

The Prediction of Maternal Serum Alpha Fetoprotein Level and Fetal Nuchal Translucency, Forcases Involving Preterm Prelabor Rupture of Membranes

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Abstract

Objective: To investigate whether the values of maternal serum alpha-fetoprotein (AFP) and fetal nuchal translucency (NT) during the first and second trimesters can predict preterm prelabor rupture of membranes (PPROM).

Methods: This retrospective case-control study analyzed the first and second trimester screening indicators and maternal outcomes of gravidas who were divided into the Non-PPROM group (594) and the PPRM group (591). Binary logistic regression analysis was used to calculate the OR and 95% CI. ROC and AUC were used for screening performance of AFP and NT.

Results: The values of NT and AFP in the PPRM group were higher than the control group and the difference was statistically significant (all $P < 0.05$). There was no significant difference with respect to PAPP-A, free- β hCG, and the calculated risk of trisomy 18 (all $P > 0.05$). Binary logistic regression showed that NT and AFP MoM were risk factors for PPRM, with ORs of 1.719 and 3.549, respectively. The AUC according to the ROC for NT, AFP MoM and maternal weight in the first trimester +NT+AFP were 0.552, 0.618 and 0.630.

Conclusions: NT and AFP during the first and second trimesters were identified as risk factors for PPRM and valuable markers to predict PPRM in late pregnancy. Multi-index joint prediction was more effective.

Introduction

The definition of prelabor rupture of membranes (PROM) is rupture of membranes before the onset of labor [1]. Membrane rupture before 37 weeks of gestation is considered as preterm prelabor rupture of membranes (PPROM). PROM that occurs preterm complicates roughly 2–3% of all pregnancies, representing a percentage of preterm births, whereas term PROM occurs in approximately 8% of pregnancies in the United States [2]. Although PROM can result from a normal physiologic weakening of the membranes associated with shearing forces created by uterine contractions, PPRM can result from a wide array of pathologic mechanisms that act individually or in concert [3]. Sepsis, necrotizing enterocolitis and intraventricular hemorrhage also are associated with prematurity. PPRM has been associated with an increased risk of neurodevelopmental impairment [4–5].

AFP is the dominant serum protein early in fetal life, which is synthesized in the fetal liver and yolk sac, reaching a maximum concentration in the first and second trimester, thereafter, its concentration drops steadily [6]. The combined examination of AFP, PAPP-A, free β -hCG and fetal NT has been extensively used to screen fetal Down syndrome (DS), open neural tube defects (ONTD), Edwards syndrome (ES) and other diseases [7–8]. However, there are few researches on the prediction of PPRM for AFP, free β -hCG, PAPP-A levels, and fetal NT values. In the retrospective studies conducted by Öztürk H et al., a significant relationship was found between abnormal elevation of hCG and AFP levels and adverse pregnancy outcomes (APO), such as pre-eclampsia, intrauterine growth retardation (IUGR), placenta accreta, PPRM [9].

An accurate assessment of gestational age and knowledge of the maternal, fetal risks are important to appropriate evaluation, counseling, and care of patients with PROM. Expectant management in pregnancies complicated by PPRM is related with an overall neonatal survival rate of 49.0%, high risk of considerable maternal risks [10]. The aim of this study was to assess whether AFP and fetal NT are associated with PPRM and provide evidence for predicting PPRM in pregnancy women.

Methods

Study population

This retrospective case-control study was conducted in Hangzhou Women's Hospital between January 2015 and April 2020. We analyzed the first and second trimester screening indicators and maternal outcomes of gravidas who were divided into the Non-PPROM group (594) and the PPRM group (591). PPRM was diagnosed according to the elevated vaginal fluid, vaginal power of hydrogen measurement, ultrasound examination et al [1, 11]. The exclusion criteria were as follows: (1) alcohol abuse and cigarette smoking; (2) special history of pregnancy, such as infants conceived in vitro, plural gestations; (3) co-existing medical conditions, such as chronic hypertension, diabetes mellitus, hyperthyroidism, heart disease, kidney disease, autoimmune disease, blood diseases and a history of immunotherapy or blood transfusion or special medications during pregnancy; (4) DS, ES, ONTD and other congenital birth defects; (5) missing more than 3 incomplete information; After careful screening, The Non-PPROM group were randomly selected according to the number of the PPRM group (1:1). The study group included 591 with PPRM and the control group included 594 pregnant women of normal labor. All participants provided written informed consent before participating in the study. The study was approved by the local Ethics Committee (Reference number: 2018-004-01).

