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Original Article

Fetal Brain Midline Structure Measurements: Cavum Septi Pellucidi and Corpus Callosum Indices in Multiple Views at Late Gestation

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Abstract

Background: The literature lacks studies specifically focusing on cavum septum pellucidum (CSP) and corpus callosum (CC) indices in late gestation. In this study, various CSP and CC indices, as well as frontal lobe measurements were assessed and the potential correlations among these parameters were evaluated.

Methods: A cross-sectional study was conducted at Imam Khomeini Hospital Complex on 150 fetuses at gestational age of 36 weeks and later. CSP length, width, trace length, and area in the axial view; frontal lobe thickness measured from the most anterior and posterior parts of CSP in horizontal and vertical directions; CC lengths and thickness; and CSP trace length and area in midsagittal view were all obtained via transabdominal ultrasound. SPSS version 23 was used for data analysis.

Results: The gestational age, biparietal diameter (BPD), head circumference (HC), and estimated fetal weight (EFW) Mean±SD were 37.56±1.08 weeks, 90.73±3.20 *mm*, 326.58±10.59 *mm*, and 2987.82±333.68 *gr*, respectively. In the trans-thalamic axial view, medians (IQR) CSP length, width, trace length, and axial surface area were 8.92 *mm* (7.79-9.90), 6.05 *mm* (5.10-6.71), 2.81 *mm* (2.51-3.11), and 0.49 mm^2 (0.39-0.59), respectively. Median CSP height in the coronal view was 5.85 *mm* (4.93-7.00). In midsagittal view, mean±SD CC outer-to-outer length, inner-to-inner length, CSP trace length, and area were 41.31±4.81 *mm*, 29.73±4.17 *mm*, 2.56±0.63 *mm*, and 0.36±0.19 mm^2, respectively. All CC and CSP indices correlated significantly with HC ($p<0.05$). Vertical and horizontal distances from the frontal bone to the anterior and posterior CSP parts in axial and near-field views also showed significant correlations with HC ($p=0.0001$).

Conclusion: This study presented normative data and new indices including fetal CC, CSP, and frontal lobe measurements in prenatal sonographic exam of fetuses at gestational age of 36 weeks and later.

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Introduction

Cavum Septum Pellucidum (CSP) and Corpus Callosum (CC) are two important midline fetal brain structures. CSP development begins at 10-12 weeks of gestation and reaches its maximum size at 17-18 weeks (1, 2). CC or CSP detected abnormalities in prenatal imaging can indicate important fetal central nervous system (CNS) developmental disorders such as holoprosencephaly, CC agenesis, or septo-optic dysplasia (SOD) (1-4). According to the American Institute of Ultrasound in Medicine (AIUM) and many other valid international guidelines, visualization of CSP is an essential element in assessing fetal brain anatomy during prenatal period, specifically in the trans-thalamic axial view. Its presence should be confirmed from 18 to 37 weeks of gestation (5). The septum pellucidum leaflets begin to fuse in a posterior-to-anterior direction around the 24th week of gestation, and by six months after birth, the cavity of the septum pellucidum is obliterated in more than 85% of infants (6). Additionally, CSP and CC embryologic development is closely interrelated. Primary callosal axons also appear around the 74th day of embryonic life. The genu and splenium segments begin to form by approximately the 84th day, and the adult-shaped corpus callosum becomes detectable by around the 115th day of embryonic life (7).

Various imaging modalities have been used to assess CC in prenatal period, with two-dimensional ultrasonography (2D US) being the simplest. However, it has some main limitations such as difficulties in acquiring true midsagittal views, mostly in non-vertex fetal positions (8). Three-dimensional ultrasonography (3D US) has overcome some of the limitations of 2D US. It could create a virtual midsagittal reconstructed image from axial views. Despite this, 2D US provides images with higher quality. Moreover, in 3D imaging, only the external counter of CC could be visualized, which limits accurate measurement of CC thickness (9). Other studies have also used MRI for CC visualization both prenatally and postnatally (3, 10-12). Moreover, a deep attention network, based on U-Net using encoders and post-processing techniques, has been introduced to automatically measure CSP indices from trans-thalamic axial US images. Although US measurement of CC and CSP had been the focus of many previous studies, research evaluating CC or CSP indices in late gestation is lacking and most studies have provided normative references in a wide gestational range, such as 17 to 41 weeks. Therefore, the sample sizes of subgroups were limited, especially in late gestation. Moreover, fetal MRI is not available in many settings, is more expensive, and requires expertise in image interpretation, which is lacking in resource-limited countries. The accurate assessment of fetal brain midline structures during late gestation is critical for understanding neurodevelopment and ensuring optimal prenatal care. As advancements in imaging technologies continue to evolve, various indices have been developed to measure these structures from different views, each offering unique insights into fetal health. Understanding the correlation between different indices of fetal brain midline structure and clinical outcomes is critical, as each parameter contributes valuable information to a comprehensive understanding. Accurate measurements can help the early identification of at-risk infants, providing a golden opportunity for timely interventions. For instance, when abnormalities are detected, targeted therapies can be implemented ahead of schedule, potentially improving neurodevelopmental outcomes (13). Hence, the biometric indices of CC and CSP using 2D US were investigated in the current study, specifically from 36 weeks of gestation and later. Different indices, including length, width, height, area, and trace diameters, were measured across multiple views (trans-thalamic, midsagittal, and coronal). Additionally, the potential association between the indices of these two main midline brain structures and fetal frontal lobe measurements was assessed.

