous circumstances where high-risk pregnancies frequently occur, women often arrive late for their initial antenatal appointment or present themselves during labor. This creates difficulties in managing complications, evaluating fetal development, and implementing evidence-based treatments, such as in situations of threatened preterm labor, where the administration of corticosteroids is essential for fetal lung maturation (2).

The age of a fetus can be assessed using several measurements, including the biparietal diameter, abdominal circumference, head circumference, kidney length, crown-rump length, long bone lengths, and ossification centers. The bones of the human skeleton originate from distinct ossification centers, divided into primary and secondary types (3).

The femur is positioned at an angle of 30 to 70 degrees relative to the body's long axis. Its accuracy in determining gestational age is comparable to biparietal diameter. The typical growth rate of the femur is less than 2 mm per week. The emergence of limb buds during the fourth week of gestation signals the

onset of femur development in the embryo. This process is followed by mesenchymal proliferation and endochondral ossification by the eighth week of gestation. The primary ossification center forms in the femoral shaft. By six months of age, the secondary ossification begins at the upper part of the bone. The head of the femur starts to ossify between the fourth and fifth months of gestation. The seventh month of gestation is characterized by the appearance of the ossification center in the distal femur (4).

Epiphyseal ossification centers are formed later in pregnancy when standard biometry calculations are unreliable. As a result, when a mother comes for her first antenatal checkup and ultrasound examination late in the third trimester with uncertain dates, these ossification centers can help establish the gestational age. The Distal Femur Epiphysis (DFE) is located at the lower end of the femur and appears as a bright area on an ultrasound. It is found along the inner and outer surfaces of the distal epiphysis in the axial plane and is measured from the outer edges. The DFE is not typically visible on ultrasound before 28 weeks and generally becomes apparent, on average, between 32 and 33 weeks of pregnancy. By the 34th week of gestation, 94% of fetuses are observed to display the DFE. Therefore, if the DFE is not visible, it is highly unlikely that the fetus is younger than 34 weeks (5,6).

This research aimed to establish a connection between the diameter of the ossification center of the distal femoral epiphysis in sonograms and the gestational age estimated using crown rump length (CRL) from 28 to 40 weeks of gestation.

Patients and methods

This prospective observational study was con-

Table 1. Demographic data of the studied patients

Study population	N=74
Age (years)	
Mean ± SD	33 ± 4.4
Weight (kg)	
Mean ± SD	65 ± 9
BMI (kg/m ²)	
Mean ± SD	28 ± 1.3

ducted on 74 pregnant women attending the obstetrics and gynecology outpatient clinic at Badr University Hospital, Helwan University, from January 2024 to June 2024. The study gained ethical committee approval from the Faculty of Medicine, Helwan University, number 58-2023. All procedures were done per the Declaration of Helsinki. The study complies with STROBE guidelines for observational studies.

Inclusion criteria: Pregnant women between the ages of 20 and 40 with a single pregnancy and a gestational age ranging from 28 to 40 weeks. Participants should have had consistently regular menstrual cycles in the three cycles before becoming pregnant and should not have used hormonal contraception during those three cycles. All women should have

routine ultrasonography 1st trimestric scan done between 11 and 14 weeks, confirming the dates.

Patients were excluded if they had fetal congenital anomalies, fetal growth restriction, fetal macrosomia, hypertension, Diabetes with pregnancy, and amniotic fluid abnormalities.

Procedure: All pregnant women involved in the study underwent the following methods: Each participant received an explanation of the study and provided informed consent. They also underwent a comprehensive history-taking and clinical examination. Gestational age was calculated from the first day of the last menstrual period (LMP) and confirmed by first-trimester ultrasound (Crown-rump length).

All women included in the study received transabdominal ultrasound examinations conducted by two sonographers to prevent any bias from the operator. The first sonographer measured the fetal biparietal diameter, femur length, abdominal circumference, and head circumference. Fetal gestational age was estimated using the Hadlock formula ($\log_{10} \text{weight} = 1.335 - 0.0034 \text{AC} \times \text{FL} + 0.0316 \text{BPD} + 0.0457 \text{AC} + 0.1623 \text{FL}$). All ultrasound examinations were performed by two sonographers who were unaware of the clinical features of the women being studied, using a standard ultrasound machine, the LOGIQ P5 from GE Medical Systems, USA. The machine used a 2D abdominal transducer probe with a 1.5-4.5 MHz frequency.