Blood Samples And Screening Indicators

Take fasting venous blood from pregnant women at 11 weeks to 13 weeks and 6 days of gestation and 15 weeks to 20 weeks and 6 days of gestation. After 30 minutes separation, the serum samples were stored in a refrigerator at 2–8 °C and were sent for examination within 1 week. Screening indicators and programs: (1) Free β -hCG, PAPP-A levels and fetal NT thickness by ultrasound in first trimester; (2) Free β -hCG levels and AFP in second trimester; (3) Joint Screening Program: The results of AFP and free β -hCG in second trimester were matched with those of PAPP-A and NT in first trimester; and the risk rates of T21 and T18 were calculated.

Reagents And Apparatuses

The apparatuses are applied in the study included 1235 Automatic Immunoassay System (DELFLIA) (PerkinElmer, Shelton, USA) for detection with a single labeling kit (PAPP-A, free β -hCG) in first trimester and a double labeling kit (AFP/free β -hCG) in second trimester, enhancement solution and washing liquor

as well as the quality control sample and VolusonE8 ultrasonic apparatus (GE, Boston, USA) and the standard (WallacOy, Turku, Finland).

Nt Thickness Measurement

Fetal NT thickness was screened according to the standards issued by Fetal Medicine Foundation (<https://fetalmedicine.org/education/the-11-13-weeks-scan>). The fetus was normal in case of NT thickness < 2.5 mm, abnormal in case of NT thickness ≥ 2.5 mm. A midsagittal view of the fetus was taken and in a natural posture, the soft tissues overlying the cervical spine was measured by magnifying the image to display the head and upper chest of the fetus only and the maximum thickness of the translucent space between the skin [12]. NT was measured by ultrasound (VolusonE8, GE) at 11 weeks to 13 weeks and 6 days of gestation.

Definition And Calculation Of The Multiple Of Median (mom)

The original concentration levels of PAPP-A AFP, free β -hCG and the values of NT were replaced by MoM value using model analysis and calibrate the MoM values with maternal weight and gestational age [13].

Statistical analysis

All statistical analyses were performed using IBM SPSS Statistics (version 24.0; Chicago, USA). We used a one-sample Kolmogorov-Smirnov test to test data for normality. Skewed data are expressed as medians and percentiles M(P2.5–P97.5). The maternal age and NT values were normally distributed and expressed as mean \pm standard deviation ($\bar{x} \pm s$). Independent t-test was employed between or among groups. The Mann-Whitney U test were used to compare two groups. Binary logistic regression analysis was used to calculate the odds ratio (OR) and 95% confidence interval (CI) for each variable. The best cut off value and AUC were determined by ROC. Besides, the youden index was calculated. $P < 0.05$ was considered to be statistically significant.

Results

Comparison of demographic data between the Non-PPROM group and the PPROM group

Maternal weight in the first and second trimester and gestational age at testing in the second trimester were all significantly higher in the PPROM group than the control group ($Z = 3.584$, $P < 0.001$; $Z = 3.491$, $P < 0.001$; $Z = 2.007$, $P = 0.045$; respectively; Table 1). There was no significant difference with respect to maternal age, gestational age at testing in the first trimester between the two groups ($P > 0.05$)

Table 1
Comparison of demographic data between the non-PPROM group and the PPROM group.

Groups	Non-PPROM (n = 594)	PPROM (n = 591)	t or Z	P
Maternal age (years)	28.40 ± 2.85(28.17–28.63)	28.64 ± 2.85(28.40–28.66)	1.399	0.162
Maternal weight in the first trimester (kg)	52.00(42.40–68.56)	53.50(41.50–70.24)	3.584	< 0.001**
Maternal weight in the second trimester (kg)	53.50(43.50–69.41)	55.00(43.00–71.30)	3.491	< 0.001**
Gestational age at testing in the first trimester(days)	90.00(80.00–97.00)	90.00(79.00–97.00)	0.397	0.691
Gestational age at testing in the second trimester(days)	117.00(108.00–128.00)	118.00(108.00–128.00)	2.007	0.045*
Note: Data are presented as median (P2.5–P97.5). PPRPOM, preterm prelabor rupture of membranes. *P < 0.05; **P < 0.001.				

