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Amniocentesis Results of Van and Surrounding Provinces Between 2018 And 2020: A Tertiary Center Experience

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ABSTRACT

Amniocentesis is one of the safest procedures of prenatal diagnosis. This study aimed to show amniocentesis indications, rate of successful amniocyte culture, complications and outcomes of these pregnancies undergoing prenatal diagnosis in eastern part of Turkey, especially Van province and its nearby. Between 2018 and 2020, 253 patients were referred to our center for amniocentesis and 120 patients giving consent were enrolled from Van and its surrounding provinces. The most frequent indication was high risk in prenatal screening tests whereas the highest chromosomal abnormality was found in fetuses with abnormal ultrasonographic findings. Overall, 11.6% of fetuses had chromosomal anomaly. The most abundant chromosomal abnormality was Trisomy 21, followed by trisomy 18 and 13. Except one case with amniotic leakage no complications were found. Eleven pregnancies were terminated whereas one fetus was born with Turner syndrome and two pregnancies resulted with stillbirth. In conclusion, fetuses with abnormal ultrasound screening had higher chromosome anomaly diagnosed by amniocentesis and compatible with the literature, amniocentesis is a safe and successful method of prenatal diagnosis of chromosomal abnormalities. Van and its surrounding provinces showed a high rate of chromosomal anomaly when prenatal diagnosis was done.

Keywords: Amniocentesis, chromosome abnormality, prenatal diagnosis, Van

Introduction

Amniocentesis is the procedure of transabdominal amniotic fluid aspiration. Since 1988 amniocentesis has been available and initially done for Red cell alloimmunisation and gender determination. It has been used for chromosome analysis fom 1970s(1-3). All of these information is reached via fetal cells of dermis and urinary system found in the amniotic fluid. (4). Most common amniocentesis indications are high risk in first and second trimester screening tests and noninvasive prenatal test, a history of previous newborn with chromosome abnormality, maternal anxiety, translocation in parental chromosomes and abnormal findings on ultrasound exam. Amniocentesis is ideally performed between 15-20 weeks of gestation in order to decrease fetal loss, prevent talipes and amniotic fluid leakage and to gain live fetal cells adequately.(5, 6).

In this study, amniocentesis results and demographic properties of patients referred to our perinatology clinic for further genetic evaluation between 2018 and 2020 from Van and surrounding provinces(Mus, Agri, Bitlis, Hakkari, Igdir) were evaluated.

Materials and Methods

Beginning form January 2018 till November 2020, 253 pregnants referred to Van Yuzuncu Yil University Perinatology Clinic for prenatal karyotyping were included. Both the patient and her family were informed about amniocentesis including the details of the procedure, the risk of complications and alternative management. After given information, 120 patients gave informed consent for amniocentesis. Amniocentesis timing varied between 15-20 gestational weeks. All pregnancies were searched negative for Hepatitis B, Hepatitis C and HIV infections. Maternal blood type workup is performed

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in order to detect Rh alloimmunisation and Rh negative patients were given 300 mcg of Rh IgG after the procedure. Demographic characteristics, fetal ultrasonographic findings, indication of amniocentesis, quantitative fluorescent polymerase chain reaction (QF-PCR) and cytogenetic results of amniocyte cultures were evaluated with pregnancy outcome. Amniocentesis was performed according to Invasive Techniques for Prenatal Diagnosis of International Society of Ultrasound in Obstetrics and Gynecology (ISUOG)) published in 2016(7). Prophylactic sefazoline (2 gr) was done priorly to procedure. During the procedure convex ultrasound transducer of 2-5 MHz of GE Voluson E6 (General Electric Healthcare, ABD) was used by the same operator. Prior to the procedure presence of fetal heart beat, placentation, fetal biometry, amniotic fluid index, the safest region of puncture were evaluated by ultrasound. Spinal needle of 20 G (BD) was used to aspirate amniotic fluid. First 2 ml of amniotic fluid was disposed in order to exclude maternal contamination. Then, 20 mL of amniotic fluid was aspirated to 2 injectors without pistons. Fetal heart beat presence and amniotic fluid index and any hemorrhage were reevaluated after the procedure before procedure cessation. Patients were referred to Medical Genetic Division with the amniotic fluid aspirated. All the samples were undergone QF-PCR and chromosome analysis.

Statistical Analysis: Descriptive statistics for the categorical variables were presented as count, median, interquartile range and percentage. SPSS 22 (IBM Corp., NY, USA) statistical program was used for all statistical computations.

