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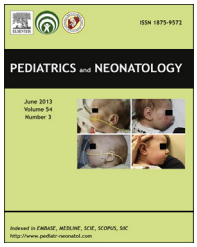

ORIGINAL ARTICLE

# Discrepancy of Cytogenetic Analysis in Western and Eastern Taiwan

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**Key Words**
amniocentesis;
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*Objective:* This study aimed at investigating the results of second-trimester amniocyte karyotyping in western and eastern Taiwan, and identifying any regional differences in the prevalence of fetal chromosomal anomalies.
*Methods:* From 2004 to 2009, pregnant women who underwent amniocentesis in their second trimester at three hospitals in western Taiwan and at four hospitals in eastern Taiwan were included. All the cytogenetic analyses of cultured amniocytes were performed in the cytogenetics laboratory of the Genetic Counseling Center of Hualien Buddhist Tzu Chi General Hospital. We used the chi-square test, Student *t* test, and Mann–Whitney *U* test to evaluate the variants of clinical indications, amniocyte karyotyping results, and prevalence and types of chromosomal anomalies in western and eastern Taiwan.
*Results:* During the study period, 3573 samples, 1990 (55.7%) from western Taiwan and 1583 (44.3%) from eastern Taiwan, were collected and analyzed. The main indication for amniocyte karyotyping was advanced maternal age (69.0% in western Taiwan, 67.1% in eastern Taiwan). The detection rates of chromosomal anomalies by amniocyte karyotyping in eastern Taiwan (45/1582, 2.8%) did not differ significantly from that in western Taiwan (42/1989, 2.1%) ($p = 1.58$). Mothers who had abnormal ultrasound findings and histories of familial hereditary diseases or chromosomal anomalies had higher detection rates of chromosomal anomalies (9.3% and 7.2%, respectively). The detection rate of autosomal anomalies was higher in eastern Taiwan (93.3% vs. 78.6%, $p = 0.046$), but the detection rate of sex-linked chromosomal anomalies was higher in western Taiwan (21.4% vs. 6.7%, $p = 0.046$).

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*Conclusion:* We demonstrated regional differences in second-trimester amniocyte karyotyping results and established a database of common chromosomal anomalies that could be useful for genetic counseling, especially in eastern Taiwan.
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## 1. Introduction

Second-trimester amniocentesis for amniocyte karyotyping is a common procedure for prenatal diagnosis of chromosomal anomalies.[1] Indications for amniocyte karyotyping are advanced maternal age, abnormal maternal serum screening results, abnormal prenatal ultrasound findings, and known family history of chromosomal anomalies or hereditary diseases.[2,3] In recent years, Taiwanese women have married later and become pregnant at older ages. After the implementation of national health insurance, antenatal care usage also increased.[4] Therefore, second-trimester amniocentesis for advanced maternal age and abnormal maternal serum screening test are routinely diagnostic recommendations at Taiwan. Karyotyping and risk calculation results not only offer the opportunity for pregnant women to have comprehensive genetic counseling, but also significantly decrease the prevalence of newborn babies affected by chromosomal anomalies.

We report the analysis of amniocyte karyotyping results of 3573 fetal amniocyte specimens from a cytogenetic laboratory in Taiwan. We investigated the indications for amniocyte karyotyping, prevalence of fetal chromosomal anomalies, and regional differences between western and eastern Taiwan.

## 2. Materials and Methods

### 2.1. Sample collection

From 2004 to 2009, pregnant women who underwent amniocentesis in their second trimester at three hospitals (Taipei Branch, Taichung Branch, and Dalin Branch of Buddhist Tzu Chi General Hospital) in western Taiwan and at four hospitals (Hualien, Yuli Branch, and Kuanshan Branch of Buddhist Tzu Chi General Hospital, and Mennonite Christian Hospital) in eastern Taiwan were included. The Ethics Review Board of the hospital reviewed and approved the study protocol. All cytogenetic analyses of cultured amniocytes were performed in the cytogenetics laboratory of the Genetic Counseling Center of Hualien Buddhist Tzu Chi General Hospital. We retrospectively reviewed and analyzed these results.