Comparison of screening indicators (MOM) and the calculated risk of T21,T18

AFP and NT in the PPROM group were 1.03 MoM and 0.96 MoM in Table 2 and these were significantly higher than the Non-PPROM group (0.93MoM, Z=5.995, P< 0.001; 0.93 MoM, t= 2.352, P= 0.019, respectively). In addition, the calculated risk of T21 in the PPROM group was 1/15393 and this was higher than the Non-PPROM group (1/13714; Z = 1.438, P= 0.015). Conversely, the MoM for maternal PAPP-A, free β-hCG and the calculated risk of T18 in the PPROM group did not show any significant differences when compared with the Non-PPROM group.

Table 2
Comparison of screening indicators (MOM) and the calculated risk of T21,T18

Groups	Non-PPROM (n = 594)	PPROM (n = 591)	<i>t or Z</i>	<i>P</i>
PAPP-A MoM	0.95(0.33–2.41)	1.00(0.28–2.35)	0.707	0.480
Free β -hCG MoM in the first trimester	1.07(0.34–3.48)	1.02(0.39–3.44)	0.381	0.703
NT MoM	0.93 \pm 0.25(0.90–0.95)	0.96 \pm 0.24(0.94–0.98)	2.352	0.019*
AFP MoM	0.93(0.52–1.72)	1.03(0.55–1.87)	5.995	< 0.001**
Free β -hCG MoM in the second trimester	1.03(0.34–3.49)	1.02(0.36–3.77)	0.741	0.459
Calculated risk of T21	1/13 714(1/339–1/100 000)	1/15393(1/493–1/100 000)	1.438	0.015*
Calculated risk of T18	1/100 000(1/22092–1/100 000)	1/100 000(1/13914–1/100 000)	0.507	0.612
Data are presented as median (P2.5–P97.5). AFP, α -fetoprotein; hCG, human chorionic gonadotropin; PAPP-A, pregnancy-associated plasma protein A; NT, nuchal translucency; T21, trisomy 21; T18, trisomy 18; MoM, multiple of median. * <i>P</i> < 0.05; ** <i>P</i> < 0.001.				

Results Of Binary Logistic Regression Analysis

Binary logistic regression analysis showed that the ORs and 95% CIs for maternal weight, AFP and NT were 1.030 (95%CI:1.011–1.049), 3.549 (95%CI:2.323–5.421) and 1.719 (95%CI:1.011–2.923) in Table 3, respectively, which were the risk factors of PPRM.

Table 3
Binary logistic analysis of maternal weight in the first trimester, AFP, NT and PPRM

Indicators	β	SE	Wald	df	<i>P</i>	OR	95% CI for OR	
							Lower	Upper
Weight (kg)	0.029	0.009	9.580	1	0.002*	1.030	1.011	1.049
NT(MoM)	0.542	0.271	4.002	1	0.045*	1.719	1.011	2.923
AFP(MoM)	1.267	0.216	34.336	1	< 0.001**	3.549	2.323	5.421
AFP, α -fetoprotein; NT, nuchal translucency; OR, the odds ratio; CI, confidence interval; SE, standard error; MoM, multiple of median; Weight, Maternal weight in the first trimester (kg). * <i>P</i> < 0.05; ** <i>P</i> < 0.001.								

Predictive value of AFP, NT and maternal age separate screening and joint screening for PPRM

ROC analysis was performed to determine the predictive diagnosis value of maternal age, NT and AFP for PPRM (see Table 4 and Fig. 1). At the best cutoff level of 0.98 MoM, AFP predicted PPRM AUC = 0.618(95%CI: 0.583–0.653, $P < 0.001$), with a sensitivity, specificity and youden index of 0.583, 0.603, and 0.186, respectively. Besides, at the best cutoff level of 0.91 MoM, NT accurately predicted PPRM AUC = 0.552 (95%CI: 0.516–0.588, $P = 0.005$), with a sensitivity, specificity and youden index of 0.579, 0.531, and 0.111, respectively. After combining AFP, NT and maternal weight, the AUC increased to 0.630 (95%CI: 0.595–0.664, $P < 0.001$).

Table 4
The value of AFP, NT and maternal age separate screening and joint screening for PPRM.

Indicators	AUC	95%CI	<i>P</i>	cut-off	Sensitivity	Specificity	Jordan index
Weight (kg)	0.556	0.520–0.592	0.002*	52.45	0.591	0.517	0.108
NT(MoM)	0.552	0.516–0.588	0.005*	0.91	0.579	0.531	0.111
AFP(MoM)	0.618	0.583–0.653	< 0.001**	0.98	0.583	0.603	0.186
Weight + NT + AFP	0.630	0.595–0.664	< 0.001**	0.47	0.685	0.508	0.194
AFP, α-fetoprotein; NT, nuchal translucency; AUC: area under curve; CI, confidence interval; MoM, multiple of median; Weight, Maternal weight in the first trimester (kg); Predicted probability, Weight + NT + AFP. * $P < 0.05$; ** $P < 0.001$.							