The distal femoral epiphyseal ossification center was identified by the second sonographer as a slit-like, oval, or egg-shaped echogenic structure centrally located within the hypochoic epiphyseal cartilage of the femur at its distal end. It was observed to be 1mm in size and was detected by carefully guiding the transducer along the longest axis of the femoral diaphysis to avoid oblique sectioning. Measurements were obtained millimeters from the outer edges along the mediolateral surfaces of the epiphyses (Figure 1). Once the ossification center in



Figure 1. Distal femoral epiphyseal measured in millimeters from the outer-to-outer margins along the mediolateral surfaces of the epiphyses

Table 2. Different fetal biometric measurements at different gestational ages

G.A (weeks)		DFOC in mm	BPD in cm	HC in cm	AC in cm	FL in cm
28	Range	0-0	7.0-7.4	25.9-26.2	23.04-23.5	2-5.3
	Mean±SD	0-0	7.2±0.1	26.05±0.075	23.27±0.115	5.25-0±025
32	Range	1.8-3.6	8.4-8.7	31.1-31.6	29.3-29.7	6.5-6.6
	Mean±SD	2.7±0.45	8.5±0.07	31.35±0.125	29.5±0.1	6.55±0.027
36	Range	4.9-6.1	9.0-9.2	32.3-32.6	32.2-32.5	7.1-7.3
	Mean±SD	5.5±0.3	9.1±0.05	32.45±0.075	32.35±0.073	7.2±0.05
P value		0.0001*	0.210	0.507	0.199	0.731

DFOC =distal femoral epiphyseal ossification center (mm), BPD=biparietal diameter (cm),FL= femur length (cm),

AC=abdominal circumference (cm), HC=head circumference (cm)

P value <0.001 is highly significant.

the distal femoral epiphysis was identified, three measurements were taken, and the largest diameter in millimeters was recorded. Each patient was measured three times for the ossification center in the distal femoral epiphysis at 28 weeks, 32 weeks, and 36 weeks of gestation.

Primary outcome: The correlation between DFOC and gestational age and its diagnostic accuracy in the prediction of Gestational age

Secondary Outcomes: Ultrasound biometry of studied cases (BPD, AC, HC and FL)

Sample size justification

The Power Analysis and Sample Size Software (PASS 2021) has calculated the required sample size. The primary outcome measure was the distal femoral epiphyseal ossification center for gestational age (GA) estimation. A previous study by Ahmed *et al.* (6) reported that the patients with DFE had a mean adjusted gestational age by USG of 34.39± 2.54 weeks. Pearson Correlation was applied to adjusted gestational age by the US, showing a value of 0.973 for femur length and 0.889 for the size of DFE, indicating a positive correlation. Based on these assumptions, a sample size of 74 pregnant women achieves 99% power to detect a change in slope from 0 under the null hypothesis to 70 under the alternative hypothesis.

Statistical analysis of data: The data was analyzed using SPSS version 22. Qualitative data was presented

in terms of number and percentage. For quantitative data, normality was assessed using the Shapiro-Wilk test. Then, it was described as the mean and standard deviation for normally distributed data and the median and range for non-normally distributed data. The appropriate statistical test was chosen based on the data type, with the suggested tests as follows: Chi-Square for the categorical variable. The accepted level of significance was set at $P \leq 0.05$.

Results

Table (1) presents the demographic data regarding age, weight, and BMI.

Table 2 presents the average values of the distal femoral ossification center (DFOC), biparietal diameter (BPD), head circumference (HC), abdominal circumference (AC), and femur length at 28, 32, and 36 weeks of gestation. The data shows a statistically significant DFOC(mm) increase at 36 weeks. The table indicates no statistically significant difference in BPD, HC, AC, and FL(cm) between weeks 28, 32, and 36. Table 1 shows a significant DFOC (mm) increase in the 36th week.

At 32 weeks of gestation, the distal femoral epiphysis showed a sensitivity of 68.2%, indicating its ability to detect positive cases accurately. Additionally, its specificity was 67.5%, demonstrating its capability to identify negative cases correctly. The positive

Table 3. Diagnostic Accuracy Values of Distal Femoral Epiphysis as an indicator of gestational age at the time of greatest accuracy

Gestational age (weeks)	Sensitivity	Specificity	+PPV	-PPV	Accuracy
32	68.2	67.5	95.13	18.5	72.4
36	98.3	82.7	97.6	81.0	96.9

+PPV: positive predictive value, -PPV: negative predictive value.

predictive value (PPV) was computed at 95.13%, indicating the likelihood that a positive test result shows the condition's presence. In contrast, the negative predictive value (NPV) was 18.5%, showing the probability that a negative test result correctly indicates the absence of the condition. Overall, the indicator's accuracy at 32 weeks was 72.4%.

