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Genetic Amniocentesis : 10 years experience at Songklanagarind Hospital

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ABSTRACT

Objective To evaluate the outcome of genetic amniocentesis.
Design Descriptive study.
Setting Songklanagarind Hospital, Prince of Songkla University.
Subjects Pregnant women at risk of fetal chromosomal abnormalities who underwent genetic amniocentesis from January 1988 to December 1997.
Main outcome measures The prevalence of fetal chromosomal abnormalities and pregnancy outcomes.
Methods Genetic amniocentesis and fetal cell culture were performed and the outcomes were followed.
Results One thousand and sixteen pregnant women underwent genetic amniocentesis. Advanced maternal age was the most common indication for genetic amniocentesis (78.8%). The success rate of cell culture was 97.9%. There were 45 cases (4.5%) of abnormal chromosomes. Forty cases were terminated and five cases continued their pregnancies. The indication of abnormal sonographic findings had the highest percentage of abnormal chromosomes (27.8%). Trisomy 21 was the most prevalent. The abortion rate within 14 days after the procedure was 0.3% (3/1016). Pregnancy outcome of the fetuses with normal chromosome included spontaneous abortion 1.3%, dead fetus in utero 1.1%, premature delivery 9.0%, and term delivery 88.6%. Genetic counselling seemed to be difficult in some cases with rare chromosomal abnormalities.
Conclusion The prevalence of fetal chromosomal abnormalities was 4.5%. The fetal loss rate was low. Genetic counselling is very important and should be done before the procedure. It should be provided by a team of nurse counsellors, obstetricians, paediatricians, paediatric surgeons and clinical geneticists.

Key words: amniocentesis, genetic, chromosome abnormalities

Amniocentesis is the most commonly performed invasive test for prenatal diagnosis of genetic diseases. The relative safety and accuracy of the procedure lead to the welcomed acceptance of amniocentesis as a method of genetic prenatal diagnosis.[1] The clinical use of amniocentesis and fetal chromosome analysis was introduced in Songklanagarind Hospital in 1988. Up to 1997, the number of amniocenteses carried out in the Perinatology Unit, Department of Obstetrics and Gynaecology, Songklanagarind Hospital dramatically increased. Fetal cell cultures were performed in the Human Genetics Unit, Department of Pathology, Songklanagarind Hospital. These services have been organised for ten years. This study was conducted to assess the prevalence of fetal chromosome abnormalities and the pregnancy outcomes of high risk pregnant women who underwent amniocentesis.

Materials and Methods

All of the amniocenteses in this study were performed on patients in the Perinatology Unit, Department of Obstetrics and Gynaecology, Songklanagarind Hospital. These patients included those referred from other provinces in Southern Thailand.

From January 1988 to December 1997, a total of 1016 cases of genetic amniocenteses were performed on pregnant women at risk of having a child with chromosome abnormalities. Each patient had an ultrasound examination to confirm gestational age, placental localization, and multiple gestation or anomalies. Genetic counselling had been provided for every pregnant woman before undertaking the procedure. The risks of the procedure, the likelihood of finding an abnormality and the response to the diagnosis of an abnormality had been discussed.

Real-time ultrasound (Toshiba Sonolayer SSA 250A, 3.75 MHz Curvilinear probe) was used for guidance of the needle and the fetal heart tones were monitored before the procedure. Amniocentesis was performed under ultrasound guidance with a No 20- to 22-guage spinal needle. Initially, the first 1-2 ml of amniotic fluid was discarded in order to avoid maternal cell contamination and then 20 ml of amniotic fluid was aspirated in a separate syringe and sent for cell culture. A 20-guage spinal needle had been used in the first three years. We had found a high incidence of blood-stained amniotic fluid, therefore, a 22-guage was used since then. Fetal karyotyping was performed using Giemsa-Trypsin-G-banding technique at the Human Genetics Unit, Department of Pathology, Songklanagarind Hospital.

All women were followed within 14 days after the procedures when the results had been obtained. All of them were asked to report complications and their pregnancy outcomes until delivery.

