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PRENATAL SCREENING MARKERS FOR DOWN SYNDROME: SENSITIVITY, SPECIFICITY, POSITIVE AND NEGATIVE EXPECTED VALUE METHOD

SENZITIVNOST, SPECIFIČNOST, POZITIVNA I NEGATIVNA PREDVIĐENA VREDNOST MARKERA PRENATALNOG SKRININGA NA DAUNOV SINDROM

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Summary

Background: Genetic screening for chromosomopathy is performed in the first trimester of pregnancy by determining fetal nuchal translucency (NT), and the pregnancy associated plasma protein-A (PAPP-A) and free human chorionic gonadotropin (free-beta HCG) biomarkers in maternal serum.

Methods: We tested the sensitivity, specificity, positive and negative expected values of each marker with the aim of setting a model for prenatal screening readings. Statistical data treatment has been performed on a sample of 340 pregnant women with positive results of prenatal screening.

Results: Sensitivity of PAPP-A was 0.6250 (probability 62.50%), free beta HCG 0.5893 (58.93%), NT 0.1785 (17.85%). Specificity of PAPP-A was 0.5106 (probability 51.06%), free beta HCG 0.5246 (52.46%), NT 0.9718 (97.18%). Positive expected value of PAPP-A was 0.2011 (probability 20.11%), free beta HCG 0.1964 (19.64%), NT 0.556 (55.56%). Negative expected value of PAPP-A was 0.8735 (probability 87.35%), free beta HCG 0.8662 (86.62%), NT 0.8571 (85.71%). The NT marker has a significantly higher specificity, which means that its normal value will significantly reduce the final risk of trisomy 21. The sensitivity of NT is much lower than that of biochemical markers, which means that a pathological value of NT does not have a significant influence on the final risk, i.e. the significantly higher sensitivity of biochemical markers will reduce the final risk of trisomy 21.

Kratak sadržaj

Uvod: Prenatalni skrining na Daunov sindrom u prvom trimestru trudnoće radi se ultrazvučnim merenjem nuhalne translucencije fetusa (NT) i određivanjem fetoplacentalnih biomarkera u maternalnom serumu: pregnancy associated plasma protein-A (PAPP-A) i free human chorionic gonadotropin (free beta HCG).

Metode: Ispitana je senzitivnost, specifičnost, pozitivna i negativna predviđena vrednost svakog markera u cilju postavljanja modela tumačenja prenatalnog skrininga i interpretacija patoloških vrednosti. Ispitivanje je rađeno na uzorku od 340 trudnica sa pozitivnim nalazom prenatalnog skrininga gde je amniocentezom dobijen kariotip ploda.

Rezultati: Senzitivnost PAPP-A bila je 0,6250 (verovatnoća 62,50%), free beta HCG 0,5893 (58,93%), NT 0,1785 (17,85%). Specifičnost PAPP-A bila je 0,5106 (verovatnoća 51,06%), free beta HCG 0,5246 (52,46%), NT 0,9718 (97,18%). Pozitivna predviđena vrednost PAPP-A bila je 0,2011 (verovatnoća 20,11%), free beta HCG 0,1964 (19,64%), NT 0,556 (55,56%). Negativna predviđena vrednost PAPP-A bila je 0,8735 (verovatnoća 87,35%), free beta HCG 0,8662 (86,62%), NT 0,8571 (85,71%). Uticaj PAPP-A i free beta HCG na konačan rizik za trizomiju 21 je približno jednak. NT marker ima značajno veću specifičnost što znači da će njegova normalna vrednost značajno oboriti konačan rizik za trizomiju 21. Senzitivnost NT je mnogo manja od biohemijskih markera, što znači da patološka vrednost NT ne

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Conclusions: The analyses stress the importance of using a software which has the possibility to separate the level of a biochemical risk by correlating PAPP-A and free beta HCG and, by adding the NT marker, calculate the level of a final risk of Down syndrome.

Keywords: prenatal screening, Down syndrome, sensitivity, specificity

utiče značajno na konačan rizik, odnosno značajno veća senzitivnost biohemijskih markera će oboriti konačan rizik za trizomiju 21.

Zaključak: Ove analize ukazuju da je veoma važno koristiti softver za prenatalni skrining koji ima mogućnost da razdvoji posebno nivo biohemijskog rizika korelacijom PAPP-A i free beta HCG i dodavanjem NT markera izračuna nivo konačnog rizika za Daunov sindrom.

