Retrospective Analysis of 356 Amniocentesis Results Performed for Karyotype Analysis

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Abstract

Objective: The aim of this study is to evaluat the indications, karyotype results and maternal fetal complication of amniocentesis performed in our clinic.

Methods: We retrospectively analyed the results of 356 amniocentesis cases performed in our clinic between january 2001 and june 2005 for different indications.The cases are evaluated in respect to amniocentesis indication, complications, cell culture success and genetic results. After performance of ultrasound, amniocentesis of done by free hand technicue, 20-22 G needle in used and 1ml of amniotic fluid is taken for every week of pregnancy.

Results: The most frequent indication for genetic amniocentesis was found as advanced maternal age (%45). The cell cultures were succesful in 350 of cases and there were only 6 cases in which cell culture were unsuccesful. Abnormal caryotypes were detected in 12 of 350 (%3.3). Abnormal caryotypes were; one Trisomy 13 (%0.2), one Trisomy 18(%0.2) and 6 trisomy 21 (%1.7). Other chromosomal abnormalities were thought to be normal variants (%1.1). In amniocentesis done for advanced maternal age (35) and high risk in triple test chromosomal abnormality were found %1.2 (2/158) and %3.7 (5/134) respectively. In amniocentesis done for abnormalities diagnosed by ultrasound choromosomal abnormality was reported as %4 (1/25).

Conclusion: Amniocentesis is still a widely used technique in prenatal diagnosis due to low fetal loss rate and high diagnostic ability. Complication risk of amniocentesis is low for both mother and fetus should be done in advanced maternal age and in high risk in triple test as a prenatal diagnostic test.

Keywords: Amniocentesis, indication, complication, chromosomal analysis.

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Karyotip analizi amacıyla genetik amniyosentez uygulanan 356 olgunun retrospektif analizi

Amaç: Kliniğimizde amniosentez uygulanan olguların endikasyonlarını, karyotip sonuçlarını ve işleme bağlı feto-maternal komplikasyonları incelemektir.

Yöntem: Ocak 2001 ve Haziran 2005 tarihleri arasında kliniğimizde çeşitli endikasyonlar ile amniyosentez uygulanan 356 olgunun karyotip sonuçları retrospektif olarak değerlendirildi. Bu kapsamda olgular amniyosentez için endikasyon, komplikasyon, hücre kültürü başarısı ve genetik sonuçlar yönünden değerlendirildi. Ultrasonografiyi takiben 16-18. gebelik haftasında 20-22 G iğne kullanılarak serbest el tekniği ile amniyosentez işlemi uygulandı ve her işlemde gebelik haftası başına 1ml amnion sıvısı alındı.

Bulgular: En sık endikasyon ileri anne yaşı olarak tespit edildi (%45). Olgularımızdan 6'sı dışında 350 sinde kültürde hücre üretildi (%98). Karyotip analizleri yapılan 350 olgunun 12'sinde (%3.3) çeşitli kromozom anomalileri saptandı. Bunlardan 6 olguda Trizomi 21 (%1.7) birer olguda Trizomi 13 (%0.2) ve Trizomi 18 (%0.2) tespit edildi. Tespit edilen diğer anomaliler normal varyant (%1.1) olarak yorumlandı. İleri anne yaşı (35) nedeniyle amniosentez uygulanan olguların %1.2'sinde (2/158), üçlü testte yüksek risk (1/270) nedeniyle amniosentez yapılan olguların %3.7 sinde (5/134) kromozom anomalisi tespit edildi. Ultrasonografide (USG) anomali saptanan olguların %4'ünde (1/25) kromozom anomalisi saptandı.

Sonuç: Amniosentez yüksek tanı ve düşük fetal kayıp oranları ile prenatal tanıda halen en sık kullanılan yöntemlerdendir. Amniosentezin anne ve fetus için komplikasyon riski düşük olup, ileri yaş gebeliklerinde, üçlü tarama testinde yüksek risk tespit edilmesi durumunda prenatal tanı amaçlı amniosentez yapılmalıdır.

Anahtar Sözcükler: Amniosentez, endikasyon, komplikasyon, kromozom anomalileri

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Introduction

It is possible to obtain information about fetal karyotype nowadays by means of interventional processes used for prenatal diagnosis. Amniocentesis is the oldest well-known prenatal diagnosis method and fetal sex determination was first done by "Barr" corpuscle existence in fetal cells which was obtained by means of amniocentesis by Fucs and Riis in 1956.[1] Steel and Breg showed in 1966 that fetal karyotype determination is possible in amniotic fluid.[2]

Amniocentesis for genetic purpose was first performed by transvaginal way and was performed blindly in following 60s by transabdominal way. It was performed in areas that placenta did not exist by static ultrasonography in the beginning of 80s.[3] In the last three decades, the most frequent indication for genetic amniocentesis has been advanced age gestation. Many studies have been done in our country and centers shared their experiences about this subject. Cengizoglu et al reported that 109 amniocenteses were performed due to advanced maternal age in 46 cases and increased risk in triple test in 19 cases.[4]

