Analysis of Triple Test Results in 27 Cases of Twin Pregnancies

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Abstract. The study comprises 889 pregnant women between 14 and 21 weeks of gestation. The control group consisted of 862 pregnant women with unburdened obstetric anamnesis with an uneventful singleton pregnancy. The examined group consisted of 27 pregnant women with uncomplicated twin pregnancy. In the sera of pregnant women AFP (Microparticle Enzyme Immunoassay AxSYM Abbott), total β-hCG (Microparticle Enzyme Immunoassay AxSYM Abbott) and unconjugated estriol (Radioimmunoassay Amerlex-M. 2T Johnson & Johnson Ortho Clinical Diagnostics Ltd.) were determined. The risk of fetal trisomy 21 was calculated with the use of PRISCA 3.0 software, which corrected the MoM values for twin pregnancy. Ulm Index was also calculated. In the majority of twin pregnancies increased concentrations of AFP, total β-hCG and uE₃ in the range over 1,0 MoM was noted. In the group of women below 35 years of age with singleton pregnancies using PRISCA 3.0 software it approximated to 95%. For women older than 35 optimum index for fetal trisomy 21 risk calculation was Ulm Index with the specificity 93,8%. The specificity of AFP determination in the detection of fetal open NTD in singleton pregnancy was 99%. In the group of women with twin pregnancy the obtained specificity of 77,8% for PRISCA 3.0 software is low, a more advantageous way to calculate the risk of fetal trisomy 21 is Ulm Index with the specificity of 85,2%. The specificity of AFP determination as a screening for fetal open NTD in twin pregnancy was 96,3%.

Key words: Triple test, α-fetoprotein, β-hCG, Unconjugated estriol, Twin pregnancy, Fetal trisomy 21, Open neural tube defect

INTRODUCTION

The number of multiple pregnancies grows together with the broader use of assisted reproduction techniques. The women in whom the pregnancies have been achieved in
this way are most often older than 30 years of age. That is why prenatal diagnosis aimed mainly at the detection of chromosomal aneuploidies is indicated. The would-be-mothers prefer non-invasive diagnostic modalities, that is “genetic ultrasound” and the triple test especially when the pregnancy was preceded by long-term therapy of infertility. The triple test is a screening examination performed in pregnant women between 14 and 21 weeks of gestation aimed the detection of fetal trisomy 21 and open NTD. It is based on the measurement of the concentrations of AFP, hCG and uE3 in maternal serum. Gestational age is determined according to fetal ultrasound biometry [9, 10]. The result of the test is expressed of a likelihood ratio of fetal trisomy 21 and open NTD. In cases of pregnancies with fetal trisomy 21 the concentrations of AFP and uE3 decrease below 0.7 MoM, whereas the concentrations of hCG and free β-hCG increase over 2.5 MoM [3, 4, 5, 8]. In cases of pregnancies with open NTD AFP concentrations increase over 2.5 MoM [6, 7]. In twin pregnancies the increase of all three examined parameters is observed [7]. This leads to the increase in the false positive results ratio of the triple test; both in the detection of fetal trisomy 21 (caused by the increase of hCG concentration) and fetal open NTD (caused by the increase of AFP concentration). Thus, adequate correction of MoM is warranted so as to adjust the risks of fetal trisomy 21 and open NTD.

MATERIALS AND METHODS

The study comprised 889 pregnant women between 14 and 21 weeks of gestation. The control group consisted of 862 pregnant women with unburdened obstetric anamnesis with an uneventful singleton pregnancy. The examined group consisted of 27 pregnant women with uncomplicated twin pregnancy. In each patient a detailed medical and obstetric anamnesis were obtained. Every patient was weighted. The pregnancy dating was based on biparietal diameter (BPD) and femur length (FL) Sonoline SI-450 Siemens ultrasound scanner, 3.5 MHz convex probe. Measured on the day of the collection of the sample or a day before. In all patients 8 ml of peripheral venous blood was obtained. The sera were stored in -65°C until the determination. After bringing the sample to room temperature the concentrations of AFP, total β-hCG (Microparticle Enzyme Immunoassay AxSYM Abbott) and unconjugated estriol (Radioimmunoassay Amerlex-M, 2T Johnson & Johnson Ortho Clinical Diagnostics Ltd.) were determined. The risk of fetal trisomy 21 was calculated with use of PRISCA 3.0 software, which corrected the MoM values for twin pregnancy. Ulm Index was also calculated according to the following equation [2]:

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Ulm \text{ Index} = \frac{(hCG \text{ MoM})^2}{(AFP \text{ MoM})^2 \cdot uE3 \text{ MoM}}
\]

The cut-off level for singleton pregnancy for the detection of open NTD was 2.5 AFP MoM, whereas for twin pregnancy it was 3.5 AFP MoM [6].
RESULTS

The distributions of AFP, total β-hCG and uE₃ concentrations in the control group showed typical values for the examined period of gestation. AFP and uE₃ concentrations increased gradually with gestational age, whereas total β-hCG concentrations decreased. The above determinations were used as a basis for the calculation of the medians for the consecutive weeks of gestation. In the group of women with twin pregnancy, an increase of all examined parameters in comparison to singleton pregnancy was observed. In the majority of twin pregnancies, concentrations of AFP, total β-hCG and uE₃ in the range over 1.0 MoM showed an increase. The median of AFP concentration for twin pregnancy equalled 2.22 MoM comparing to singleton pregnancy (Fig. 1). The median of total β-hCG for twin pregnancy equalled 2.44 MoM (Fig. 2) and the median of uE₃ equalled 1.78 MoM comparing to singleton pregnancy (Fig. 3).

The specificity of the triple test was shown to decrease together with maternal age. In the group of women below 35 years of age with singleton pregnancies using PRISCA 3.0 software it approximated to 95%. For women older than 35 optimum index for fetal trisomy 21 risk calculation was Ulm Index with the specificity 93.8%. The specificity of AFP determination in the detection of fetal open NTD in singleton pregnancy was 99%. In the group of women with twin pregnancy there were 6 cases of false positive results in the screening for fetal trisomy 21 using PRISCA 3.0 software (1:250 cut-off level) and 4 cases using Ulm Index (8.1 cut-off level).

There was also 1 false positive result in the screening for fetal open NTD (2.5 AFP MoM cut-off level). In the present study the specificity of the triple test in twin pregnancy as a screening for fetal trisomy 21 (1:250 cut-off level) was 77.8% for PRISCA 3.0 software and 85.2% for Ulm Index. The specificity of AFP determination as a screening for fetal open NTD in twin pregnancy was 96.3%.

Fig. 1 - The values of AFP MoM in cases of twin pregnancies.
DISCUSSION

Because of the above described changes of the AFP total β-hCG and uE₃ concentrations, in twin pregnancy different attitudes exist towards the triple test in such cases. This is caused most often by the decrease of specificity and sensitivity of this test comparing to singleton pregnancy. Such cases require lowering of the cut-off level even to 1:50 [1]. The values of AFP MoM (2, 22) and β-hCG MoM (2, 44) obtained in the present study
are higher than the corresponding results obtained by Barnabei et al. which equalled 1, 91 AFP MoM and 1, 99 hCG MoM [1].

In the examined group of women with twin pregnancy neither cases of fetal trisomy 21 nor fetal open NTD was identified, which allowed us to calculate only the specificity. The obtained specificity of 77.8% for PRISCA 3.0 software is low; a more advantageous way to calculate the risk of fetal trisomy 21 is Ulm index with the specificity of 85.2%. While analysing the results of the triple test in a case of twin pregnancy it is very important to know whether it is a dizygotic or monozygotic one. In case of dizygotic twins when trisomy 21 is present in one of the twins low concentrations of AFP and uE$_3$ and high concentrations of hCG in maternal serum may be obscured by the healthy twin. The similar situation may occur in presence of open NTD in one twin: high AFP concentrations coming from the affected twin, may be masked by the normal concentration coming from the healthy one. Wald et al. described 73% sensitivity in the triple test for monozygotic twin pregnancy and 43% for dizygotic one at 5% false positive results ratio [11]. Barnabei et al. reported the mean sensitivity (not differentiating twins into mono-and dizygotic) of 80.3% for the Caucasians and 64.9% for the Blacks [1].

CONCLUSIONS

1. The value of AFP, total β-hCG and uE$_3$ in twin pregnancies (AFP MoM – 2, 22, β-hCG MoM – 2,44, uE$_3$ MoM – 1,78) obtained in the present study are higher in comparison to singleton pregnancies.

2. The obtained specificity of 77.8% for PRISCA 3.0 software is low, a more advantageous way to calculate the risk of fetal trisomy 21 in twin pregnancies is Ulm Index with the specificity of 85.2%.

REFERENCES


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