The indications for amniocentesis included advanced maternal age, suspected Down syndrome, suspected trisomy 18, suspected neural tube defect, familial hereditary disease or chromosomal anomaly, abnormal ultrasound findings, elective choice, and others. Mothers aged 35 years or more at the time of delivery were defined as having advanced maternal age. Suspected Down syndrome, suspected trisomy 18, and suspected neural tube defect were classified according to maternal serum screening results.[5–7] Amniocentesis performed due to maternal anxiety or for personal purposes was defined as elective.

### 2.2. Cytogenetic analysis

The high-resolution banding technique for R-bands by BrdU using Giemsa staining was performed using a standard *in situ* protocol.[8,9] Fifteen colonies were analyzed routinely for each karyotype report.

### 2.3. Statistical analysis

For evaluating the demographic data, we obtained the sample size ($n$), mean, standard deviation (SD), median, and range. We used the chi-square test to evaluate the independence of variances and Student $t$ test to evaluate the mean differences of the variances such as maternal age. We also used the nonparametric Mann–Whitney $U$ test to evaluate the differences of the variances without normal distributions in gravidity, spontaneous abortion, and artificial abortion. We used SPSS 16.0 to perform these analyses.

## 3. Results

From 2004 to 2009, we analyzed 3573 amniocyte cultures, 1990 (55.7%) from western Taiwan and 1583 (44.3%) from eastern Taiwan. Two samples failed karyotyping (0.056%) and were excluded from the analysis.

Maternal nationality was not significantly different in western and eastern Taiwan ($p = 0.699$). Pregnant women who underwent amniocentesis in eastern Taiwan were younger and had greater gravidity, spontaneous abortions, and artificial abortions (Table 1). The main indication for amniocyte karyotyping was advanced maternal age (69.0% in western Taiwan, 67.1% in eastern Taiwan). Suspected Down syndrome was the second indication and was almost equal in both western and eastern Taiwan (15.5% vs. 15.3%). The indications of amniocyte karyotyping, owing to suspected trisomy 18, suspected neural tube defect, familial hereditary disease, or chromosomal anomaly, and abnormal ultrasound findings were more frequent in eastern Taiwan, but advanced maternal age and elective karyotyping were more frequent in western Taiwan (Table 2).

Foreign-born mothers had a greater abnormal karyotype rate (7/134, 5.2%) than Taiwan-born mothers (80/3437, 2.3%) ($p = 0.033$). Mothers who had elective amniocyte karyotyping had significantly lower abnormal karyotype rates (1/374, 0.3%) than others (86/3197, 2.7%) ($p = 0.004$). Mothers younger than 34 years did not have

**Table 1** Demographic data of pregnant women who received amniocyte karyotyping in western and eastern Taiwan.

| | Western Taiwan | | | Eastern Taiwan | | | |
|---|---|---|---|---|---|---|---|
| Maternal nationality | *n* | % | | *n* | % | | *p* |
| Taiwanese | 1917 | 96.3 | | 1521 | 96.1 | | 0.699 |
| Foreigner | 73 | 3.7 | | 62 | 3.9 | | |
| Maternal age | Mean | SD | | Mean | SD | | |
| | 34.3 | 3.7 | | 33.8 | 4.5 | | <0.001 |
| Pregnancy history | Median | Range | Mean rank | Median | Range | Mean rank | |
| Gravidity | 2 | 1–9 | 1616.5 | 2 | 1–11 | 1931.5 | <0.001 |
| Spontaneous abortion | 0 | 0–5 | 1589.5 | 0 | 0–5 | 1651.3 | <0.001 |
| Artificial abortion | 0 | 0–8 | 1552.6 | 0 | 0–9 | 1683.7 | <0.001 |

*n* = sample size; SD = standard deviation.