Methods

Study design: This cross-sectional study was done during 2021-2023 at Maternal and Fetal Clinic of Vali-E-Asr Hospital, Imam Khomeini Hospital Complex, Tehran, Iran, which is a tertiary referral center for fetal and maternal medicine. The Ethical Committee of Tehran University of Medical Sciences approved the study (IR.TUMS.IKHC.REC.1400.463) and informed consent was taken from all participants.

Sampling: Pregnant women who came to our center at the gestational age of 36 weeks or later to undergo a third trimester fetal biometric ultrasound scan were enrolled.

Inclusion and exclusion criteria: The inclusion criteria were as follows: Iranian ethnicity; maternal age between 18 to 45 years; willingness for participation; absence of significant pregnancy complications such as gestational diabetes mellitus or gestational hypertension requiring medication; absence of severe intrauterine fetal growth restriction (IUGR); IUGR with accompanying oligohydramnios, or abnormal umbilical or middle cerebral artery Doppler indices; absence of any maternal chronic diseases or uncontrolled thyroid disorders; and absence of any fetal structural malformations or high risk aneuploidy screening results in the current pregnancy.

Participants were excluded if acceptable ultrasound images could not be obtained or if the patient was unwilling to cooperate. As this cross-sectional study aimed to determine the mean values of CSP measurements at term, according to Jou et al.'s study which reported a mean value of 6.37±5 *mm* considering an error of 1 from the standard deviation at a 95% confidence interval, a sample size of 105 participants was required (14).

Transabdominal ultrasound exams were done using Affiniti 70 system (Philips, USA) equipped with a 5–9 *MHz* curvilinear transducer. Cavum septum pellucidum was visualized in an axial trans-thalamic view, with the falx cerebri seen in the midline. The image was completely symmetrical, and the two leaflets of the septum pellucidum appeared as parallel echogenic lines resembling an "equal" sign. The image was then maximally magnified so that CSP occupied more than two-thirds of the screen. The maximum transverse distance between the inner surfaces of the septum pellucidum leaflets, measured perpendicular to the midline, was defined as the transverse diameter of the CSP. The maximum longitudinal diameter of the cavity was measured as the greatest anterior-posterior distance along the falx cerebri. The CSP trace length was measured manually by continuous tracing at this section, and the CSP area was automatically calculated during the trace measurement.

Additionally, the vertical and horizontal distances from the most anterior and posterior points of CSP leaflets in the near field to the inner surface of fetal skull were measured in a standard trans-thalamic axial view. In several cases, fetal ultrasounds performed after 37 weeks of pregnancy for the assessment of fetal weight, biophysical profile, and other routine evaluations also included CSP measurements. CSP height was measured in a magnified trans-thalamic coronal view, with the CSP and anterior horns of both lateral ventricles clearly visualized. These measurements were performed in fetuses that did not have any structural or genetic anomalies before birth, and the data obtained from the study could significantly help identify cases outside the normal range, thereby facilitating the early recognition of infants at risk.

Additionally, after obtaining a standard brain midsagittal view of the fetal brain, the image was magnified so that the entire CC occupied more than two-thirds of the screen. CSP trace length and area were measured manually and automatically, respectively. Regarding the CC, inner-to-inner, outer-to-outer diameters, trace length, and thickness at the splenium segment were measured. All measurements were repeated twice by a feto-maternal subspecialist with over 5 years of experience in fetal ultrasound imaging, and the mean values were recorded.

Statistical analysis: The data analysis was performed using SPSS version 23 (EBM, USA). Descriptive statistics for qualitative variables were expressed as absolute and relative frequencies. Quantitative variables, which were found to be non-normally distributed based on the Kolmogorov-Smirnov test, were summarized using medians, minimum and maximum values, and interquartile ranges. To compare quantitative variables between two groups, Man-Withney U test was applied. The correlation test was used to determine the relationship between two quantitative variables such as CC or CSP indices with BPD, HC, EFW, and body mass index (BMI). To control for potential confounders, a multivariate analysis using general linear model was performed. P-values less than 0.05 were considered statistically significant.