Discussion

There are few researches about fetal NT values, maternal serum screening indicators and PPRM. Therefore, we used a retrospective case-control study method to explore the correlation and predictive value of AFP and NT with PPRM. The result of the study showed that the level of NT, AFP and the calculated risk of T21 in the PPRM group were higher than the Non-PPROM group and the difference was statistically significant ($P < 0.05$) in Table 2, which is consistent with the report by Başbuğ D et al. Adverse pregnancy outcomes, such as preterm delivery, preterm premature rupture of membranes (PPROM), oligohydramnios and intrauterine growth restriction (IUGR) rates were increased in the elevated AFP group [14]. Conversely, the MoM for maternal PAPP-A and free β-hCG in the PPRM group did not show any significant differences when compared with the control group ($P > 0.05$). Besides, the results of other researches are similar with our study. There were no differences in any major APO between pregnant women with low free β-hCG levels and control subjects, including preterm birth, low birth weight at term, intrauterine fetal death, abruptio placentae, preeclampsia, or PPRM [15–16]. Contrary to our study, low maternal serum PAPP-A levels during the first trimester may reflect a trophoblast invasion defect in the maternal-fetal interface, resulting in subsequent preterm delivery in a retrospective study, particularly PPRM [17].

Binary logistic regression analysis showed that the ORs and AUC of AFP and NT were 3.549, 0.618 and 1.719, 0.552. However, the AUC of Weight + AFP + NT was 0.630 in Table 3 and Table 4, which presented that multi-index prediction combining AFP, NT and maternal age is more effective. The previous researches just mention PPRM as one outcome of adverse pregnancy. There was an established association between an inflammatory intrauterine environment and APO which is independent of gestational age and mediated by the fetal inflammatory response [18]. A recent morphological study showed that inflammation/infection and malperfusion were the two most common pathological changes in the placentas of preterm birth [19]. Placental inflammation did not increase the expressions of AFP in maternal liver, fetal liver and placenta, but reduced the fetal serum AFP levels in a rat model. Placental inflammation can be the potential cause linking APO and AFP [20]. However, there are limitations to this study. Most studies of second-trimester and PPRM are retrospective and include only expectantly managed cases because of selection bias. The basic mechanisms are unclear at present. Therefore, further studies are required to validate and confirm the findings of this study.

The pregnant women in the PPRM group have higher levels of maternal weight in the first and second trimester and the gestational age at testing in the second trimester than women in the Non-PPROM group ($P < 0.05$, Table 1). Maternal obesity is associated with increased systemic inflammation and an increased risk of preterm premature rupture of membranes [18]. A retrospective cohort study presented that a higher risk for preterm premature rupture of the membranes at the upper extremes of BMI (OR 1.6; 95% CI = 1.1–2.3; $P = 0.02$) [21]. In a population-based retrospective cohort study, the incidence rate of PROM was correlated inversely with body mass index (BMI) and waist circumference. We considered that their low BMI (< 17.5 and $17.5–19.9$ kg/m²) and low WC (< 60 and $60.0–64.0$ cm) groups were different from the present study [22].

Conclusion

In conclusion, the increase of AFP level, NT and maternal weight are risk factors of PPRM, which may be helpful for the prediction of PPRM in the pregnant women. However, PAPP-A and free β -hCG in first and second trimester had no association with PPRM.

Abbreviations

PAPP-A: pregnancy-associated plasma protein A; free- β hCG: free beta human chorionic gonadotropin; NT: nuchal translucency; PPRM: predict preterm prelabor rupture of membranes; MoM: multiple of the median; ROC: receiver operating characteristic; AUC: area under curve; OR: Odds ratio.

Declarations

Conflict Of Interest

The authors have no conflicts of interest to declare.

Statement Of Ethics

The study was approved by Hangzhou Women's Hospital (Hangzhou Maternity and Child Health Care Hospital) ethics committee, in accordance with the ethical standards as laid down in the 1964 Declaration of Helsinki and its later amendments or comparable ethical standards. And the approval number was [2018] medical ethics (004) No.01. Consent you obtained from study participants was written.

Data Availability Statement

Data sharing is not applicable to this article as no new data were created or analyzed in this study.

