RESEARCH

True umbilical cord knot detection via active scanning: a prospective study on accuracy and visualization factors

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Abstract

Background True umbilical cord knot (TUCK) is frequently missed in prenatal ultrasound (US), hindering standardized management and risk assessment of adverse perinatal outcomes. This study aimed to assess TUCK detection accuracy using active umbilical cord (UC) scanning and identify factors affecting prenatal visualization.

Methods A prospective study of 378 pregnant women (11–40 weeks) was conducted. Experienced and novice physicians sequentially scanned the full UC, grading umbilical cord ultrasonic image quality (UCUIQ) as sufficient (scale 1), restricted (scale 2), or poor (scale 3). Factors affecting UCUIQ were analyzed using multiple logistic regression, and diagnostic accuracy was evaluated. Cases for diagnosis were confirmed at delivery.

Results Interobserver agreement for UCUIQ grading was excellent (K = 0.979). Gestational week emerged as the primary factor influencing UC visualization (P < 0.05), with ultrasound achieving a diagnostic accuracy of no less than 89.3% for TUCK detection during the 17–26 weeks gestational period.

Conclusions Gestational week significantly influenced TUCK detection, with high accuracy at 17–26 weeks. Active UC scanning during this period improved detection accuracy of TUCK.

Keywords Pregnancy, Umbilical cord knot, Ultrasound, Prenatal diagnosis, Gestational weeks

Introduction

The true umbilical cord knot (TUCK) is an obstetrical phenomenon observed in 0.61%- 3.5% of all deliveries [1-3]. It has been associated with advanced maternal age, obesity, maternal anemia, previous miscarriages,

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multiparity, prolonged pregnancy, male fetus, and long umbilical cord (UC) [1, 2, 4–6]. Although direct evidence remains limited, some experts hypothesize that TUCK may form during early gestation, potentially as early as 9–12 weeks [7–9], when the amniotic fluid volume significantly exceeds fetal size. Studies have demonstrated that TUCK is associated with fetal distress, fetal hypoxia, longterm neurological damage, and a reported 4-to-10-fold increased risk of stillbirth [2, 10-13]. Furthermore, coexisting TUCK and nuchal cord exhibit a synergistic effect on the risk of perinatal death [14]. Recent evidence suggests that excellent obstetrical outcomes can be achieved through antenatal detection of TUCK and appropriate fetal monitoring during pregnancy and delivery [12]. However, not all TUCKs are associated with adverse



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perinatal outcomes. The unpredictable nature of TUCKrelated complications can challenge clinical decisionmaking, potentially leading to unnecessary iatrogenic interventions, such as early induction of labor or cesarean delivery, as well as increased maternal anxiety due to uncertainty regarding fetal well-being [1]. Consequently, further exploration of prognosis and standardized management strategies for TUCK is warranted. Despite the first ultrasonographic diagnosis of TUCK being reported nearly 34 years ago [15], TUCK is frequently identified after delivery [3, 6, 13, 16]. Given that ultrasound (US) is a routine prenatal examination, missed diagnoses of TUCK may impede clinicians'ability to effectively evaluate outcomes and develop evidence-based clinical management guidelines. Therefore, increased attention should be directed toward improving the prenatal detection accuracy of TUCK through optimized US protocols.

However, most of the research articles were focused on improving the diagnostic ability of US to solve the problem of false-positive [17], there were limited studies on the reasons for missed diagnosis of the TUCK by US and how to avoid it. Therefore, we designed a prospective study to assess the detection accuracy of TUCK using active UC scanning and to identify factors affecting the prenatal visualization of TUCK.

Methods

Study design

This prospective study aimed to evaluate the detection accuracy of TUCK using active UC scanning and to

investigate factors influencing the prenatal visualization of TUCK. A detailed flow chart of the study design is presented in Fig. 1.

Study population

Pregnant women aged over 18 years who underwent obstetric US assessment and were scheduled for delivery at our institution were recruited for the study. The study protocol received approval from the institutional review board of our medical center. All participants provided written informed consent before enrollment, explicitly authorizing the publication of de-identified ultrasound images and clinical data."