In contrast, at 36 weeks of gestation, the sensitivity significantly increased to 98.3%, highlighting its improved ability to detect positive cases accurately. The specificity also increased to 82.7%, indicating a moderate ability to identify negative cases correctly. The positive predictive value was notably high at 97.6%, suggesting a high probability that a positive test result indicates the condition's presence. Similarly, the negative predictive value was 81.0%, indicating a high probability that a negative test result correctly shows the absence of the condition. Overall,

the accuracy of the distal femoral epiphysis as an indicator of gestational age at 36 weeks was substantially high at 96.9%.

The correlation between the distal femoral epiphysis diameters in millimeters and the gestational age in weeks was determined using the nomogram and regression curve shown in Figure 2. The correlation between the diameter of the distal femoral epiphysis and gestational age was positive.

There was a statistically significant association between the detection of DFOC and GA detected by FL. DFOC was first detected at 29th – 30th weeks gestation, FL was 58 ± 10 mm, and DFOC was 2 mm. At ≥ 37 th weeks' gestation, FL was 70 ± 20 mm, and DFOC was 5 mm with a ratio of 7% (CI: 0.83 to 0.95) with a sensitivity of 82.8%, specificity of 79.3%, positive predictive value (PPV) of 95% and overall accuracy of 80%

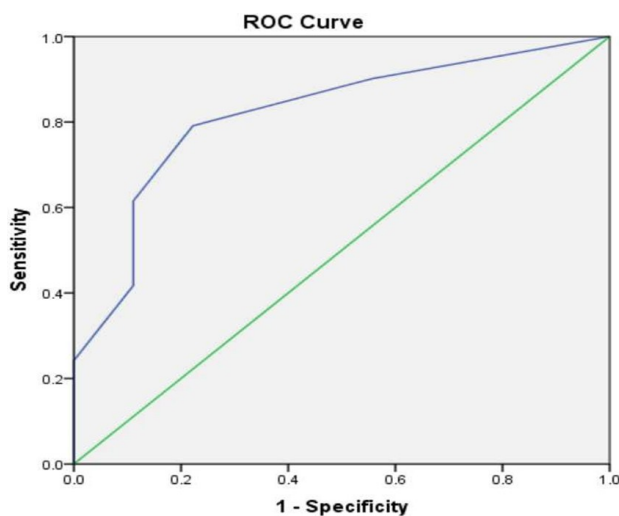


Figure 2. ROC Curve showing diagnostic performance of DFOC in assessment of gestational age

Discussion

Estimating gestational age is crucial in obstetric management. Reliable gestational age estimation is particularly important in regions with high rates of neonatal and maternal mortality. Many women are uncertain about their last menstrual period (LMP) or have irregular cycles, which can lead to discrepancies in gestational age estimation (7).

Ultrasound is safe for fetal measurements, but traditional measurements like CRL, FL, BPD, and AC are less accurate in the 3rd trimester. In the late stages of pregnancy, epiphyseal ossification centers start to appear. This can help determine gestational age when a woman is unsure of her dates, especially dur-

ing her first visit in the third trimester. The femoral epiphysis appears around 28 weeks, and its size can be observed to correspond to fundal height in an uncomplicated pregnancy, making it a potential marker for determining gestational age in the third trimester in the future (8-9).

DFOC reference charts are commonly utilized in certain nations. In Egypt, minimal research has been conducted on the correlation between the diameter of the distal femoral epiphysis ossification center and the advancement of gestational age from 28 to 40 weeks using sonography. Thus, it was essential to conduct a study within the population of Helwan City.

Comparison of our results and similar studies

Consistent with our findings, an earlier study by Goldstein *et al.* (1988) involving 228 pregnant women in the United States revealed that the distal femoral epiphysis was not visible until after 28 weeks. However, this structure was observed in 72% of fetuses at 33 weeks and 94% at 34 weeks gestation. In 87.0% of the cases, the presence of a distal femoral epiphysis measuring between 1 and 2 mm was associated with a gestational age exceeding 33 weeks, while an epiphysis measuring 3 mm or larger was correlated with a gestational age greater than 37 weeks in 85% of the fetuses (10).

Several studies correlated DFOC with other modalities, such as fetal weight and fetal lung maturity.

According to our study, research by Udoh *et al.* (2020) involving 999 Nigerian women showed that the distal femoral epiphysis ossification center (DFOC) was absent during the 28th week of gestation. A correlation coefficient 0.85 indicates a strong positive association between DFOC and gestational age (GA). Furthermore, a correlation coefficient of 0.83 suggests that DFOC increases as fetal weight rises. The formula $GA = 30.84 + (1.25 \times DFOC \text{ in mm})$ defines the relationship between gestational age and DFOC (7).