Amniocentesis for genetic purpose is generally performed in between 16^{th} – 20^{th} gestational week after 15th week. Even though it is a reliable diagnosis method in the hands of experienced people, it has fetal loss and fetal-maternal complication risks. Total fetal loss, spontaneous abortus and intrauterine death rates were reported in change from 2.4% up to 5.2% in a multi-centered study done by Ager and Oliver.[5] It was found in the randomized controlled study published by Tabor et al in 1986 that fetal loss risk was increased 1% as to control group.[6] Being used of scanning tests and becoming widespread of determination by ultrasonography for diagnosis of chromosomal anomalies in recent years caused amniocentesis count to rise.

Indication distribution, complications and fetal karyotype results of genetic amniocentesis applications done by different indications within approximately last five years in our clinic were evaluated retrospectively in this study.

Methods

Results of 356 cases were evaluated retrospectively whose karyotype determinations were done in Medical Biology and Genetic Department and who were applied amniocentesis due to finding anomaly and aneuploid markers in ultrasonography (USG), high risk in triple test (≥1/270) in between pregnants above 35 years applied to polyclinic of Obstetrics and Gynecology Department. Cases were evaluated in terms of indication, complication, cell culture success and genetic results for amniocentesis.

Written consent was taken from couples who accepted application before intervention. All cases were evaluated before intervention in terms of being Hepatitis transporter and Rh impropriety. Hitachi EUB 520 model ultrasonography device and 3.5 MHz transabdominal probe were used for amniocentesis process. All fetuses were evaluated in detail by ultrasonography before process and placenta localization was determined. After ultrasonography, amniocentesis was performed to cases in localization far from placenta by using 20-22 G injector which were possible and was performed to cases by passing transplacental which were not possible in between 16^{th} – 18^{th} gestational weeks and 1 ml amnion fluid was taken per week in each process.

The material taken from amnion fluid for cytogenetic examination was sent to genetic laboratory of Genetic Illnesses Department. The protocol that Hoehn et al[7] used was performed in cell culture. Materials were examined by using 20 metaphases display analysis system after 15-20 days of cell culture. 350 cases whose karyotypes were determined were taken into the study. Family anamneses of cases with chromosomal anomaly were retrospectively researched. Complementary statistic was used as statistical method.

Results

Average gestational week of cases that had been applied karyotype analysis was found as 18.33±1.43 and age was found as 34.96±6.7. Karyotype results of 6 (1.6%) of 356 cases were could not obtained due to previous bleeding and contamination. Amniotic fluid infiltration lasting 24-48 hours was found after the process in 6 cases which were applied amniocentesis. Amniocentesis indications were found as following; advanced maternal age (45%), high risk in triple test (38%), anomaly and marker in ultrasonography (7.1%), birth with anomaly history (0.5%).

Chromosome anomaly was found in 12 (3.4%) of 350 cases after the result of karyotype analyses in produced cells. Trisomy 21 was found in 6 of

cases (1.7%), trisomy 13 was found in one case (0.2%) and trisomy 18 was found in one case (0.2%). Anomalies interpreted as normal variant were found in remaining cases (1.1%) (Table 1). Chromosome anomaly was found in 2 (1.2%) of 158 cases which were applied amniocentesis due to advanced maternal age (≥35) and both of them were interpreted as trisomy 21. Chromosome anomaly was found in 5 (3.7%) of 134 cases which were applied amniocentesis due to high risk in triple test (≥1/270). Trisomy 21 was found in two of them, trisomy 13 was found in one case and trisomy 18 was found in one case. Polydactyly and ventriculomegaly were found in detailed USG examination of case found trisomy 13. Age risk existed in addition to high risk in triple test in 11 of 356 cases which were applied amniocentesis. No chromosomal anomaly was found in these 11 cases.

Chromosome anomaly (trisomy 21) was found only in one case within 25 cases which were applied amniocentesis due to anomaly or marker in ultrasonography. Polyhydroamnios, acid in fetal abdomen and shortness equal to femoris length were found in ultrasonography of this case in 16th week. They were multiple anomalies which were found in USG in cases with anomalies. Markers were those which were found in trisomies (short femoris, short humerus, nape thickness, cardiac, renal, gastrointestinal and other anomalies). No repeating anomalies were found in histories of cases which were found chromosomal anomalies.

Table 1. Chromosomal anomalies found by amniocentesis.

