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Do Triple Test Results Predict the Risks for Adverse Pregnancy Outcomes?

Hale Goksever Celik[1], Engin Celik[1], Gokhan Yildirim[1]

ABSTRACT:

Do triple test results predict the risks for adverse pregnancy outcomes?

Objective: Of the second trimester maternal screening tests, the triple screening test, is used to determine the risk of certain fetal chromosomal abnormalities, particularly Down's Syndrome. Besides the risk assessment for chromosomal abnormalities, many adverse pregnancy outcomes (APO) can be predicted with this test. In our study, we aimed to determine the relationship between the triple screening test results with APO and to define the cut-off values in serum markers for these APO.

Material and Methods: A total of 1372 pregnant women who had a triple screening test between April 2014 and 2015 and then gave birth in our hospital during the period of one year were included in the study. Complete demographic and clinical characteristics were obtained from records including age, weight, obstetric history, presence of any comorbid conditions, in vitro fertilization pregnancies, smoking, gestational weeks on triple screening test and at delivery, birth weight, type of delivery, gender of baby.

Results: The mean age of our study population was 27.9±5.6, whereas the median age was 28 years. The mean gestational week was 17.3±1.0 during the triple screening test, and 38.3±2.7 at the time of delivery. Most patients were multiparous and had vaginal delivery. The APO were encountered mostly in the older age multiparous patients and in cesarean group. The cut-off values for APO were determined to be > 0.935 MoM for AFP, < 0.945 MoM for E3 and > 0.945 MoM for hCG.

Conclusions: The second trimester screening test is applied to all patients in our clinic, due to its low cost and high effectiveness in predicting the chromosomal abnormalities. In fact, determination of risk for the APO with the first trimester screening test is more meaningful, because of the feasibility of prevention strategies such as bed rest and use of aspirin. Nevertheless, it is important to inform the patients about the benefits of the second trimester screening test to be predicting APO and recommend it.

Keywords: Gestational diabetes mellitus, intrauterine growth restriction, preeclampsia, triple test

ÖZET:

Üçlü tarama testi sonuçları kötü gebelik sonuçlarını öngörür mü?

Amaç: İkinci trimestır maternal tarama testlerinden üçlü tarama testi özellikle Down Sendromu olmak üzere bazı fetal kromozomal anormalliklerinin riskini tayin etmek için kullanılır. Bunun yanında pek çok kötü gebelik sonuçları da bu testle öngörülebilmektedir. Çalışmamızda kötü gebelik sonuçları ile üçlü tarama testi sonuçları arasındaki ilişkiyi tespit etmek ve bu kötü obstetrik sonuçlar için serum belirteçlerinde kesim noktası değerleri tanımlamayı amaçladık.

Gereç ve Yöntemler: Nisan 2014 ve 2015 tarihleri arasında bir yıllık sürede üçlü tarama testini hastanemizde yaptırmış ve doğum yapmış 1372 gebe kadın çalışmaya dahil edildi. Yaş, kilo, obstetrik öykü, komorbid durumların varlığı, yardımcı üreme tekniği kullanımı, sigara içme, üçlü tarama testi ve doğumdaki gestasyonel hafta, doğum şeklini içeren hastaların tüm demografik ve klinik özellikleri kayıtlardan elde edildi.

Bulgular: Populasyonun ortalama yaşı 27.9±5.6 olup ortanca yaş 28 idi. Üçlü tarama testinin yapıldığı gebelik haftası ortalama 17.3±1.0 iken doğumun gerçekleştiği hafta ortalama 38.3±2.7 idi. Çoğu hasta multipar olup vajinal yolla doğum gerçekleşmişti. Kötü gebelik sonuçlarına çoğunlukla daha ileri yaş multipar hastalarda ve sezaryen olmuş grupta rastlanıldı. Kötü gebelik sonuçlarına yönelik kesim noktaları AFP için > 0.935 MoM, E_3 için < 0.945 MoM, hCG için > 0.945 MoM olarak tespit edildi.

Sonuçlar: Düşük maliyeti ve kromozomal anormallikleri öngörmede yüksek etkinliğe sahip olduğu için ikinci trimestır tarama testi kliniğimizde tüm hastalara uygulanmaktadır. Aslında birinci trimestır tarama testi ile kötü gebelik sonuçlarının öngörülmesi yatak istirahati, aspirin kullanımı gibi önleyici yaklaşımların uygulanabilirliği nedeniyle daha anlamlıdır. Yine de hastaları ikinci trimestır tarama testinin kötü gebelik sonuçlarını öngörmedeki faydası konusunda bilgilendirmek ve yapılmasını önermek önemlidir.