Inclusion criteria included

- 1. Singleton pregnancies with regular menstrual cycles;
- 2. Gestational age between 11 and 40 weeks;
- 3. Menopausal weeks in line with the estimated gestational age by US.

Exclusion criteria included

- Oligohydramnios (amniotic fluid index ≤5 cm or single deepest pocket <2 cm);
- 2. Single umbilical artery;
- Uncertain dating, where reliable gestational age could not be established by either last menstrual period or first-trimester US;

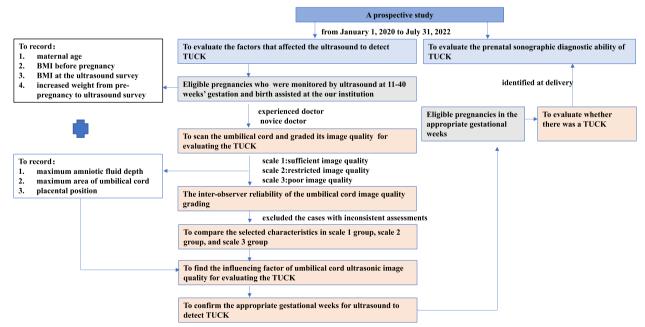


Fig. 1 The flow chart of the prospective study design. TUCK, true umbilical cord knot; BMI, Body Mass Index

4. Extreme obesity (pre-pregnancy body mass index [BMI] >50 kg/m²), which significantly impaired the visualization of the fetal UC on US.

Data collection

The following maternal demographics and pregnancy characteristics were collected at inclusion:

- 1. Maternal age;
- 2. Gestational age (in weeks), calculated from the first day of the last menstrual period to the date of enrollment;
- 3. Pre-pregnancy BMI, calculated within one month prior to pregnancy;
- 4. BMI at the US survey, calculated within one week of the US assessment.

Sonographic examinations

Sonographic examinations were performed by two physicians with a 4–8 MHz curved array volume transducer (GE Voluson E10). One physician had more than 8 years of experience in fetal US examinations, while the other had less than 3 years of experience. Both physicians independently performed US scans and assessed the imaging quality of the entire length of the UC, from the fetal insertion site to the placental insertion site, with a 10-min interval between the two examinations. The experienced physician additionally measured the maximum cross-sectional area of the UC along its outer border and assessed the maximum depth of amniotic fluid at maximum magnification using the US machine's software. The placental position was also recorded, categorized as anterior wall, posterior wall, or other uterine wall locations.

The umbilical cord ultrasonic image quality (UCUIQ) for evaluating TUCK was graded as follows:

- 1. Sufficient image quality (scale 1): The umbilical vessels were clearly identified, and the full length of the UC could be clearly traced, allowing effective evaluation of TUCK.
- 2. Restricted image quality (scale 2): The full length of the UC could be traced, but the umbilical vessels were not clearly identifiable, limiting the evaluation of TUCK.
- 3. Poor image quality (scale 3): Part of the UC was covered by the fetus, preventing effective evaluation of TUCK.

For the cases with sufficient ultrasonic image quality (scale 1), both physicians evaluated the presence of TUCK. Discrepancies were resolved through discussion to reach a consensus.

Diagnostic Criteria for TUCK

- 1. Two-dimension ultrasound (2DUS) Findings: When UC entanglement was detected during prenatal 2DUS examination, especially when a transverse section of the UC was encircled by a loop of UC forming the characteristic"hanging noose sign" [18], it was essential to identify the intersection point of the UC loop and confirm that one end of the UC truly entered into this closed loop (schematic diagram of TUCK is illustrated in Fig. 2). This step was critical for differentiating TUCK from UC twisting or false knot of UC.
- 2. Three-Dimensional High-Definition flow (3D HD-flow) (if applicable): If 2DUS was inconclusive, 3D HD-flow was employed to confirm the diagnosis. The power Doppler mode was activated, and the image was enhanced with optimization of the gain and brightness. Volume data were acquired with a mechanical probe using an angle sweep of 35°to 60°. Digital information was stored for post processing and posterior analysis using the built-in hardware.
- 3. Confirmation Criteria: All cases for diagnosis were confirmed by direct visualization after delivery.