a significantly lower abnormal karyotype rate (39/1399, 2.8%) than those aged 34 years or older (48/2171, 2.2%) ($p = 0.275$). The detection rate of chromosomal anomalies by amniocyte karyotyping in eastern Taiwan (45/1582, 2.8%) did not differ significantly from that of western Taiwan (42/1989, 2.1%) ($p = 1.58$) (Table 3). Mothers who had abnormal ultrasound findings and histories of familial hereditary disease or chromosomal anomaly had higher detection rates of chromosomal anomalies (9.3% and 7.2%, respectively) (Table 2). The types of chromosomal anomalies are listed in Table 4. The detection rate of autosomal anomalies was higher in eastern Taiwan (42/45. 93.3%) than in western Taiwan (33/42, 78.6%) ($p = 0.046$). Detection rates of sex-linked chromosomal anomalies were higher in western Taiwan (9/42, 21.4%) than in eastern Taiwan (3/45, 6.7%) ($p = 0.046$). *De novo* abnormal chromosomal detection rate was 8% (7/87).

## 4. Discussion

This study showed differences in amniocyte karyotyping results between western and eastern Taiwan. The detection rate of chromosomal anomalies in our center was similar to those of previous studies undertaken in Taiwan.[10,11] According to previous studies, newborns of foreign-born mothers had better neonatal outcomes in Taiwan.[12,13] Surprisingly, these mothers had higher rates of chromosomal anomalies in our study. However, it is well known that advanced maternal age is a risk factor for chromosomal anomalies. However, we found that it was not associated with higher risk of chromosomal anomalies in our study. Besides the socioeconomic and biological aspects of sample collection, selection bias could also explain the paradox.

The first three most common reasons for amniocyte karyotyping were advanced maternal age (57.29–58.85%), abnormal maternal serum screening results (21.34–26.17%), and abnormal ultrasound findings (4.50–8.86%) in previous studies. In our study, more mothers with advanced age underwent amniocyte karyotyping both in western (69.0%) and in eastern Taiwan (64.8%), but fewer mothers had abnormal maternal serum screening results and abnormal ultrasound findings in both western (15.8%, 2.4%, respectively) and eastern (20.0%,

**Table 2** Indications and detection rate of chromosomal anomalies by amniocyte karyotyping for various indications.

| | Indications* | | | | | | Detection rate of chromosomal anomalies† | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Western Taiwan | | Eastern Taiwan | | Total | | Western Taiwan | | Eastern Taiwan | | Total | |
| | *n* | % | *n* | % | *n* | % | *n* | % | *n* | % | *n* | % |
| Advanced age pregnancy | 1373 | 69.0 | 1025 | 64.8 | 2398 | 67.1 | 31 | 2.3 | 23 | 2.2 | 54 | 2.3 |
| Suspected Down syndrome | 309 | 15.5 | 239 | 15.1 | 548 | 15.3 | 5 | 1.6 | 9 | 3.8 | 14 | 2.6 |
| Suspected trisomy 18 | 1 | 0.1 | 44 | 2.8 | 45 | 1.3 | 0 | 0.0 | 1 | 2.3 | 1 | 2.2 |
| Suspected neural tube defect | 3 | 0.2 | 34 | 2.1 | 37 | 1.0 | 0 | 0.0 | 2 | 5.9 | 2 | 5.4 |
| Familial hereditary disease or chromosomal anomaly | 33 | 1.7 | 36 | 2.3 | 69 | 1.9 | 3 | 9.1 | 2 | 5.6 | 5 | 7.2 |
| Abnormal antepartum sonography | 47 | 2.4 | 50 | 3.2 | 97 | 2.7 | 2 | 4.3 | 7 | 14.0 | 9 | 9.3 |
| Elective | 224 | 11.3 | 150 | 9.5 | 374 | 10.5 | 1 | 0.4 | 0 | 0.0 | 1 | 0.3 |
| Others | 0 | 0.0 | 5 | 0.3 | 5 | 0.1 | 0 | N/A | 1 | 20.0 | 1 | 20.0 |
| Total | 1990 | | 1583 | | 3573 | | 42 | 2.1 | 45 | 2.8 | 87 | 2.4 |

Chi-square test: * $p < 0.001$, † $p = 0.177$. *n* = sample size; N/A = not available.