Results

A total of 1016 cases underwent genetic amniocentesis from 1988 to 1997. The number of cases increased dramatically from 13 in 1988 to 281 in 1997 (table 1). The mean age of the patient was 37.7 $\pm$ 4.5 years (ranging from 19 to 53 years). Approximately 50% were between 35-39 years of age (table 2). The mean gestation age at the time of amniocentesis was 17.6 $\pm$ 2.9 weeks (ranging from 14 to 31 weeks). Most cases were performed between 16-18 weeks (74.7%). Only 2.4% of cases were conducted after 24 weeks (table 3). Most of the amniotic fluid was clear (92%). Bloody tap was found in 72 cases (7.1%), and discoloured amniotic fluid was found in 0.9% of the cases.

Advanced maternal age was the most common indication for amniocentesis (78.8%). Some cases had two indications for prenatal diagnosis such as advanced maternal age with a history of an abnormal child, advanced maternal age with abnormal sonographic findings, and advanced maternal age with a family history of an abnormal child (table 4).

The success rate of cell culture was 97.9% (995/1016). Twenty one cases (2.1%) had cell culture failure. The prevalence of fetal chromosome abnormalities based on the first attempt of amniocentesis with successful cell culture was 4.5% (45/995). Of 995 cases, 50 were reported as abnormal results. Only 45 cases were confirmed to be abnormal. Five cases were reported as questionable, but all were proven to be normal on repeated procedures. Among 45 cases of

abnormal chromosomes, trisomy 21 was the most common. The remaining were various abnormalities such as translocation, mosaicism, 45,X and other rare abnormal chromosomes (table 5). Genetic counselling was provided after the results had been obtained in those cases with fetal chromosome abnormalities. It seemed to be difficult in some cases particularly those with rare fetal chromosome abnormalities because of lack of knowledge of those genetic disorders. The clinical geneticist and paediatricians were consulted to participate in counselling in order to provide accurate information for the couples.

Of 21 cases of cell culture failure, 18 were accepted to repeat a second procedure. The cell culture was successful in all cases and the results were reported as normal.

Among various indications for amniocentesis, abnormal sonographic findings had the highest percentage of fetal chromosome abnormalities (27.8%). The frequency of abnormal chromosomes was 6.5% (2/31) in those who had two indications for prenatal diagnosis (table 6).

Of 45 cases proven to be abnormal chromosomes, 40 were terminated and 5, whose fetal chromosomes were balanced translocations (3 cases), 46,XX / 45,X mosaicism (1 case) and trisomy 21 (1 case), decided to continue their pregnancies. The trisomy 21 case delivered prematurely and the fetus was complicated by duodenal atresia. This anomaly was prenatally diagnosed before performing amniocentesis. The couple chose to continue the pregnancy though all information had been informed.

The complications were reported within 14 days after the procedure in all cases. Abortion occurred in 3 cases (0.3%). All of them had normal chromosomes. However, two of them had abnormalities. One fetus had omphalocele and the other was complicated by hydramnios. Amniotic fluid leakage was found in 2 cases, but only one case had spontaneous abortion. Vaginal bleeding was reported in one case, but it then stopped without any complication. Chorioamnionitis was found in one case whose fetus was complicated by omphalocele with normal chromosomes. The pregnancy resulted in spontaneous abortion. No other serious complications occurred.

The pregnancy outcomes of those with normal fetal chromosomes were followed in only 475 cases. The fetal loss rate was 2.4% including spontaneous abortion (1.3%) and dead fetus in utero (1.1%). Spontaneous abortions occurred more than a month after the procedure in 3 out of 6 cases. Premature and term delivery were 9.0% and 88.6% respectively (table 7).

Discussion

The number of genetic amniocentesis performed annually in our institution has dramatically increased. Advanced maternal age was the most common indication for prenatal diagnosis, which is similar to that of other studies.[2-4] The mean gestational age was 17 weeks. We scheduled to perform procedures between 16 and 18 weeks when the amniotic fluid volume is approximately 150 ml and the ratio of viable to nonviable cell is greatest.[5] Some cases who were referred from other hospitals with the indication of abnormal findings in the late second trimester such as suspicion of fetal anomaly, hydramnios, or intrauterine growth retardation were performed at later gestational ages.