Anahtar kelimeler: Gestasyonel diyabetes mellitus, intaruterin gelişim kısıtlılığı, preeklampsi, üçlü tarama testi

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[1]Kanuni Sultan Suleyman Training and Research Hospital, Department of Gynecology and Obstetrics, Istanbul - Turkey

Address reprint requests to / Yazışma Adresi:
Hale Göksever Çelik,
Kanuni Sultan Suleyman Training and Research Hospital, Department of Gynecology and Obstetrics, Istanbul - Turkey

E-posta / E-mail:
hgoksever@yahoo.com

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INTRODUCTION

Antenatal screening tests have being used widely all over the world (1). The triple test is performed between 14 and 21 weeks of gestational age and determine the risk for fetal chromosomal abnormalities, combining maternal serum levels of α-fetoprotein (AFP), total β-human chorionic gonadotropin (hCG) and unconjugated estriol (E_3) levels, and maternal age although it is not diagnostic (2). Alvarez-Nava et al. (3) found that elevated second trimester maternal serum markers were also indicators for Turner syndrome. There have been many developments during recent years to increase the detection rate of chromosomal abnormalities. According to recent guidelines, quad marker test which includes inhibin A in addition to other markers of triple test serum markers should be recommended as a second trimester screening test. But the evidence for quad market test being superior to triple test is limited (4).

Another benefit of the triple test is that many adverse pregnancy outcomes (APO) such as preeclampsia, gestational diabetes mellitus (GDM), intrauterine growth restriction (IUGR) may be understood with these serum markers (5). We currently know that in case of any suspicion of APO in an early gestational week would let us recommend several precautions such as close follow-up, bed rest or aspirin (6).

We aimed to determine the relationship between triple test results with APO and to define the cut-off values for these APO. We compared parameters of triple test serum markers and found that AFP/E_3 ratio is meaningful to determine the risk for APO.

MATERIAL AND METHODS

This is a case-control study. The triple antenatal screening test has been carried out in our hospital for many years. The results of triple test and pregnancy outcomes in 1372 pregnant women were investigated. These women were chosen from women who had triple test performed between April 2014 and April 2015, and gave birth in Kanuni Sultan Suleyman Training and Research Hospital. Inclusion criteria were viable fetus between 14 and 21 weeks of gestation, singleton pregnancy, availability of the results of triple test. We excluded multiple pregnancies, women having a fetus with known neural tube defect or any chromosomal abnormality and women with a history of APO. We did not include women who had done triple test and then not come for follow-up.

All maternal serum markers were studied with solid-phase competitive enzyme immunoassay method. All of them were converted and analyzed as multiples of the median (MoM). Fasting blood tests were taken from an antecubital vein from pregnant women between 14 and 21 weeks of gestation. Blood samples were allowed to clot for 20-30 minutes at room temperature and then centrifuged at 3000 x g for 5 minutes and frozen up to below -80°C until analysis.

Complete demographic and clinical characteristics were obtained from records, including age, weight, obstetric history, presence of any comorbid conditions, in vitro fertilization pregnancy, smoking, gestational weeks on triple test and birth, birth weight, type of delivery, gender of baby. We also investigated the association of these characteristics with APO.

Our study was designed retrospectively and conducted according to the Helsinki Declaration. There was no ethical approval because we collected data of the patients from the records in archive and we did not documented any personal information. Also in our hospital, informed consent is taken from every patient regarding the medical information, which may be used in scientific publications.

