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The Importance of Screening Tests and Amniocentesis in Approach to Pregnant Women Over the Age of Thirty-Five

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ABSTRACT

Objective: Our goal was to evaluate the approach to the increased risk of fetal chromosomal anomaly in pregnant women over thirty-five years of age.

Methods: We retrospectively examined pregnant women over the age of 35 who underwent interventional procedures for fetal karyotype analysis in the Perinatology Clinic of University of Health Sciences Turkey Kanuni Sultan Süleyman Training and Research Hospital. Regardless of the indication for karyotype analysis, pregnant women over the age of 35 who underwent karyotype analysis were included in the study.

Results: Abnormal karyotype was detected in 76 (8.7%) of a total of 867 pregnant women examined in the study. Of 76 abnormal karyotypes, 51 were found as trisomy 21 (67.7%), 15 as trisomy 18 (21%), and 3 as trisomy 13 (4%). Three fetal karyotype analyzes revealed Turner syndrome (4%), 1 Klinefelter syndrome (1.3%), 2 mosaic trisomy 21 (2.6%) and 1 mosaic Turner syndrome (1.3%). In the patients who underwent interventional procedures primarily because of advanced maternal age, abnormal karyotype was detected in 6 (1.9%) of them. The rates of abnormal karyotype results for double, triple and quadruple tests were 6.4%, 1.9% and 5.8%, respectively.

Conclusion: The use of a triple test has no definite impact on the results of pregnant women of older age. When there is no fetal abnormality in the ultrasound, double and quadruple tests may be requested or since the fetal death rate is minimal in amniocentesis, it can be recommended directly without performing any screening tests in pregnant women with older age.

Keywords: Advanced maternal age, amniocentesis, Down sendrome, chromosomal anomaly, karyotype analysis

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INTRODUCTION

The rate of pregnancies in older ages is increasing day by day. Advanced maternal age is a significant risk factor of chromosomal abnormalities (1,2). While trisomy 21 is the most prevalent chromosomal abnormality in live births, other genetic abnormalities such as trisomy 13, trisomy 18, Turner syndrome, and Klinefelter syndrome may also be found in newborns (3,4). To determine the likelihood of a fetal chromosome abnormality, double test, triple test, or quad test may be used. In these tests, in addition to prenatal ultrasonography, certain hormone levels in the maternal blood with maternal age are considered when determining the risk ratio. Advanced maternal age also causes high-risk screening test results by causing changes in biochemical parameters that increase the likelihood of chromosomal abnormalities (5). Tests and/or fetal ultrasonographic evaluation will identify pregnancies with a high probability of chromosomal anomalies (6). In high-risk pregnancies, a fetal chromosome analysis is recommended.

The interventional methods used to determine chromosomes differ depending on the week of pregnancy. Between 11-14 weeks of gestation, chorionic villus sampling (CVS); between 16-20 weeks, amniocentesis and >20 weeks, cordocentesis can be performed. Amniocentesis is the most common procedure (7). While interventional procedures are often used to diagnose chromosomal abnormalities, fetal DNA analysis [non-invasive prenatal test (NIPT)] in maternal blood has gained popularity in recent years. This non-invasive procedure is not widely performed in our country because it is a costly test with no government warranty. Since it cannot be used to make a conclusive diagnosis, it can be used as a powerful screening tool for the time being.

METHODS

Ethical approval for our study was obtained from İstanbul Esenyurt University, dated 17.02.2022 and numbered 2022/02-14. We retrospectively examined pregnant women over the age of 35 who underwent interventional procedures for fetal karyotype analysis in the Perinatology Clinic of University of Health Sciences Turkey Kanuni Sultan Süleyman Training and Research Hospital between 2014-2017. The study protocol was reviewed and approved by the local ethics committee. The patients' gestational weeks ranged from 12 to 22 weeks. The research involved pregnant women who underwent karyotype analysis only because they were over 35 years old, had a fetal anomaly on ultrasound examination, had an elevated risk (>1/250) in their double, triple, or quadruple test results or had a history of fetal chromosomal anomaly in their previous pregnancies. Patients who were pregnant to twins and who had a positive fetal DNA (cell-free DNA) analysis for chromosomal abnormalities were excluded from the study.

The research involved a total of 867 patients. Amniocentesis was performed in 685 patients, CVS in 148, and cordocentesis in 34. The interventional procedures were carried out by perinatology specialists.