Results

In this study, a total of 120 patients applied to our perinatology clinic for 2 years were included. Demographic characteristics of the participants are illustrated in Table 1. Only one of the pregnancies was dichorionic diamniotic twin pregnancy and one of the twins amniotic sample was taken.

The most common indication for amniocentesis was detection of high risk of trisomy in prenatal tests (Table 2). The median risk of first and second trimester screening tests was 1/89 (IQR:1/50-1/155) and one amniocentesis was indicated because of Turner syndrome risk in noninvasive prenatal test. The second indication was fetal abnormality detection on ultrasound, most common abnormalities were increased nuchal translucency (%10), non-immune hydrops fetalis (%10) fetal heart (%4,2), urinary system (%3,3), multiple system abnormalities(%3,3), omphalocele (%1,7) and multiple soft marker detection (%1,7). The other fetal abnormalities included, cleft lip and palate, central nervous system abnormalities (ventriculomegaly, Dandy-Walker malformation) and nasal hypoplasia.

Five patients had previous newborns diagnosed with mucopolysaccharidosis type 6, GM1 gangliocidosis, Pompe Disease, X linked lymphoproliferative syndrome (Duncan Disease) and Alström Syndrome. In addition one patient had a child with Down Syndrome.

The vast majority of patients had adequate amniotic fluid index and placenta was places anteriorly. First puncture attempt was successful in 91,7% of patients. Puncture number was not significantly related to age, gestational week, amniocentesis indication and placentation

Result could not be obtained in 9,2% of QF-PCR and final diagnosis was obtained from amniocyte cultures. Most frequent abnormalities were Trisomy 21, Turner syndrome and Trisomy 18. One patient had Trisomy 13 and one patient had maternal contamination.

In 2,5% of patients amniocyte culture was unsuccessful. Amniocyte culture was successfully obtained in 97,5% (117/120) of patients. Most frequent diagnosis were Trisomy 21, Turner syndrome and Trisomy 18, coherent with QF-PCR. In addition, triploidy, trisomy 13 and inversion were also detected (Table 5). Genetic pathologies detected were given in Figure 1, most being Down Syndrome.

Three fetuses had balanced translocation [t(5;18)(q23.2;q23) ; 46,inv(X)(p22.1;q13) (26)/46,--(24) and 45,—,rob(14;21)(q10;q10)], 4 had polimorfism [inv(9) ve 9qh+]. Eighty-eight percent of fetuses' karyotype were either euploid, with polimorphism or balanced translocation.

The patients had a child with history of mucopolisaccharidosis type 6, GM1 gangliosidosis, Pompe Disease, X linked lymphoproliferative syndrome (Duncan Disease) and Alström Syndrome, all diagnosed molecularly. Those pregnancies were studied of these known mutations with prenatal molecular genetic testing. Only the fetus having a sibling diagnosed with Alström Syndrome had homozygote mutation of c.6828 C>A on ALMS1 gene; pregnancy outcome is unknown because patient was lost to follow-up.

More than one third of fourteen pregnancies that were diagnosed with chromosomal abnormality had non-immune hydrops fetalis. All of the patients were informed on prognosis, outcome of their pregnancies and pregnancy termination. Eleven pregnancies were terminated with consent. Remaining patients had delivery or stillbirth (Table 6). Parents of fetuses with balanced translocation had genetics consultation.

Table 1. Characteristics of Pregnant With Amniocentesis Indication

Characteristics	Median	Inter quartile range
Maternal age (year)	33	23-39
Gestational age (week)	18	16-19

Table 2. Indications of Amniocentesis

Indication	Number of patient (n)	Percentage (%)
High risk in prenatal screening test	50	41,7
Fetal abnormality on ultrasound	46	38,3
Maternal anxiety	10	8,3
Genetic disease in previous child	5	4,2
Advanced maternal age	4	3,3
Fetal growth restriction	3	2,5
Culture failure after chorion villus sampling	2	1,7

Table 3. Ultrasonographic Features and Number of Puncture of Patients

Ultrasonographic feature	Classification	Number of patient (n)	Percentage (%)
Placenta	Anterior	70	58,3
	Posterior	35	29,2
	Lateral	15	12,5
Amniyotik fluid index	Adequate	119	99,2
	Polyhydramniosis	1	0,8
Number of puncture	1	111	91,7
	2	8	6,7
	3	1	0,8

One of the patients with an euploid fetus had amniotic leakage. During the follow-up leakage continued for sixteen days and oligohydramniosis got severe till anhydramniosis. On the 17th day

leakage stopped and on 21st day the amniotic fluid was adequate. No apperent stillbirth complication observed after amniocentesis.