The prevalence of chromosome abnormality in our institution was high (4.5%) when compared with other studies.[2-4] It is probably because Songklanagarind Hospital is a referral centre and is the only place in Southern Thailand where fetal karyotyping is provided. Fifty five percent of these cases were referred from other hospitals.

The indication of abnormal ultrasound findings had the highest frequency of chromosomal aberrations (27.8%). This risk is much higher than advanced maternal age which is the most common reason for referral. Our data confirmed the previously reported high incidence of chromosome aberrations in fetuses with a sonographic diagnosis of structural anomalies, with figures ranging from 15.5 to 45 per cent. [6-11]

Several large collaborative trials have been undertaken to establish the safety of midtrimester

amniocentesis. The procedure-related loss rate was 0.5%.[12] The first randomised trial was conducted by a Danish collaborative study in 1986. They found a statistically higher rate of spontaneous abortion in the amniocentesis group. The observed difference of 1% corresponded to a relative risk of 2.3 of the amniocentesis group.[13] Most trials were non-randomised. The fetal loss rate was reported as approximately 3% to 5%.[2] In this study, the abortion rate within 14 days after procedures was low (0.3%). However, we are unable to show the procedure-related loss because of lack of controls. The overall fetal loss rate in our study was 2.4% which is not higher than that of other reports.[3,14]

It has been reported that factors found to be associated with increased risk of fetal loss included needle gauges larger than 20-gauge, placental perforation, the presence of discoloured amniotic fluid, and more than two needle insertions at any given time.[12] Regarding the placental perforation, Bombard and colleagues have stated that transplacental amniocentesis does not appear to increase the fetal loss rate in second-trimester procedures in the hands of experienced operators.[15]

In order to minimise fetal loss rate, we suggest that the procedure be performed by continuous ultrasound guidance, using a small gauge needle (number 20-22), avoiding placental perforation, and no more than two needle insertions performed on any given occasion.

Discoloured amniotic fluid was found in 0.9% of cases in our study. It has been reported in 1% to 7% of all second-trimester amniocentesis.[12] The significance of discoloured amniotic fluid of amniocentesis has been the topic of many debates. Fetal loss rates between 7% and 16% have been reported for women with discoloured amniotic fluid.[13,16,17] However, some authors have reported no differences.[18] We could not confirm this association in our study because most cases were unable to be followed until delivery.

The culture failure rate was 2.1% in our study. It should be approximately 0.5% to 1%.[19] We found that those cases were associated with bloody tap, small amount of fluid obtained, discoloured amniotic fluid, and contaminated culture media. Some authors have reported that amniotic fluid culture failure is more likely in fetuses with chromosomal abnormalities.[20,21] We could not confirm this association. Those cases where procedures were repeated had normal fetal chromosomes. Sample size may be too small to show this significance.

The limitation of this study must be acknowledged. The pregnancy outcome of more than 50% of cases could not be obtained though we have tried to collect that data. We concluded that is because they were referred from other hospitals and then referred back when the results had been reported. Therefore, only immediate complications, usually within 14 days after procedures, were reported in all cases. The real pregnancy loss rate might be higher or lower. In addition, we did not confirm postnatal chromosome studies, in particular, those cases with normal chromosomes.

Genetic counselling is of importance. It must be done in a non-directive way. Moreover, this service should be provided by various professionals including obstetricians, paediatricians, paediatric surgeons, nurse counsellors, clinical geneticists, and social workers. In our practice, the nurse counsellor provides genetic counselling before performing the procedure. If the result was reported as abnormal, an obstetrician and other professionals played a role in counselling. We suggest that an effective counselling team be organised in order to provide accurate and complete information for the couples.

In conclusion, amniocentesis is a relatively safe and reliable procedure. It should be performed by an experienced operator. All factors associated with increased risk of fetal loss must be avoided. Genetic counselling must be done before performing the procedure and all aspects should be discussed. It should be provided by various professionals. Improvement of laboratory is also needed to achieve the highest yield of successful cell culture.

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