No	Indication	Chromosome anomaly	
1	Advanced maternal age	47, XX, +21	(Down syndrome)
2	Advanced maternal age	47, XX, +21	(Down syndrome)
3	High risk in triple scanning	47, XX, +13	(Patau Syndrome)
4	High risk in triple scanning	47, XX, +18	(Edward's syndrome)
5	High risk in triple scanning	47, XX, +21,	inv 9 (Down syndrome)
6	High risk in triple scanning	47, XY, +21	(Down syndrome)
7	High risk in triple scanning	47, XY, +21	(Down syndrome)
8	High risk in triple scanning	46,XY, Yqh +	(Normal variant)
9	High risk in triple scanning	46,XY, Yqh+	(Normal variant)
10	High risk in triple scanning	46,XY, 22pstk+	(Normal variant)
11	High risk in triple scanning	45,XY,t(14:21)	Balanced translocation

Discussion

Chromosomal anomaly was found in 3.4% of cases in karyotype determination after genetic amniocentesis in retrospective examination of our cases. Chromosome anomaly rate was found as 2.5% which were determined pathologically. Both these and our other rates are close and compatible with rates reported in the literature. Period of 5 years of this work also covers our training period.

As known, states such as advanced maternal age, parental balanced translocation, child history with chromosome anomaly and fetal anomy existence, high risk in triple test in ultrasonography are indications of amniocentesis.[8] Even though it is known as a safe procedure in the hands of experienced people; there are rates of complication reported as changing related with centers.

Sener et al reported in their work that there may be amnionitis about 0.1% and amniotic fluid infiltration about 1-2%.[9] Also, maternal mortality related with *E.coli* sepsis was reported within 48 hours after amniocentesis.[10] Possible complications belonging to mother are rare in amniocentesis. These are; perforation in visceral organs, amnion fluid emboly and Rh sensitizasyon.[11]

Amniotic fluid infiltration complaint lasting 24-48 hours was found in our eight cases and amniotic fluid infiltration stopped within 36-48 hours by resting in bed without applying any medication in our series as complication. No oligohydroamnios or ascendant infection was found in any case after amniotic fluid infiltration. No fetal loss was found in first three weeks in any our cases after applied amniocentesis procedure. It was not possible to give any rate in terms of other complications due to the fact that all monitoring process of cases was not done in our clinic. Also no maternal complication was reported to us.

Our success for producing fetal cell from amnion in our series was found as 98%. This rate was found compatible with 98% success rate reported by Guven et al.[12] The reason for being unable of production in amniocyt cell cultures in 6 cases was thought it may be related with previous bleeding and contamination as reported by Yayla et al.[13]

Chromosome anomaly risk increases dramatically in advanced maternal age of gestation. Chromosome anomaly was found in two (1.2%) of 158 cases which were applied amniocentesis due to only advanced maternal age (≥35). This rate is low when it is compared with chromosome anomaly about 5.8% of that Taner et al examined amniocentesis results in 359 advanced maternal age

cases.[14] The result of this may be age interval of cases which were applied amniocentesis due to advanced age.

Amniocentesis was applied to 134 cases due to high risk in triple test (≥1/270) and chromosome anomaly was found in 5 cases (3.7%). Two of found chromosome anomalies were trisomy 21 (1.4%) and one of them was trisomy 13 (0.7%) and one of them was trisomy 18 (0.7%). Trisomy 21 was found in six cases (1.3%), trisomy 18 was found in two cases (0.4%) and trisomy 13 was found in one case (0.2%) in the study of Kim et al which was performed on 458 cases.[15] This rate was found as compatible with our results.

Rizzo et al[16] found chromosome anomaly in 16.8% of 273 fetuses that they found anomaly in ultrasonography and Dallaire et al[17] chromosome anomaly in 27.1% in fetal anomalies. Chromosome anomaly was found only in 4% of 25 cases that we applied amniocentesis due to anomaly in ultrasonography in our series. Polyhydroamnios and acid in fetal abdomen was found in ultrasonography in this mentioned case and risk in Triple test was mentioned as 1/450 in again this case. Our rate difference series were ultrasonography anomalies deemed as aneuploid marker that no fetal anomalies were observed such as oligohydroamnios in 6 cases and polyhydroamnios in 7 cases. Amniocentesis was applied to four cases of other fetal anomalies due to nuchal edema; it was applied to one case due bilateral fissure lip, it was applied to two cases due to renal anomaly, it was applied to two cases due to fetal cardiac anomaly, it was applied to one case due to short extremity, it was applied to one case due to hyperechogenic intestinal loops. Nose root was as flattened in remaining case and amniocentesis was applied on demand of the family.

Gestations were terminated on demand of family of all cases which were found chromosome anomaly except cases found as normal variant. Pregnancy of cases which were found normal variant was monitored up to the end of gestation and no perinatal complication was found.

Consequently; complication risk of amniocentesis is low for mother and fetus and amniocentesis for prenatal diagnosis purpose should be applied to cases which were found anomaly in ultrasonography in advanced age gestations, in the existence of high risk in triple scanning. Its most important disadvantage is to get results later than other perinatal methods. Detailed ultrasonographic examination should be applied to cases which were found low risk at triple scanning. Our prenatal diagnosis success is 98%. Chromosome anomaly rate we obtained with this study is 3.3%.

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