Statistical analysis

Data normality was assessed using the Shapiro-Wilk test. Continuous data were presented as median (interquartile range) for non-normally distributed data or mean ± standard deviation for normally distributed data. Differences in categorical variables were evaluated using the chi-square test or Fisher's exact test, as appropriate. Cohen's kappa coefficients were calculated to evaluate the interobserver agreement of the UCUIQ grading. The interobserver agreement was described as unacceptable (K < 0), poor ($0 \le K < 0.4$), fair ($0.4 \le K$ < 0.6), good (0.6 \leq K < 0.8), and excellent (K \geq 0.8). The comparison of continuous parametric data in three groups grouped according to the UCUIQ was performed using variance analysis, and the Kruskal Wallis test was used when comparing nonparametric data. Additionally, the Odds ratios (OR) and their 95% confidence interval (CI) of the factors affecting the ultrasonic detection accuracy of TUCK were computed by a multivariate logistic regression model. Significance was accepted at p < 0.05, but the multiple comparisons between groups were performed with p-value adjustments according to Bonferroni. Statistical analyses were conducted using the IBM Statistical Package for the Social Sciences (IBM SPSS v.23; IBM Corporation Inc, Armonk, NY, USA).

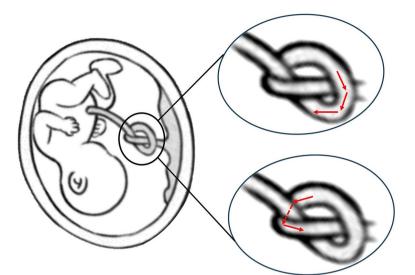


Fig. 2 The diagram of TUCK. When the UC is traced by ultrasound (US) and the "hanging noose sign" is observed, it is crucial to identify the intersection of the UC loop. Additionally, it is necessary to confirm that one end of the UC truly enters into a closed loop. This finding is indicative of a TUCK. UC, umbilical cord; US, ultrasound; TUCK, true umbilical cord knot

Results

The inter-observer reliability of the UCUIQ grading for evaluating the TUCK

A total of 378 singleton pregnant women were scanned by two physicians. The mean maternal age was 29.95 \pm 4.36 years, and the mean BMI before pregnancy and at the US survey were 21.06 \pm 2.82 kg/m²and 23.55 \pm 3.44 kg/m² respectively. The mean gestational age was 24.10 \pm 8.51 weeks of gestation. Interobserver agreement between the experienced and novice physicians for grading the UCUIQ in the evaluation of TUCK was excellent (K = 0.979, *P* < 0.001). A total of 4 cases with inconsistent UCUIQ grading were concentrated before 13 gestational weeks, and the other 1 case was at 30 weeks of gestation. Distribution of cases is presented in Table 1.

The influencing factor of UCUIQ for evaluating the TUCK at 11–40 weeks of gestation

After excluding the inconsistent cases, a total of 373 cases with consistent evaluation were included for analysis. Table 2 presents the selected characteristics of the three groups. Gestational age in scale 1 group was significantly higher than that in scale 2 group (P < 0.001), but lower than that in the scale 3 group (P < 0.001). Higher BMI at the US survey, maximum amniotic fluid depth and maximum cross-sectional area of UC were observed in the scale 1 and 3 groups as compared with the scale 2 group (all P < 0.001). However, whether UC was clearly displayed was not associated with maternal age(P = 0.69), pre-pregnancy BMI (P = 0.271), or placental location(P = 0.190).