Results

During 2021-2023, a total number of 150 pregnant women were included in the study. Demographic features of the participants and their fetal biometry indices are shown in table 1.

Of 150 fetuses, 81 (54%) were female and 69 (46%) were male. Regarding pregnancy complications, 87 cases (58%) were low risk uncomplicated pregnancies. The two most common comorbidities were gestational diabetes mellitus on treatment, observed in 25 cases (18%) and well-controlled hypothyroidism, observed in 20 cases (13.3%). Less common comorbidities among the study population included gestational hypertension without medication (4 cases, 2.7%), epilepsy (2 cases, 1.3%), gestational thrombocytopenia (2 cases, 1.3%), and asthma (2 cases, 1.3%).

In the trans-thalamic axial view, the medians (interquartile ranges) of CSP length, CSP width, CSP trace length, and CSP axial surface area were 8.92 *mm* (7.79-9.90), 6.05 *mm* (5.10-6.71), 2.81 *mm* (2.51-3.11), and 0.49 mm^2 (0.39-.59), respectively. The median CSP height in the trans-thalamic coronal view was 5.85 *mm* (4.93-7.00). Other CSP and CC indices in the midsagittal view are pre-

Table 1. Demographic features of the participants and their fetal biometry indices

	Maternal age (years)	Gestational age (weeks)	Gravidity (N)	Live birth (N)	Maternal BMI (*kg/m²*)
Mean (±SD)	30.00(±6.31)	37.56(±1.08)	2.20(±1.20)	0.84(±0.93)	29.84(±5.21)
Median	30.00	37.3	2.00	1	29.26
Minimum-maximum	18-44	36.1-41.00	1.00-6.00	0.00-4.00	20.17-48.7
Interquartile range (25-75%)	25-35	36.00-38.40	1-3	0.00-1.00	26.30-32.82
	BPD (*mm*)	**HC (*mm*)**	**AC (*mm*)**	**FL (*mm*)**	**EFW (*grams*)**
Mean (±SD)	90.73(±3.20)	326.58(±10.59)	324.77(±17.52)	71.35(±3.68)	2987.82(±333.68)
Median	90.40	326.00	325.25	71.15	2980.00
Minimum-maximum	76.50-100.76	279.50-355.40	258.50-375.10	60.6-98.60	2300.00-4002.00
Interquartile range (25-75%)	88.700-92.92	320.100-332.77	313.37-336.42	69.600-72.82	2711.50-3178.25
	AFI (*mm*)	**MCA PSV (*mm*)**	**MCA PI (*mm*)**	**MCA RI (*mm*)**	
Mean (±SD)	12.46(±4.29)	55.98(±11.36)	1.89(±1.20)	0.80(±0.06)	-
Median	11.65	56.15	1.86	0.79	-
Minimum-maximum	5.8-23.6	32.00-81.30	1.20-2.87	0.6-0.98	-
Interquartile range (25-75%)	9.4-14.94	46.97-63.70	1.67-2.03	0.76-0.85	-

BMI: Body Mass Index, BPD: Biparietal Diameter, HC: Head Circumference, AC: Abdominal Circumference, FL: Femur Length, EFW: Estimated Fetal Weight, AFI: Amniotic Fluid Index, MCA PSV: Middle Cerebral Artery Peak Systolic Velocity, PI: Pulsatility Index

sented in table 2.

To assess the correlation between CSP and CC indices with fetal biometric parameters, Pearson's correlation was applied. Briefly, it was found that CC and CSP indices in midsagittal view were correlated with fetal head circumference ($p<0.05$) as shown in table 3.

The same tests were used to find out any possible correlation between maternal age, gestational age, and BMI with CC and CSP indices but no significant correlation was observed (all $p<0.05$). For frontal lobe measurements, the horizontal distance from the most anterior part and vertical distance from the most posterior part of the CSP leaflets to the inner surface of the frontal bone in the near field were correlated with gestational age. ($p=0.023$ with $r=0.185$ and $p=0.046$ with $r=0.163$, respectively).

CSP width ($p=0.036$, $r=0.074$), the vertical distance from the frontal bone at the most anterior ($p=0.0001$, $r=0.368$) and posterior parts of CSP ($p=0.0001$, $r=0.473$) in the near field, and the horizontal distance from the frontal bone at the most anterior ($p=0.037$, $r=0.171$) and posterior parts of CSP ($p=0.032$, $r=0.176$) in the axial view were all correlated with BPD.