Statistical Analysis

Frequency analysis was used to characterize nominal and ordinal parameters, frequency and percentage were used to describe categorical data.

RESULTS

In our research, 867 patients were included. There were 308 patients who underwent interventional procedures primarily because of advanced maternal age (Table 1). Abnormal karyotype was detected in 6 (1.9%) of them. In the group with advanced maternal age and serum biochemical screening tests; 1) Double test: Karyotype analysis was performed in 132 patients and 9 (6.4%) had abnormal karyotype. 2) Triple test: Abnormal karyotype was observed in 2 (1.9%) of 105 patients. 3) Quadruple test: Trisomy 21 was observed in 6 of 103 patients (5.8%).

Karyotype analysis was performed in 180 patients due to advanced maternal age and pathological ultrasonographic findings. Normal karyotype was found in 128 (71.2%) and abnormal karyotype was determined in 52 (28.8%) patients. In 24 patients with advanced maternal age and a birth history with chromosomal anomalies in their previous pregnancy, trisomy 21 was detected in only 1 (4.2%) patient. Abnormal karyotype was detected in 76 (8.7%) of a total of 867 pregnant women examined in the study and the distribution of chromosomal abnormalities was summarized in Table 2.

DISCUSSION

As the number of advanced maternal age pregnancies increases, screening and diagnosis of fetal chromosomal abnormalities have become more important. The prevalence of fetal chromosome abnormalities increases with the increase of maternal age (2).

Table 1. The distribution of karyotype analysis based on indications

Karyotype analysis indication	Normal karyotype	Abnormal karyotype	Total
Only advanced age	308 (98.1%)	6 (1.9%)	314
Advanced age + double test	132 (93.6%)	9 (6.4%)	141
Advanced age + tripple test	103 (98.1%)	2 (1.9%)	105
Advanced age + quadruple test	97 (94.2%)	6 (5.8%)	103
Advanced age + pathological ultrasound finding	128 (71.2%)	52 (28.8%)	180
Advanced age + history of chromosomal anomalies in previous pregnancy	23 (95.8%)	1 (4.2%)	24
Total	791 (91.3%)	76 (8.7%)	867

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Table 2. The result of karyotype analysis

Chromosomal anomaly	Number
Trisomy 21	51 (67.1%)
Trisomy 18	15 (19.7%)
Trisomy 13	3 (4%)
Turner syndrome	3 (4%)
Klinefelter syndrome	1 (1.3%)
Mosaic trisomy 21	2 (2.6%)
Mosaic turner send	1 (1.3%)
Total	76 (100%)

Chromosome abnormalities are detected by invasive methods after genetic counseling in pregnant women, which are required according to the results of double, triple, and quadruple tests, in which different biochemical analytes are used. The most common invasive test is amniocentesis (7).

The most important risk factor for chromosome number abnormalities is maternal age. The indications for interventional procedures that we conduct are similar to those reported in the literature (8). Apart from positive prenatal screening tests, the most important reason for amniocentesis is advanced maternal age (9,10). Advanced maternal age was shown to be the most common indication for amniocentesis in a sample of 6,041 pregnant women, and positive serum screening tests were found to be the second most common cause (11).

The incidence of chromosomal anomalies in amniocentesis, which is the most frequently performed interventional procedure, has been given between 1-6% in the ACOG guideline and in the literature (8,10,12). In our study group, this rate was found to be 8.7%. We believe that this elevated incidence was attributed to pregnant women referred to our clinic after a fetal anomaly was detected in an ultrasound scan. Excluding the pregnant women with fetal anomaly on ultrasound, abnormal karyotype was observed in 24 (3.5%) of the remaining 687 pregnant women, which was similar to the literature.

In our study, we applied interventional procedures to 308 pregnant women only because of advanced maternal age and we detected chromosome anomalies in 6 of them (1.9%). Xiao et al. (13) found the rate of detecting chromosomal abnormality as 2.79% in older pregnant women and 2.23% in those with biochemical marker abnormalities in maternal serum and showed that advanced maternal age was an independent indication for amniocentesis. In another study, Zhu et al. (14) found that the incidence of trisomy 21 increased significantly in women aged 39 and over. According to the literature, fetal abnormalities identified by prenatal ultrasonography have the highest positive predictive value for identifying chromosomal anomalies in amniocentesis (6). Danisman et al. (8) found the rate of chromosomal abnormality to be 10.5% in this group. In another study by Kagan et al. (5), chromosome anomalies were found in 15% of fetuses diagnosed as having congenital abnormality by prenatal ultrasonography.