Discussion

Amniocentesis is a safe procedure of prenatal diagnosis of genetic abnormalities between 15-20th gestational weeks in which fetal loss risk 0,1-1% Besides unsuccessful culture risk is %0.1 which is lower than that of chorionic villus sampling(7). Thus amniocentesis is preferred rather than chorionic villus sampling. In this study, only one unsuccessful culture was observed, this small risk of 0,08% might resulted because of hemorrhagic amniotic fluid. Successful culture rate was 99,2%. This rate is higher than Yayla et al. and similar to the rate of Guven et al. and Arikan et al. (93%, 98% and 97,9%, respectively)(9-11).

The most frequent amniocentesis indication was high risk in prenatal screening test. This was similar to Timur(%29,9), Arikan(%65,5) and Guven(%43) et al; however different than Yayla et al; in which advanced maternal age was the leading cause, in this study this

Table 4. QF-PCR Results of Patients

Result	Number of patient (n)	Percentage (%)
No aneuploidy	96	80,0
No result	11	9,2
Trisomy 21	6	5,0
Turner Syndrome	3	2,5
Trisomy 18	2	1,7
Trisomy 13	1	0,8
Maternal contamination	1	0,8

Tablo 5. Amniocyte Culture Results of Patients

Result	Number of patient (n)	Percentage (%)
Euploid	100	83,4
Trisomy 21	7	5,8
Turner Syndrome	3	2,5
Unsuccessful amniocyte culture	3	2,5
Trisomy 18	2	1,7
46,--,t(5;18)(q23.2;q23)	1	0,8
46,inv(X)(p22.1;q13) (26)/46,-- (24)	1	0,8
45,--,rob(14;21)(q10;q10)	1	0,8
Triploidy	1	0,8
Trisomy 13	1	0,8

indication corresponded to 3.3% of pregnancies in this study group(9-12). Among the pregnancies with high risk prenatal screening test, 6% had chromosomal anomaly.

Abnormal fetal ultrasonography accompanied 38% abnormal amniocentesis result. This rate was very high with respect to rate of foreign studies which is 27%(11, 13). Studies including different regions of Turkey, declared this rate between 4,7-13%(9-12). The higher rate detected in this study might be a result of high percentage of non-immune hydrops fetalis that has 20% risk of fetal chromosomal anomaly(14, 15).

This study showed that 11.6% of patients had chromosomal anomaly. Previous national studies' chromosomal abnormality rates lied between 3,8-6,1%, which showed a higher rate in our study(9-12). Compared to those studies, our data contained 120 patients which were lower than the other four studies including a minimum of 165 and a maximum of 561 patients; this might lead higher rates of chromosomal abnormality. Besides, studies of comparison were of different regions of Turkey therefore, Van and surrounding provinces might have higher chromosomal abnormality rate. The closest region to Van that has been studied by Yayla et al. was at Diyarbakir and showed 4,2% chromosomal anomaly after amniocentesis. Other studies was performed in İzmir, Ankara and Kahramanmaras(9-12). In addition to regional factors, the studies might differ in chromosomal abnormality rates as indications of amniocentesis and patient characteristics were not the similar.

In the study, only one patient had fetal loss, on the sixth day after amniocentesis with no apparent procedural complication. Chromosomal abnormality

Table 6. Indication of Amniocentesis of Pregnancies With Abnormal Karyotype and Pregnancy Outcomes

Karyotype	Maternal Age (year)	Gestational Age (week+day)	Indication	Abnormal finding	Pregnancy Outcome
Trisomy 21	33	20+0	Fetal anomaly	NT: 3,5 mm	Termination
Trisomy 21	40	16+1	Fetal anomaly	Non-immune hydrops fetalis	Termination
Trisomy 21	35	16+2	Fetal anomaly	Non-immune hydrops fetalis	Termination
Trisomy 21	35	19+5	Fetal anomaly	Right aortic arch	Termination
Trisomy 21	44	18+3	High risk in prenatal test + Fetal anomaly	Scoliosis+ diastometamyelia	Termination
Trisomy 21	39	21 + 5	Fetal anomaly	Non-immune hydrops fetalis	Termination
Trisomy 21	20	20 + 3	Fetal anomaly	Non-immune hydrops fetalis + fetal growth retardation	38 weeks, 3040 g, ex fetus
45, X0	22	15 + 3	Fetal anomaly	Cystic higroma	Delivery
45, X0	24	17+6	Fetal anomaly	NT: 6 mm + AVSD	Termination
45, X0	25	15+6	Fetal anomaly	Non-immune hydrops fetalis	Termination
Trisomy 18	39	19+6	Fetal anomaly	bilateral radial agenesis, choroid plexus cyst ,club foot, unilateral vm (10.4)	Termination
Trisomy 18	22	12+6	High risk in prenatal test + Fetal anomaly	Tri 13/18 risk: 1/151 + cardiyac anomaly	Stillbirth at 15 weeks
Trisomy 13	34	16+2	Fetal anomaly	Holoprosencephaly + cystic hygroma + renal pelviectasia	Termination
Triploidy	26	22+2	Fetal anomaly	Fetal growth retardation + AVSD + ARSA	Termination