Statistical Analysis

Statistical analyses were performed with the Statistical Package for the Social Sciences (SPSS Inc; Chicago, IL, USA) statistics 22.0 version for Windows. Differences in mean values and characteristics between groups are analyzed with independent samples t-test and chi-square test. Means were presented with standard deviation (SD). $p<0.05$ was considered statistically significant. For the multivariate analysis, the possible factors identified with univariate analyses were further entered into

Table-1: Demographic and clinical characteristics of patients

Characteristics	Mean±SD	Min - Max values
Age	27.9±5.6	15 - 51
Weight (kg)	65.7±12.8	36 - 132
Gravidity	2.6±1.4	1 - 12
Parity	1.2±1.0	0 - 7
Gestational week on triple test (weeks±days)	17.3±1.0	15.0 - 20.6
E_3 (MoM)	1.00±0.40	0.14 - 5.08
hCG (MoM)	1.17±0.72	0.01 - 15.39
AFP (MoM)	0.99±0.55	0.13 - 11.84
Gestational week on birth (weeks›days)	38.3±2.7	21.0 - 42.6
Birth weight (gram)	3168.9±629.9	330 - 4700

E_3: estriol, hCG: human chorionic gonadotropin, AFP: alpha-fetoprotein

the logistic regression analysis to determine independent predictors of patient outcome. Thresholds for the association of abnormal maternal serum triple analytes with APO for this study were determined by initially using receiver operating characteristics (ROC) curves to ascertain the optimal cut-off for each analyte. When a significant cut-off value was observed, the sensitivity, specificity, positive likelihood ratio values were presented. While evaluating the area under the curve, a 5% type-I error level was used to accept a statistically significant predictive value of the test variables.

RESULTS

A total of 1372 women who had triple test performed between April 2014 and April 2015 and gave birth at Kanuni Sultan Suleyman Training and Research Hospital were included in the study. The mean age of our study population was 27.9±5.6, whereas the median age was 28. According to median age, the majority of women in the group aged 28-51. The mean gestational week was 17.3±1.0 weeks during triple test, and 38.3±2.7 weeks during birth (Table-1). Most patients were multiparous and had vaginal delivery. 42.6% of patients gave birth by cesarean section and the most frequent indication for cesarean section was previous cesarean history (Table-2).

Adverse pregnancy outcomes such as preeclampsia, IUGR were encountered in 113 patients (8.2% of patients). Older age women giving birth with cesarean section were more prone to

Table-2: Distribution of demographic and clinical characteristics

Characteristics	Number (%)
Age	
≤28	676 (49.3)
>28	696 (50.7)
Parity	
0	346 (25.2)
≥1	1026 (74.8)
Smoking	
Absent	1223 (89.1)
Present	149 (10.9)
Chronic health problems (DM, etc)	
Absent	1351 (98.5)
Present	21 (1.5)
In vitro fertilization pregnancy	
Absent	1360 (99.1)
Present	12 (0.9)
Poor obstetric history (abortus, preterm birth, still birth, etc)	
Absent	1363 (99.3)
Present	9 (0.7)
Route of labor	
Vaginal birth	788 (57.4)
Cesarean section	584 (42.6)
Adverse pregnancy outcome (preeclampsia, IUGR, GDM,etc)	
Absent	1259 (91.8)
Present	113 (8.2)
Indications for cesarean section	
Previous cesarean	329 (24)
Fetal distress	99 (7.2)
Obstructed labor	42 (3.1)
Malpresentation	39 (2.8)
Macrosomia	28 (2.0)
Cephalopelvic disproportion	22 (1.6)
Ablatio placenta	11 (0.8)
Placenta previa	10 (0.7)
History of myomectomy	4 (0.3)

DM: diabetes mellitus, IUGR: intrauterine growth restriction, GDM: gestational diabetes mellitus

Table-3: Distribution of characteristics according to presence of adverse pregnancy outcome

Characteristics	Adverse pregnancy outcome		
	Absent	Present	p
Age			
≤28	635 (93.9)	41 (6.1)	0.004
>28	624 (89.7)	72 (10.3)	
Parite			
0	315 (91)	31 (9)	NS
≥1	944 (92)	82 (8)	
Smoking			
Absent	1123 (91.8)	100 (8.2)	NS
Present	136 (91.3)	13 (8.7)	
Chronic health problems (DM, etc)			
Absent	1247 (92.3)	104 (7.7)	<0.001
Present	12 (57.1)	9 (42.9)	
In vitro fertilization pregnancy			
Absent	1247 (91.7)	113 (8.3)	NS
Present	12 (100)	0	
Poor obstetric history			
Absent	1252 (91.9)	111 (8.1)	NS
Present	7 (77.8)	2 (22.2)	
Route of labor			
Vaginal birth	775 (98.4)	13 (1.6)	<0.001
Cesarean section	484 (82.9)	100 (17.1)	