Elsaeed *et al.* (2017) researched 100 pregnant

women in Damanhur City and discovered that the DFOC increased significantly as gestational age increased (similar to our study). The average DFOC was notably lower in neonates with respiratory distress syndrome ($p < 0.05$). The study also revealed a significant positive correlation between DFOC and APGAR score at 5 minutes (8).

Like our research, Donne *et al.* (9) performed a study involving 377 Brazilian women between 30 and 40 weeks of gestation. They identified and measured the ossification centers of the distal femur, proximal tibia, and proximal humerus. A nomogram was created to represent fetal bone development by incorporating the measurements of the three diameters. The findings indicated a strong correlation between gestational age and the diameters of the distal femoral and proximal tibial epiphyseal ossification centers, with an even stronger connection to the combined measurements of the three centers. When the measurements of the three centers were 7, 11, and 13 mm, the positive predictive values for the fetus at least 37 weeks gestation were 82%, 94%, and 100%, respectively.

In a recent examination of 150 pregnant women in Egypt, Madkour *et al.* (1) discovered that they measured the ossification centers of the Distal Femoral and the Proximal Tibia and correlated them to BPD, HC, AC, FL, and GA. The study's findings showed that the optimal DFOC cut-off for predicting maturity ($GA \geq 37$ weeks' gestation) was ≥ 4.4 mm, with an area under the curve of 0.88 (CI: 0.83 to 0.94), a sensitivity of 82.8%, a specificity of 79.3%, a positive predictive value (PPV) of 95%, a negative predictive value (NPV) of 49%, and an overall accuracy of 80% ($p < 0.001$).

In line with our findings, Mwangirus (2020) observed comparable results within the Kenyan population, revealing that DFOC was not detectable before the 30th week of gestation but was identified in 72% of fetuses at 33 weeks, 86% at 35 weeks, and 100% at 37 weeks.

By 37 weeks, DFOC measured approximately 4 mm, while PTOC was recorded at 2.7 mm. DFOC and PTOC displayed a substantial positive predictive value for assessing gestational age during the final trimester. They utilized the dimensions of DFOC and PTOC to create charts for gestational age estimation. (11).

Abd EL-Fattah and colleagues (2018) noted that evaluating the ossification centers can confirm fetal maturity. The distal femoral epiphysis typically emerges around 32-33 weeks' gestation and increases in size as pregnancy progresses. Additionally, the proximal humeral epiphysis detection through ultrasound is associated with a mature amniocentesis lung profile. The ossification centers become visible after the 31st week of gestation (12).

Consistent with our research, Birang *et al.* (13) demonstrated that using ultrasound to observe the ossification centers of the epiphysis can serve as a valuable indicator of fetal gestational age. The Distal Femur Epiphysis (DFE) was observed in a minority of fetuses (17%) as early as the 29th week, compared to the 35 weeks in our study. A DFE measuring 3 mm or more was correlated with a gestational age of over 37 weeks in 84% of fetuses.

Suhail *et al.* (2013) analyzed the mean size of the distal femoral epiphysis (DFE), proximal tibial epiphysis (PTE), and proximal humeral epiphysis (PHE) throughout gestation. The data demonstrated that the proportion of fetuses in which epiphyses have appeared increased with increasing gestational age. The authors argue that correlating gestational age as determined by standard parameters of fetal biometry with the presence or absence of the DFE, PTE, and PHE increases the accuracy of GA estimation (14).

Scott *et al.* reported a sonographic study on 30 malnourished fetuses and observed that the DFE was absent in 37 percent (11 of 30) and none of the normal fetuses (15).

McLeary *et al.* carried out a sonographic assessment of the distal femoral epiphyseal ossification

center and identified a significant correlation between the ultrasonic and radiologic characteristics of the ossific center. They observed that the ossification of the distal femoral epiphyseal center starts at 30 weeks of gestation and achieves a 100 percent occurrence by 38 weeks, which aligns with the current study's findings (16).

In the study by Radswiki *et al.*, the distal femoral epiphyseal (DFE) ossification center was first observed at 29 weeks of gestation, with most visible by 33 weeks. A DFE measuring 7 mm or more indicates a gestational age of at least 37 weeks, while the identifiable proximal tibial epiphyseal (PTE) suggests at least 35 weeks. The study concluded that ossification centers aid in estimating fetal gestational age and lung maturity, but their absence does not imply a younger age or lack of lung maturity. It's essential to distinguish nearby structures from the epiphyseal centers, and identification of a DFE may not reflect lung maturity, particularly in complicated cases like those involving diabetic patients (17).