Table 1 The interobserver agreement of the UCUIQ grading for evaluating the TUCK

Number		Experien	Total		
		Scale 1	Scale 2	Scale 3	
Novice physician	Scale 1	183	0	0	183
	Scale 2	4	72	0	76
	Scale 3	1	0	118	119
Total		188	72	118	378

The interobserver agreement was excellent (K = 0.979, p < 0.001). TUCK, true umbilical cord knot; UCUIQ, umbilical cord ultrasonic image quality; Experienced physician, > 8 years of fetal US examination experience; Novice physician: < 3 years of fetal US examination experience; Scale 1, Sufficient image quality; Scale 2, Restricted image quality; Scale 2, Poor image quality

According to the results of multivariate logistic regression analysis (Table 3), the risk of grading the UCUIQ for evaluating the TUCK into scale 2 decreased with the increase of gestational age (OR 0.552, 95% CI 0.383– 0.795, P = 0.001). Conversely, as gestational age advanced, the risk of grading the UCUIQ for evaluating the TUCK into scale 3 increased (OR 1.497, 95%CI 1.339–1.673, P < 0.001).

Evaluation results of the appropriate gestational age to evaluate the TUCK

Table 4 shows the percentage distribution of each scale of **UCUIQ** for 373 fetuses grouped by gestational age of 2 weeks. Statistical analysis of the 15 gestational week groups based on the percentage of scale 1 revealed a significant statistical difference (P < 0.001). The scatter plot of the "percentage of scale 1" against the gestational

characteristics	scale 1 (<i>n</i> = 183)	scale 2 (n = 72)	scale 3 (<i>n</i> = 118)	P -Value
 Maternal age(year)	29.72 ± 4.31	30.10 ± 4.22	30.11 ±4.53	0.690
Gestational age(week) *	22.70 ± 4.54	13.32 ± 3.53	33.20 ± 5.47	< 0.001
BMI before pregnancy(kg/m ²)	20.90 ± 2.61	20.92 ± 2.78	21.41 ± 3.18	0.271
BMI at the US survey (kg/m ²) *	23.27 ± 2.95	21.33 ± 3.03	25.43 ± 3.46	< 0.001
Maximum amniotic fluid depth(mm) *	49.38 ± 8.84	40.06 ± 5.31	45.43 ± 9.84	< 0.001
Maximum cross-sectional area of UC(cm ²) *	1.38 ± 0.63	0.29 ± 0.40	2.07 ± 0.63	< 0.001
Placental position in uterus				0.190
Anterior wall	86	30	58	0.838
Posterior wall	90	38	56	
The other wall	7	4	4	

Table 2 The clinical characteristics of the three groups grouped according to the UCUIQ

* Indicates difference between groups (P<0.017 after Bonferonni adjustment). BMI Body Mass Index, cm² square centimeter, kg/m² kilogram per square meter, US ultrasound, UC umbilical cord, UCUIQ umbilical cord ultrasonic image quality

Table 3 Multivariate logistic regression analysis of the risk factors for affecting the ultrasonic detection of TUCK

	Risk of scale 2			Risk of scale 3		
Variables	OR	95%CI	P-Value	OR	95%CI	P-Value
Gestational age	0.552	0.383-0.795	0.001	1.497	1.339-1.673	< 0.001
BMI before pregnancy	1.091	0.643-1.851	0.748	0.991	0.729-1.346	0.953
BMI at the US survey	1.109	0.655-1.879	0.699	1.023	0.774-1.353	0.871
Maximum amniotic fluid depth	0.921	0.859-0.987	0.020	0.969	0.935-1.004	0.080
Maximum cross-sectional area of UC	2.182	0.158-30.197	0.561	0.460	0.211-1.002	0.050

BMI body mass index, US ultrasound, cm² square centimeter, kg/m2 kilogram per square meter, OR Odds ratio, 95% CI 95% confidence interval, UC umbilical cord

Table 4 Percentage distribution of UCUIQ scales across 373 fetuses categorized by 2-week gestational Age Groups