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Authors' Contributions

Y. M. Chen, Y. J. Chen, and Z. F. Zhang conceptualized and refined the study design and methodology. W. Ning conducted data collection and tested blood samples and screening indicators. Y. M. Chen and X.Y. Wang conducted all analyses and prepared tables and figures with assistance from Data Analysis Department. L. Chen assessed the ultrasound of fetal nuchal translucency. Y. J. Chen wrote the entire manuscript and performed literature review. All authors read and approved the final manuscript.

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Figures

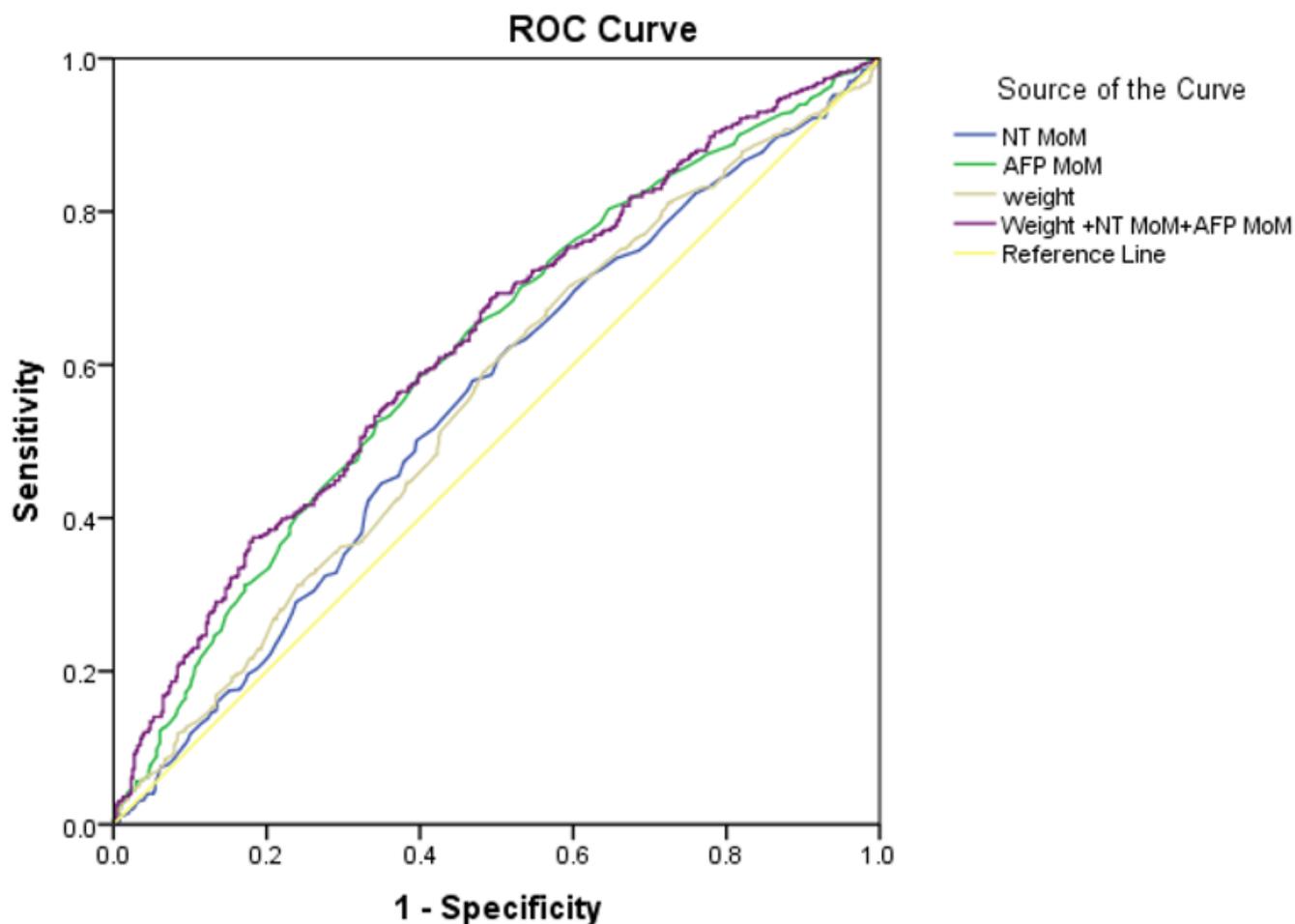


Figure 1

ROC of AFP, NT and maternal age correspond to MoM in screening PPRM AFP, α-fetoprotein; NT, nuchal translucency; Weight, Maternal weight in the first trimester.