Similarly, the vertical distances from frontal bone at the most anterior ($p=0.0001$, $r=0.362$) and posterior parts of the CSP ($p=0.0001$, $r=0.433$) in the near field, as well as the horizontal distance from the frontal bone at the most anterior ($p=0.0001$, $r=0.285$) and posterior parts of CSP ($p=0.0001$, $r=0.288$) in the axial view were all correlated with HC. A multivariate analysis using a general linear model was also applied to control for confounding effects; frontal lobe measurements remained statistically correlated with both BPD and HC, and CSP width remained correlated with BPD.

Spearman's test was also applied to assess correlation between CC trace length and outer- to-outer length in the midsagittal view, as well as CSP trace length in the same view, and CSP length and height in axial and coronal views, respectively. CC trace length in the midsagittal view showed a direct correlation with all CSP indices (Table 4).

Additionally, CC, CSP, and frontal lobe indices were compared between male and female fetuses by Mann-Withney U test, and no significant differences were observed (all $p<0.05$). Moreover, CSP trace length in axial view was significantly correlated with FL in multivariate analysis ($p=0.023$).

Discussion

CC development starts at the lamina terminalis, a bundle of fibers connecting the two brain hemispheres, at approximately 12 weeks of gestation.

Table 2. Fetal CSP and CC indices

Midsagittal view measurements				
	CC indices (*mm*)			
	Outer-to- outer	**Inner-to- inner**	**Trace length**	**Thickness**
Mean (±SD)	41.31(±4.81)	29.73(±4.17)	102.41(±9.25)	3.46(±0.60)
Median	41.2	29.30	100.15	3.40
Minimum-maximum	30.9-53.80	18.9-41.80	75.8-137.40	1.5-5.7
Interquartile range (25-75%)	37.92-44.52	26.9-32.72	96.8-108.50	3.13-3.80
	CSP indices			
	CSP trace length (*mm*)		**CSP area (*mm*^{2})**	
Mean (±SD)	2.56 (±0.63)		0.36 (±0.19)	
Median	2.49		0.33	
Minimum-maximum	1.34-4.86		0.07-1.19	
Interquartile range (25-75%)	2.14-2.90		023-0.43	
Frontal lobe measures in the trans-thalamic axial view				
	Vertical distance in the most anterior part of CSP	**Horizontal distance in the most anterior part of CSP**	**Vertical distance in the most posterior part of CSP**	**Horizontal distance in the most posterior part of CSP**
Mean (±SD)	34.74 (±2.41)	33.72 (±3.45)	37.79 (±2.84)	42.37 (±3.82)
Median	34.80	33.50	37.40	42.35
Minimum-maximum	27.90-41.1	26.80-43.30	31.30-52.20	31.80-53.20
Interquartile range (25-75%)	33.00-36.50	31.47-36.02	35.87-39.30	39.80-44.70

This process is concomitant with septum pellucidum formation and CSP visualization should be confirmed between 18 to 37 weeks of gestation. After birth, CSP closes in the majority of cases during early childhood (1, 5). Additionally, CSP might not be visualized on prenatal sonography after 37 weeks of gestation, and previous studies have reported that it is detected in nearly 50% of term neonates on postnatal sonography (15). Although several studies have focused on sonographic assessment of the cavum septi pellucidi (CSP) in term fetuses, the number of fetuses at term or beyond was small because those studies included wide gestational age ranges (15, 16). In this study, 150 fetuses at a gestational age of 36 weeks and later were examined. The results provide obstetricians with reference values for various CSP and CC indices in both axial and midsagittal views. In trans-thalamic axial view, the medians (interquartile ranges) of CSP length, CSP width, CSP trace length, and CSP axial surface area were 8.92 *mm*, 6.05 *mm*, 2.81 *mm*, and 0.49 *mm*^{2}, respectively. These findings are in line with Kertes et al.'s study, who examined CSP width and length in axial view and CSP height and width in coronal view through fetal MRI at gestational age of 25 to 41 weeks (6). Overall, our results regarding CSP measurements are similar to theirs, although CSP width in the axial view showed some differences. They measured CSP through MRI while 2D sonography was used in the current study. Additionally, 57 cases were at gestational age of 37 weeks or later in Kertes et al.'s study, while 150 fetuses were investigated in the current study and this might explain our different findings. Fetal MRI is not widely available in many resource-limited or developing countries, and even when available, expert radiologists may be lacking. Therefore, our results and measurement methods can provide valuable information for obstetricians. In this study, a comprehensive set of indices for the CSP, CC, and frontal lobe thickness across different imaging planes were presented.