Gestational week	Number of observations	scale 1%	Scale 2%	Scale 3%
11 - 12w	41	9.76%(4/41)	90.24%(37/41)	0.00%(0/41)
13 - 14w	32	15.63%(5/32)	81.25%(26/32)	3.13%(1/32)
15 - 16w	11	54.55%(6/11)	45.45%(5/11)	0.00%(0/11)
17 - 18w	15	86.67%(13/15)	0.00%(0/15)	13.33%(2/15)
19-20w	24	95.83%(23/24)	0.00%(0/24)	4.17%(1/24)
21 - 22w	36	86.11%(31/36)	2.78%(1/36)	11.11%(4/36)
23 - 24w	57	91.23%(52/57)	3.51%(2/57)	5.26%(3/57)
25 - 26w	18	83.33%(15/18)	0.00%(0/18)	16.67%(3/18)
27 - 28w	17	64.71%(11/17)	0.00%(0/17)	35.29%(6/17)
29 - 30w	23	69.57%(16/23)	0.00%(0/23)	30.43%(7/23)
31 - 32w	23	17.39%(4/23)	0.00%(0/23)	82.61%(19/23)
33 - 34w	12	16.67%(2/12)	0.00%(0/12)	83.33%(10/12)
35 - 36w	17	0.00%(0/17)	5.88%(1/17)	94.12%(16/17)
37 - 38w	37	2.70%(1/37)	0.00%(0/37)	97.30%(36/37)
39 - 40w	10	0.00%(0/10)	0.00%(0/10)	100.00%(10/10)

cm² square centimeter, n number, mm millimeter, UC umbilical cord

weeks, demonstrates the appropriate gestational age for detecting TUCK by US is between 17 and 26 weeks of pregnancy (Fig. 3). During this stage, the percentage of scale 1 reached a level above 80%.

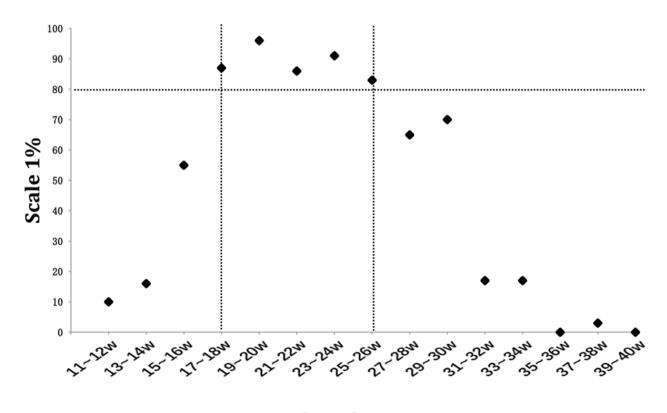
Detection accuracy of evaluating TUCK by US in the 17–26 weeks of pregnancy

A total of 150 pregnant women at 17–26 weeks of pregnancy were enrolled from the recruited 378 pregnancy women. Among them, 134 cases were classified as scale 1 and were suitable for diagnosis, while 3 cases in scale 2 and 13 cases in scale 3 were not suitable for diagnosis. In the scale 1 group, five cases found to have TUCK after delivery (Fig. 4), and 129 cases had no TUCK. All the cases in scale 1 group were correctly diagnosed by the experienced and novice physicians. The accuracy of prenatal US for detecting TUCK at 17–26 weeks of pregnancy was at least 89.3% (134/150).

Discussion

The present study aimed to evaluate the feasibility and diagnostic accuracy of systematic UC scanning for detecting TUCK during pregnancy. Our findings demonstrate that active UC scanning between 17–26 weeks of gestation is highly feasible, with a diagnostic accuracy of at least 89.3%. Gestational age is identified as the primary factor influencing UC visibility and diagnostic performance.

Our findings are particularly significant given the high prenatal sonographic missed diagnosis rate (PSMDR) and the associated risks of TUCK. TUCK has been reported to increase the risk of adverse perinatal outcome, TUCK has been reported to increase the risk of adverse perinatal outcome, particularly when combined with nuchal cord [10-13]. However, the PSMDR of TUCK remains notably high, ranging from 42.8% to 100% [3, 16]. This high PSMDR hinders clinicians from effectively evaluating the perinatal outcomes of TUCK and developing evidencebased clinical management guidelines. According to the previous literature on missed diagnoses and legal aspects [3, 6, 16, 19, 20], three types of causes for a missed prenatal diagnosis of TUCK were used to define. First, the sonographer did not systematically scan the UC. Second, the TUCK was not recognized despite being clearly visible. Third, TUCK was not clearly visible for the poor quality of the images or obstruction by the fetus, which were classified as inevitable causes.