of the fetus was trisomy 18 therefore stillbirth might be a result of amniocentesis or aneuploidy.

This study had a number of limitations. Firstly, number of patients was lower than other national studies although patients were collected in similar period of time. Although being a referral center, this low number of participants might be reflecting this region's socioeconomic and sociocultural status. More than half of the patients (133/253) disagreed to have an amniocentesis and waited an evaluation after delivery. To overcome this limitation, case collection must be elongated.

To conclude, fetuses with abnormal ultrasound screening had higher chromosome anomaly diagnosed by amniocentesis and compatible with the literature, amniocentesis is a safe and succesful method of prenatal diagnosis of chromosomal abnormalities.

References

1. Evans MI. Reproductive risks and prenatal diagnosis. 1992.
2. Fuchs F, Riis P. Antenatal sex determination. Nature. 1956;177(4503):330-.
3. BAŞARAN S. Türkiye'de Prenatal Tanının Tarihçesi. Türkiye Klinikleri Tıbbi Genetik-Özel Konular. 2018;3(1):1-4.
4. Steele M, Breg Jr WR. Chromosome analysis of human amniotic-fluid cells. The Lancet. 1966;287(7434):383-5.

5. Early TC, Group M-TATC. Randomised trial to assess safety and fetal outcome of early and midtrimester amniocentesis. The Lancet. 1998;351(9098):242-7.
6. Gerbie A, Elias S, editors. Technique for midtrimester amniocentesis for prenatal diagnosis. Semin in perinat; 1980.
7. Ghi T, Sotiriadis A, Calda P, Da Silva Costa F, Raine- Fenning N, Alfirevic Z, et al. ISUOG Practice Guidelines: invasive procedures for prenatal diagnosis in obstetrics. Ultrasound Obstet Gynecol. 2016;48:256-68.
8. Center NG. Test Rehberi İstanbul2018 [updated 2018. Available from: https://www.nextgenetic.com/tr/formlar/test-rehberi/test-rehberi.
9. Güven MA, Ceylaner S. Amniyosentez ve Kordosentez ile Prenatal Tanı: 181 Olgunun Değerlendirilmesi. Perinatoloji Dergisi. 2005;13:25.
10. Salih SADC. Üniversite kliniğinde uygulanan amniyosentez sonuçları ve retrospektif analizi. Perinatoloji Dergisi. 2013;21(2):47-52.
11. Yayla M, Bayhan G, Yalınkaya A, Alp N, Fidanboy M, Görmüş H. Yüksek riskli gebeliklerde 2. trimester genetik amniyosentez: 165 olgunun klinik değerlendirilmesi. Perinatoloji Dergisi. 1999;7(1):40-6.
12. Timur AU, İbrahim; Gülhan İbrahim; Tan Saz, Nagehan; İleri, Alper; Özeren, Mehmet. Genetik amniyosentez yapılan 16-22 haftalık gebelerin amniyosentez sonuçlarının değerlendirilmesi. Perinatoloji Dergisi. 2013;21(3):101-6.
13. Dallaire L, Michaud J, Melancon S, Potter M, Lambert M, Mitchell G, et al. Prenatal diagnosis of fetal anomalies during the second trimester of pregnancy: their characterization and delineation of defects in pregnancies at risk. Prenatal Diag. 1991;11(8):629-35.
14. Deng Q, Fu F, Yu Q, Li R, Li F, Wang D, et al. Nonimmune hydrops fetalis: Genetic analysis and clinical outcome. Prenatal Diag. 2020.
15. Beke A, Joó J, Csaba A, Lázár L, Bán Z, Papp C, et al. Incidence of chromosomal abnormalities in the presence of fetal subcutaneous oedema, such as nuchal oedema, cystic hygroma and non-immune hydrops. Fetal Diagn Ther. 2009;25(1):83-92.