The current study showed that all CSP and CC indices in midsagittal view were correlated with fetal HC. Moreover, CC thickness was also correlated with fetal BPD. Multivariate analysis also illustrated that CSP width as well as the vertical

Table 3. Correlation between CSP and CC indices with fetal biometric parameters

	*	BPD	HC	AC	Fl	EFW
CSP length (axial view)	p-value	0.198	0.282	0.167	0.077	02.252
	r	0.106	0.088	-0.114	-0.145	-0.094
CSP width (axial view)	p-value	0.366	0585	0.765	0.817	0.971
	r	0.074	-0.45	0.25	-0.019	0.009
CSP trace length (axial view)	p-value	0.574	0872	0.760	0.012	0.601
	r	0.046	0.013	-0.025	-0.204	-0.043
CSP area (axial view)	p-value	0.467	0.995	0.843	0.172	0.854
	r	0.06	0.000	0.016	-0.112	0.015
CSP height (coronal view)	p-value	0.226	0.118	0.167	0.408	0.266
	r	.100	0.128	0.128	-0.068	0.091
Outer-to-outer (midsagittal view)	p-value	0.153	0.004	0.177	0.624	0.618
	r	0.117	0.232	0.111	-0.040	0.041
Inner-to-inner (midsagittal view)	p-value	0.111	0.011	0.038	0.281	0.186
	r	0.131	0.208	0.170	0.089	0.109
CC trace length (midsagittal view)	p-value	0.332	0.037	0.969	0.688	0.902
	r	0.080	0.171	-0.003	-0.033	0.010
CC thickness (midsagittal view)	p-value	0.030	0.004	0.375	0.804	0.309
	r	0.177	0.236	0.075	0.020	0.084
CSP area (midsagittal view)	p-value	0.444	0.014	0.903	0.282	0.849
	r	0.063	0.200	0.010	-0.088	-0.016
CSP trace length (midsagittal view)	p-value	0.196	0.003	0.824	0.432	0.971
	r	0.106	0.242	-0.018	-0.065	0.003

* Pearson's correlation was applied. Bolded numbers are statistically significant values

Table 4. Correlation between CC and CSP indices

		CSP length (axial view)	CSP height (coronal view)	CSP trace length (sagittal view)
CC trace length (sagittal view)	p-value	0.002	0.01	0.0001
	r	0.257	0.209	0.435
CC outer-to-outer length (sagittal view)	p-value	0.058	0.857	0.009
	r	0.155	-0.015	0.213

distance from the frontal bone at the most anterior and also posterior parts of CSP in the near field, measured in the axial view, were all correlated with BPD (p=0.007, 0.017, 0.001, respectively). Horizontal distance from the frontal bone at the most anterior and also posterior parts of CSP in the near field, measured in the axial view, were also correlated with HC (p=0.031 and 0.010, respectively). Our findings also supported the correlation between CC indices in midsagittal view with CSP indices in midsagittal, coronal, and axial view.

To the best of our knowledge, this study is a novel one which has evaluated a narrow gestational age group (36 weeks of gestation and later) with a notable sample size of 150 fetuses which empowers generalization of the findings. Similarly, a wide range of CSP and CC indices, as well as frontal lobe thickness from midline brain structures to the inner surface of the frontal lobe, were examined, with the latter representing a novel measurement. Finally, it is important to note that single center nature of the study could be considered as a study limitation. Moreover, gestational age was not equally distributed and most cases were at 36 to 38 weeks of gestation, limiting comparisons across gestational age categories. Another limitation of our study is that it was cross-sectional and did not include complete long-term follow-up of the participants.

Conclusion

This study presented normative data for the fetal CC, CSP, and frontal lobe measurements in prenatal sonographic examinations of fetuses at gestational age of 36 weeks and later. It also introduced novel indices for two major fetal brain structures and the adjacent frontal lobes. In fact, the measurement of the fetal brain midline structures, namely CSP and CC, has significant implications for neurodevelopment and the potential diagnosis of congenital anomalies. A properly formed CSP and a structurally intact CC are indicative of normal brain development. Evaluation of the midline structures by ultrasound or MRI provides information about the health of the fetal brain and can be used to plan for the management of the baby after delivery. Additionally, the size of the CSP and the morphometry of the CC have been linked to cognitive performance. This underscores the need for careful anatomical assessment during early life to improve our understanding of cerebral connections and its implications for long-term neurological function. Nevertheless, larger studies with increased sample sizes are needed to establish normative and abnormal reference ranges for the CC and CSP.

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Conflict of Interest

The authors declare no conflict of interest.

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