Ključne reči: prenatalni skrining, Daunov sindrom, senzitivnost, specifičnost

Introduction

Prenatal screening for Down's syndrome is done in the first trimester of pregnancy between 11 and 14 weeks by the ultrasound measurement of nuchal translucency (NT-neck crease) and the determination of fetal maternal serum biomarkers: pregnancy-associated plasma protein-A (PAPP-A) and free beta human chorionic gonadotropin (free beta-hCG). The concentration of biochemical markers in maternal serum is converted to a multiple of the median (MoM) of unaffected pregnancies at the same gestation stage (1–4). The measured serum concentrations of these placental products are affected by maternal characteristics, including maternal age, racial origin, weight, diabetic status, smoking and method of conception. The risk of Down's syndrome is determined i.e. calculated by a combination of software processing of the maternal characteristics, biochemical and sonographic markers. As a cut-off risk indicating prenatal karyotyping, 1:270 is used which corresponds to a pregnant woman aged 35 (5).

There is no significant association between fetal NT and maternal serum free beta hCG and PAPP-A in either trisomy 21 or euploid pregnancies (6). It has been estimated that the false-positive rate in genetic screening is about 5%, which has resulted in an increased number of invasive diagnostic procedures of prenatal karyotyping in risk free pregnant women with respect to age (7). On the other hand, it is important to increase the sensitivity of prenatal screening to identify pregnancies suspicious for trisomy 21 at the age of non-risk in pregnant women as younger age may reduce the final risk (18). The performance of different screening methods for trisomy 21 with a combination of maternal age, sonographic and biochemical markers has been tested. It was found that effective screening in the first trimester of pregnancy should have a detection rate of about 95% and a false-positive rate of less than 3% (19). The aim of this study, however, is to define the interpretation of resulting risks and establish a model for the interpretation of pathological values of prenatal screening markers for trisomy 21. We tested the sensitivity, specificity, positive and negative expected values of each marker with the goal of setting a model for prenatal screening readings and interpretation of pathological values.

Methods

PAPP-A and free-beta HCG biomarkers have been read with IMMULITE 2000 SIEMENS which operates on the principle of chemiluminescence, using the original reagents. The processing of data and determination of the risk of trisomy 21 have been done with PRISCA 5 SOFTWARE. Statistical data treatment has been performed on a sample of 340 pregnant women with respect to age, all with positive results of prenatal screening, and the karyotyping of a fetus has been obtained with amniocentesis. Using a sensitivity analysis method, it has been determined with high probability that a pathological value of the marker implies the presence of risk. Using a specificity analysis method, it has been determined with high probability that a normal value of the marker implies the absence of significant risk. Using a positive expected value method, it has been determined with high probability that risk is present only if the marker implies so. Using a negative expected value method, it has been determined with high probability that significant risk is absent if the marker implies so.

Results

The study included a sample of 340 pregnant women with suspicious findings of genetic screening and finite risk of Down's syndrome in the PRISCA software greater than 1: 250, which is indicated based on prenatal karyotyping. In a sample of 340 high-risk findings of the screening, there were 18 (6.1%) results with the pathological values of NT markers. Pathological PAPP-A values were found in 174 (59.1%) cases. Free beta-hCG showed extreme values in 168 (57.1%) pregnant women. Values of the markers have been reported in deviation from the median – MoM (multiple of median). The risk of Down's syndrome is shown in PRISCA software at two levels, the risk of biochemical correlations of biochemical markers PAPP-A and free beta HCG and finite risk adding the ultrasound marker NT. The results show the effect of each marker in the formation of risk of Down's syndrome, influence of biochemical markers on biochemical and finite risk and impact of NT marker on the final risk.

Table I Influence of PAPP-A marker on biochemical risk.

	PAPP-A MOM normal value	PAPP-A MOM pathological value
Risk-free	98	86
Risk	68	88

Sensitivity 0.5563 (probability 55.63%)
Specificity of PAPP-A 0.4815 (probability 48.15%)
Positive expected value of PAPP-A 0.4615 (probability 46.15%)
Negative expected value of PAPP-A 0.5759 (probability 57.59%)

Table II Influence of free beta HCG marker on biochemical risk.