Gestational weeks

Fig. 3 Scatter plot of "scale 1 percentage" with the gestational week. Dotted lines are for guidance only, to demonstrate the appropriate gestational weeks for detecting true umbilical cord knot by ultrasound

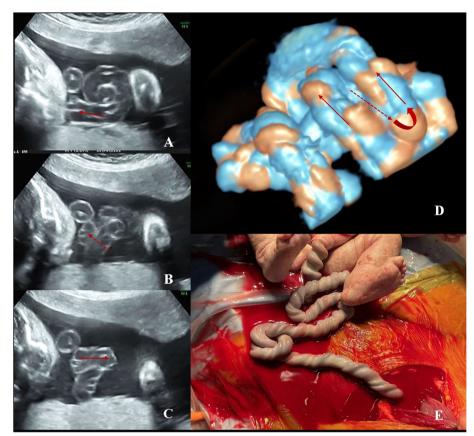


Fig. 4 A true knot of the umbilical cord. Transabdominal Ultrasonography at 25 weeks of gestation: 2DUS demonstrating a transverse section of the UC surrounded by a loop of UC that creates the "hanging noose" sign (**A**). Confirmation was made by 2DUS (**A**, **B**, **C**) and 3D HD-flow (**D**) tracking the UC passing into its loop. And this case was identified at delivery (**E**). 2DUS, two-dimension ultrasound; 3D HD-flow, three-dimensional high-definition flow; UC, umbilical cord

Our results provide actionable insights into improving the detection of TUCK during routine prenatal US examinations. In our study, we found that tracking the UC was less experience-dependent, and 17-26 weeks of gestation represents the optimal window for UC scanning. Over 80% of cases during this period achieved high-quality images (UCUIQ scale 1), enabling reliable diagnosis. Our results are consistent with the findings of Ugurlucan et al. [21]; however, their scans were limited to the second-trimester, and they did not identify the optimal gestational weeks for detecting TUCK. Although it is speculated that most of the TUCKs are likely formed between 9-12 weeks of gestation [8], it is not recommended to systematically scan before 16 weeks, because the UC is too small for effective evaluation. In contrast, during the third trimester, fetal positioning and limited amniotic fluid often obscure the UC, increasing the risk of missed or false-positive diagnoses. Therefore, we recommend systematically scanning the entire UC to avoid missed diagnoses of TUCK, particularly during 17-26 weeks of gestation. Fortunately, scanning the entire UC during second-trimester US imaging does not require significant additional time. According to previous literature [21], the mean time for this procedure is less than one minute.

Additionally, we found that the diagnostic accuracy of TUCK was as high as 89.3% when the UCUIQ was sufficient. This result aligns with the findings of Bohiltea et al., who reduced the PSMDR to 12.5% through systematic UC tracking, a significant improvement compared to their previous study [6, 22]. Therefore, systematic active scanning of the UC may effectively decrease the PSMDR. However, at present, there is an absence of clear guidelines for the recommended prenatal sono-graphic diagnosis of TUCK. Current guidelines do not encourage checking the whole UC length except for the insertion sites [23, 24], which may lead to missed diagnoses of TUCK and increase the risk of adverse obstetric outcomes.

TUCK does not have a characteristic appearance in US, making its diagnosis challenging. However, when UC entanglement is identified on 2DUS, the diagnosis

of TUCK should be approached carefully to avoid false positives. Bohiltea et al. developed an algorithm to aid in the detection of TUCK [6, 22]. When UC entanglement is identified on 2DUS, systematic tracking of the UC should be followed by 3D HD-flow, or by checking for persistent entanglement after fetal movement induction or after 1-2 weeks until reevaluation. However, fetal movement may affect the imaging quality of 3D HD-flow. We believe two key points are critical for accurate diagnosis: First, avoid tracking the entire UC by 2DUS in late gestation, as clumping/clustering of the UC is often observed in limit amniotic fluid pocket during this period, increasing the risk of false-positive diagnoses. Active scanning during 17-26 weeks of gestation is associated with a low falsepositive rate, as demonstrated by Weissmann et al. [25] (3.7%, 2/54). Second, observe whether one end of the UC truly enters into a closed loop, which plays a pivotal role in differentiating a TUCK from UC twisting or a false cord knot.