Our chromosomal anomaly prevalence was 28.8% in pregnant women with advanced age in whom pathological ultrasonography findings were observed in ultrasonography. We think our rate is high because, as a third-step referral clinic, we observe a greater number of fetal abnormalities on ultrasound than the national average. Furthermore, the majority of patients referred due to early fetal ultrasound findings such as cystic hygroma, omphalocele, and holoprosencephaly, which are frequently associated with chromosomal defects, increases our chromosomal anomaly rate.

Chromosomal abnormality was detected as 4.2% in patients who underwent interventional procedures due to fetal anomaly in previous pregnancy. In the literature, this rate has been given as 3.2% (8). In our study, fetal chromosomal abnormality was detected in only 1 of 24 pregnant women who had a birth history with chromosome anomaly in their previous pregnancy. The results were similar but for this group the number of patients was low in our study.

Detection rates will increase with the addition of ultrasonography to maternal serum biochemical markers and the use of ultrasonographic markers such as nasal bone evaluation (15). As a result, we evaluated advanced maternal age in our sample using double, triple, and quadruple tests separately. The rate of fetal chromosome anomaly was 6.4% in the double test group and 5.8% in the quadruple test group. Fetal chromosome anomaly was detected in 2 of 105 patients with a rate of 1.9% who were evaluated as high risk by examining the triple test. This was similar to the rate of the patients without any screening test. Therefore, triple test appears to be a poor test in screening for chromosomal abnormalities in advanced maternal age. In our study, as in the literature, our rate of detecting chromosomal anomalies increased in the double test in which nasal bone and nuchal translucency were evaluated compared to the triple and quadruple tests.

Trisomy 21 is the most common chromosomal anomaly among live births (3,4,16). Ocak et al. (11) found the frequency of trisomy 21 as 46% among fetal chromosome abnormalities after amniocentesis. In an analysis performed on 13,795 pregnant women, trisomy 21 was the most common, with 35.6%, among chromosomal anomalies after fetal karyotype analysis (17). In our study, among the abnormal karyotypes, Trisomy 21 was found to be the most common with a rate of 67.1%.

In the case of pregnant women with advanced maternal age who underwent both maternal serum test and NIPT, the rate of fetal chromosomal abnormality was shown to be higher than those who received only maternal serum test screening or fetal DNA screening in maternal blood (18). We did not incorporate patients who were admitted to our clinic with a positive NIPT test. In our country NIPT is not widely used as it is not paid for by insurances due to its cost. In the future, fetal chromosome determination will be made easier and with less risk in case of widespread use.

In the study of Bornstein et al. (19), the benefit of genetic amniocentesis for the only advanced maternal age, far outweighed the risk of fetal death associated with amniocentesis. In another study, performing invasive intervention only because of

advanced maternal age was found to be more costly and the risk of fetal demise was higher, therefore, the authors recommended to use maternal serum analytes and ultrasound with advanced maternal age (20). In our study, in 180 patients with pathology on ultrasound, 5 fetal demises were observed after the procedure. Three of these 5 fetuses had large cystic hygroma and 2 had hydrops fetalis. However, no fetal loss due to invasive procedures was observed in 687 pregnant women whose fetuses appeared normal on ultrasound.

Study Limitations

Our clinic is a tertiary center where fetuses with pathological ultrasound findings are frequently referred, therefore the results may be different according to a population-based study.

CONCLUSION

In conclusion the risk of chromosomal abnormalities for fetuses with a high risk of Down syndrome in the triple test was found to be comparable to that of pregnant women who received regular amniocentesis due to advanced age without any test. Therefore, it does not seem reasonable to advise triple testing for pregnant women of old age. Instead, double, or quadruple test screening may be performed, or direct amniocentesis may be recommended as the risk of miscarriage is very low.

Ethics Committee Approval: Ethical approval for our study was obtained from İstanbul Esenyurt University, dated 17.02.2022 and numbered 2022/02-14.

Informed Consent: Retrospective study.

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