	Normal value free beta HCG	Free beta HCG pathological value
Risk-free	109	77
Risk	63	91

Sensitivity of free beta HCG 0.5909 (probability 59.09%)
Specificity of free beta HCG 0.5860 (probability 58.60%)
Positive expected value of free beta HCG 0.5416 (probability 54.16%)
Negative expected value of free beta HCG 0.6337 (probability 63.37%)

Table III Influence of PAPP-A marker on the final risk (biochemical + NT).

	PAPP-A MOM normal value	PAPP-A MOM pathological value
Risk-free	145	139
Risk	21	35

Sensitivity of PAPP-A 0.6250 (probability 62.50%)
Specificity of PAPP-A 0.5106 (probability 51.06%)
Positive expected value of PAPP-A 0.2011 (probability 20.11%)
Negative expected value of PAPP-A 0.8735 (probability 87.35%)

Table IV Influence of free beta HCG marker on the final risk (biochemical + NT).

	Free beta HGC normal value	Free beta HCG pathological value
Risk-free	149	135
Risk	23	33

Sensitivity of free beta HCG 0.5893 (probability 58.93%)
Specificity of free beta HCG 0.5246 (probability 52.46%)
Positive expected value of free beta HCG 0.1964 (probability 19.64%)
Negative expected value of free beta HCG 0.8662 (probability 86.62%)

Table V Influence of NT marker on the final risk (biochemical + NT).

	NT normal value	NT pathological value
Risk-free	276	8
Risk	46	10

Sensitivity of NT 0.1785 (probability 17.85%)
Specificity of NT 0.9718 (probability 97.18%)
Positive expected value of NT 0.5556 (probability 55.56%)
Negative expected value of NT 0.8571 (probability 85.71%)

Discussion

In the last two decades, there have been numerous reports about the detection rate for different methods of screening for trisomy 21. Detection rate of the risk of maternal age and fetal NT is 75–80%, while the risk for age and biochemical screening of PAPP-A and free beta HCG is 70%. The combination of age-related risk markers NT, PAPP-A and free beta HCG increases the detection of trisomy 21 to 85–95% (27, 28). The ability to achieve a reliable measurement of NT is dependent on the appropriate training of sonographers (29). Biochemical analyzers provide automated, precise and reproducible measurements. Presenting selectively the biochemical and ultrasound screening in the first trimester and representing a separate risk of sonographic and biochemical markers give a much better insight than the pure presentation of their combination. This type of screening is achieved by a policy in which the first-stage screening is based on maternal age, fetal NT and either tricuspid or ductus venosus flow, and biochemical testing is then performed only in those with an intermediate risk. An alternative first trimester contingent screening policy consists of maternal serum biochemistry in all pregnancies followed by fetal NT only in those with an intermediate risk after biochemical testing (30).

In our study, we examined the significance of the difference between positive and negative values of risk on the one hand and, on the other hand, the normal and pathological values of markers when they apply to the analysis of contingency. It is expected that the results indicate the distinction between individual markers and combinations of risks that have emerged in the sample. According to the results of chi-square testing, there is a certain tendency for the connection between markers and risks so that the null hypothesis can be rejected at the probability level of 90%. However, specificity indicates that there are exceptions. The correlation between markers and risks is best tested through the analysis of sensitivity, specificity and the positive and negative predictive value.

The influence of PAPP-A and free beta HCG on the final risk of trisomy 21 is approximately the same. NT marker has a significantly higher specificity, which means that its normal value will significantly reduce the final risk of trisomy 21. The sensitivity of NT is much lower than that of biochemical markers, which

means that a pathological value of NT does not have a significant influence on the final risk, i.e. the significantly higher sensitivity of biochemical markers will reduce the final risk of trisomy 21. The analyses stress the importance of using a prenatal screening software which has the possibility to separate the level of a biochemical risk by correlating PAPP-A and free beta HCG and, by adding the NT marker, the level of a final risk of Down's syndrome. At these two levels a very different risk is often obtained, and the analytical methods of this study suggest a new model of reading the obtained risks.

Effective screening for Down's syndrome can be achieved in the first trimester of pregnancy with a detection rate of about 95% and a false-positive rate of less than 3%.

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Conflict of interest statement

The authors stated that they have no conflicts of interest regarding the publication of this article.

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