If a TUCK is not identified during systematic UC scanning in the second trimester, the likelihood of subsequent TUCK formation in the third trimester appears to be substantially reduced, as the available intrauterine space becomes increasingly limited for such configurations to develop. Furthermore, when a TUCK develops prior to the third trimester, spontaneous resolution of the established knot configuration during later gestational stages appears to be an uncommon occurrence, based on current understanding of fetal movement patterns and intrauterine spatial constraints. Therefore, the primary purpose of follow-up sonographic imaging is to monitor changes in umbilical venous or arterial blood flow, coexisting TUCK and nuchal cord, fetal growth restriction, and other relevant factors until fetal maturity is achieved, ensuring the best possible outcome. Regarding the frequency of arterial blood flow detection, Bohiltea et al. suggested that it should be repeated weekly in the last month until term delivery [22].

Our study has the following limitations: First, we excluded cases of oligohydramnios and extreme obesity. Although these conditions are uncommon, they can impact US visualization and warrant further investigation. In our initial study design, we chose to exclude these cases to ensure a more homogeneous study population and minimize potential confounding factors that could affect the clarity and consistency of US imaging. This approach allowed us to establish a baseline understanding of UC visualization under general conditions. In future studies, we plan to include cases of oligohydramnios and extreme obesity and analyze them as separate subgroups. Second, due to the restricted or poor UCUIQ in scale 2 and scale 3 groups, which must decrease the diagnostic ability, we did not highlight the sonographic diagnostic difficulties 17–26 weeks of pregnancy in these two groups; however, 89.3% of cases in this period had sufficient image quality, and the prenatal detection accuracy of TUCK was significantly improved.

Conclusion

In summary, the implementation of comprehensive UC assessment protocols utilizing US technology, with particular emphasis on the 17–26 weeks gestational window, has shown promising results in improving the detection accuracy of TUCK. The integration of evidence-based guidelines detailing optimal examination timing and standardized evaluation methodologies into existing prenatal sonographic protocols is strongly advocated to enhance diagnostic precision and minimize clinical oversight. While these findings suggest a potential pathway toward standardized prenatal management approaches, prospective multicenter studies are necessary to validate these observations and explore their broader clinical implications.

Abbreviations

2DUS	Two-dimension ultrasound
3D HD-flow	Three-dimensional high-definition flow
BMI	Body Mass Index
OR	Odds ratio
PSMDR	Prenatal sonographic missed diagnosis rate
TUCK	True umbilical cord knot
UC	Umbilical cord
UCUIQ	Umbilical cord ultrasonic image quality
US	Ultrasound

Supplementary Information

The online version contains supplementary material available at https://doi. org/10.1186/s12884-025-07629-6.

Supplementary Material 1.

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Authors' contributions

J. C. performed the study, analyzed the data, wrote and revised the manuscript; Z. C. performed the literature search and the statistical analysis, wrote and revised the manuscript; M. Z. and M. W. performed the literature search, analyzed the data and revised the manuscript; M. Z. revised the manuscript; Y. C. and X.L. collected the data; Q.W. performed the study and revised the manuscript; X.Z. designed the study and revised the manuscript; all authors have read and approved the final manuscript.

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Data availability

Data is provided within the manuscript or supplementary information files. the corresponding author upon reasonable request.

Declarations

Ethics approval and consent to participate

This study received approval from the local institutional review boards at the Third Affiliated Hospital of Sun Yat-sen University (Approval number: [2022]02–348-01) on December 5, 2022. Written informed consent was obtained from all participants, which included authorization for both study participation and use of de-identified data for publication purposes.

Consent for publication

Written informed consent was obtained from all participants for publication of de-identified ultrasound images and clinical data.

Competing interests

The authors declare no competing interests.

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