DM: diabetes mellitus

Table-4: Relationship between adverse pregnancy outcomes and MoM values

	Cut-off value	Area under curve	Sensitivity (%)	Specifity (%)	Positive likelihood ratio
E_3	0.945	0.53	55.8	48.5	1.08
hCG	0.945	0.52	59.3	42.9	1.04
AFP	0.935	0.58	56.6	56.1	1.29
AFP/E_3	0.94	0.58	61.1	49.9	1.22
AFP/hCG	0.84	0.54	60.2	45	1.09
hCG/E_3	0.88	0.54	69	36.9	1.09

E3: estriol, hCG: human chorionic gonadotropin, AFP: alpha-fetoprotein

Table-5: Results of logistic regression analysis

	RR (95% CI)	p
AFP	1.57 (1.17-2.09)	0.002
hCG	1.09 (0.87-1.38)	NS
E_3	0.74 (0.44-1.23)	NS
AFP/E_3	1.38 (1.17-1.64)	<0.001
AFP/hCG	0.99 (0.90-1.08)	NS
hCG/E_3	1.21 (1.05-1.40)	0.009

E3: estriol, hCG: human chorionic gonadotropin, AFP: alpha-fetoprotein, RR: risk ratio

experience APO. Although statistically not significant, APO was more frequently observed in multiparous patients (Table-3).

In our study, APO were determined with cut-off values for AFP>0.935 MoM, E_3<0.945 MoM, and hCG>0.945 MoM. If these values were compared with each other, the most helpful one was AFP/E_3 with cut-off values of 0.94 (Table-4).

Table-5 shows the results of the binary logistic regression analysis. The AFP level was identified as the strongest factor determining the risk for APO in our study population.

DISCUSSION

Second trimester maternal serum triple screening test, is being used for the detection of risk of fetal chromosomal abnormalities, especially Down syndrome, which affects approximately 1 in 800 live-born babies. This test is applied between

gestational weeks of 14 and 21. Serum AFP, hCG and E_3 values are combined with maternal age to estimate the risk (7).

Besides the risk assessment for chromosomal abnormalities, many APO such as preeclampsia, GDM, IUGR can be detected with this test.

Our primary outcome defined as APO were gestational hypertension, preeclampsia, HELLP syndrome, IUGR and GDM. Preeclampsia is defined as the blood pressure ≥140/90 mmHg during pregnancy or within the first 24 hours postpartum without a history of chronic hypertension, and is diagnosed if proteinuria exists in addition to diagnostic criteria of gestational hypertension without the signs and symptoms of preeclampsia. There is hemolysis, thrombocytopenia, elevated liver function tests in HELLP syndrome which is accepted as a variant of preeclampsia. IUGR is suspected when the estimated fetal weight falls below the 10th percentile for gestational age. Testing for GDM screening is typically performed at 24 to 28 weeks of gestation. If the screening test is positive, then a diagnostic 3-hour glucose tolerance test is performed with 100 g glucose after at least 8 hours of fasting. With abnormal fasting or any other two abnormal values, the diagnosis of GDM is confirmed (8).

We apply triple test to all patients consulted to our clinics for control during second trimester, due to its cost-effectiveness and its high rates of detection of chromosomal abnormalities (9). Triple test is feasible in multiple pregnancies, but we excluded the multiparous patients from our study (10). We can also identify women with high risks for APO with the second trimester serum markers. In fact, determination of risks for said APO with the first trimester screening test is more sensible, because prevention strategies such as aspirin, bed rest are more beneficial for these pregnant women (11). But it is not late to offer screening tests during second trimester to the patients and inform about benefits for risk assessment of APO. Yang et al. (12) evidenced that second trimester screening tests of serum markers can be helpful to identify fetal chromosomal and anatomical anomalies and to predict unfavorable pregnancy outcomes, which are similar to our results. In another study, abnormal results of second trimester screening test could be associated with APO in women with normal appearing fetus (6). Gu et al. (13) concluded that combination of parameters (pregnancy-associated plasma protein-A (PAPP-A), E_3 and AFP) are more helpful for prediction of preeclampsia than individual parameters, whereas Hume et al. (11) preferred the serum markers in early pregnancy in prediction of preeclampsia.

Decisions may vary by personal preference about having knowledge and taking precautions for APO. Our responsibility should be to inform patients about benefits of screening tests. Our aim was to emphasize the importance of these tests. In time, developments will allow us to understand which test is superior. The application of screening tests at earlier gestational weeks must be the subject for further studies.

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