In the study of Nemec *et al.*, they investigated femur development in fetuses with fetal growth restriction (FGR) due to placental insufficiency, using prenatal MRI to analyze femur morphometrics and epimetaphyseal features. It included 111 FGR cases and 111 matched controls, with a mean gestational age of 27 + 2 weeks. The results showed significant differences in femur morphometrics between FGR cases and controls ($P < 0.001$), indicating bone shortening in FGR. However, no significant differences were found in the epimetaphyseal features (epiphyseal shape, $P = 0.341$; metaphyseal shape, $P = 0.782$; epiphyseal ossification, $P = 0.85$). Epiphyseal ossification occurred later in FGR cases, with a median of 33.6 weeks compared to 32.1 weeks in controls ($P = 0.008$). They concluded that FGR cases exhibited femur shortening on MRI, but comparable distal epimetaphyseal features were observed in both groups (18).

A study by Partiot *et al.* reassessed the develop-

ment of distal femoral and proximal tibial epiphyses in a sample of 248 individuals aged 26 to 42 gestational weeks (GW) using medical CT scans from Marseille (2008-2017). It was found that 11% of those aged 26-33 GW had developed distal femoral epiphysis, while individuals aged 38-42 GW exhibited both femoral and tibial epiphyses. These findings indicate that epiphyseal maturation is not a reliable indicator of term status in archaeological contexts. Additionally, no delayed ossification was observed in those who did not survive to 42 GW or in individuals with severe developmental issues, challenging the idea that delayed maturation indicates morbidity (19).

Clinical implications of our study

In the present study, the relationship between the diameters of the distal femoral epiphysis ossification centers and gestational age from 28 weeks to 40 weeks was evaluated. The results indicated a positive correlation between the distal femoral epiphysis ossification center and gestational age, presenting a linear trend from 28 to 40 weeks. This correlation could serve as a reliable metric for determining gestational age in women uncertain about their dates and detecting fetal developmental issues, thus aiding the obstetrician in creating an effective management plan. Incorporating the DFOC measurement in standard prenatal care could improve current methods for estimating gestational age. These methods can be helpful when a woman is unsure of her dates, especially when she makes her first visit in the third trimester.

Strengths and limitations of our study

The effectiveness of this study is based on the utilization of a straightforward and easily quantifiable sonographic parameter for determining gestational age. This approach demands less extensive training than fetal biometry and is more efficient in terms of time. Another advantageous aspect is that all ultrasound assessments were carried out by two sonog-

raphers, which reduced the statistical bias. The limitations of this research are that it did not suggest replacing other anthropometric metrics, such as bi-parietal diameter, abdominal and head circumference, or femur length. By examining the diameters of the DFOC and PTOC, we aim to highlight the potential of this straightforward marker of fetal development as a reliable sign of fetal lung maturity. Identifying and measuring the epiphyseal ossification centers might be less affected by fetal growth restriction or excessive growth than other anthropometric metrics. Conversely, a deficiency in calcium metabolism might occasionally postpone the emergence of the two epiphyseal ossification centers.

Recommendation for further studies

The measurement of the DFOC and PTOC is essential for assessing fetal maturity and gestational age during the third trimester. Consequently, future research could concentrate on developing growth charts for our population in the United States, Canada, and other countries. Future studies might also aim to illustrate the connection between gestational age and secondary epiphyseal ossification centers (DFOC and PTOC) in complicated cases of pregnancy.

Conclusion

As measured by ultrasound, the diameter of the ossification center in the distal femoral epiphysis seems to be a potential non-invasive approach for estimating gestational age during the third trimester. This method can improve prenatal care by providing precise gestational age assessments and facilitating prompt medical interventions.

Authors contributions

All authors jointly contributed to the conception and design of the study.

Abdelsalam Karim: Design of the study, helped in the review of literature, revision of results and data analysis, writing the manuscript, and submission to the journal

Ahmed Sherif Abdel Hamid aided in revising the study.

Hossam Abdelmgeed Abdou; design of the study, revision of review of literature, and revision of the manuscript

Yasmeen Anwar Ramadan: obtaining ethical committee approval, reviewing the literature, sharing in the collection of Data, patient recruitment

Mai Raafat Hussien design of the study, revision of the review of literature, and revision of the manuscript

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Disclosure of Interest

The authors declare no conflict of interest.

Ethics Approval and Informed Consent to Participate

Following local regulations, The study gained ethical committee approval from the Faculty of Medicine, Helwan University, no 58-2023. All procedures were done under the Declaration of Helsinki.

Data Sharing

The datasets used and analyzed during the current study are available from the corresponding author upon reasonable request.

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Not applicable.

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