**Table 3** Results of amniocyte karyotyping in western and eastern Taiwan.

| | Western Taiwan | | Eastern Taiwan | | Total | |
|---|---|---|---|---|---|---|
| | *n* | % | *n* | % | *n* | % |
| Normal female | 781 | 39.3 | 601 | 38.0 | 1382 | 38.7 |
| Normal female variant | 149 | 7.5 | 137 | 8.7 | 286 | 8.0 |
| Normal male | 881 | 44.3 | 668 | 42.2 | 1549 | 43.4 |
| Normal male variant | 136 | 6.8 | 131 | 8.3 | 267 | 7.5 |
| Autosomal anomalies | 33 | 1.7 | 42 | 2.7 | 75 | 2.1 |
| Sex chromosomal anomalies | 9 | 0.5 | 3 | 0.2 | 12 | 0.3 |
| Total | 1989 | | 1582 | | 3571 | |

Chi-square test: $p = 0.048$. $n$ = sample size.

3.2%, respectively) Taiwan than those reported in the previous studies. Among women with abnormal serum screening results, abnormal karyotypes were found in 1.6% (5/313) in western and 3.8% (12/317) in eastern Taiwan. The detection rate of chromosomal anomalies by amniocyte karyotyping for women with abnormal ultrasound findings was significantly higher in eastern Taiwan (14.0%) than in western Taiwan (4.3%) and in previously reported data (1—8.86%).[10,11] Therefore, detailed antepartum sonography and family history review are important to identify the mothers at risk for chromosomal anomalies.

We found that the detection rate for autosomal anomalies was higher in eastern Taiwan and that of sex chromosomal anomalies was higher in western Taiwan, according to second-trimester amniocyte karyotyping. This finding was comparable to the data in the population-based birth registry of Taiwanese newborns in 2002. Advanced maternal age was the most important risk factor of chromosomal anomalies.[14] Since there was a discrepancy between different areas, other than socioeconomic status, personal habits, environmental factors, and occupational factors, a possible explanation was that there were still pregnant women, especially aboriginal women, at risk of chromosomal anomalies who did not receive antepartum genetic screening examinations in eastern Taiwan.[4,15]

There were some limitations to this study. We did not have information about some important risk factors such as socioeconomic status, ethnicity, smoking, alcohol consumption, and occupation. Although we included seven hospitals around Taiwan, selection bias could not be avoided completely.

**Table 4** Types of chromosomal anomalies in western and eastern Taiwan.

| | Western Taiwan | | Eastern Taiwan | | Total | |
|---|---|---|---|---|---|---|
| | *n* | % | *n* | % | *n* | % |
| Autosomal anomalies | | | | | | |
| Numerical anomalies | | | | | | |
| Trisomy 21 | 13 | 31.0 | 16 | 35.6 | 29 | 33.3 |
| Trisomy 18 | 1 | 2.4 | 3 | 6.7 | 4 | 4.6 |
| Numerical and structural anomalies | 1 | 2.4 | 2 | 4.4 | 3 | 3.4 |
| Structural anomalies | | | | | | |
| Translocation—reciprocal | 4 | 9.5 | 7 | 15.6 | 11 | 12.6 |
| Translocation—Robertsonian | 3 | 7.1 | 1 | 2.2 | 4 | 4.6 |
| Inversion | 1 | 2.4 | 8 | 17.8 | 9 | 10.3 |
| Addition | 2 | 4.8 | 1 | 2.2 | 3 | 3.4 |
| Derivative | 1 | 2.4 | 2 | 4.4 | 3 | 3.4 |
| Duplication | 1 | 2.4 | 0 | 0.0 | 1 | 1.1 |
| Insertion | 0 | 0.0 | 1 | 2.2 | 1 | 1.1 |
| Marker | 1 | 2.4 | 0 | 0.0 | 1 | 1.1 |
| Mosaicism | | | | | | |
| Numerical | 3 | 7.1 | 0 | 0.0 | 3 | 3.4 |
| Structural | 2 | 4.8 | 1 | 2.2 | 3 | 3.4 |
| Subtotal* | 33 | 78.6 | 42 | 93.3 | 75 | 86.2 |
| Sex chromosomal anomalies | | | | | | |
| Numerical anomalies | | | | | | |
| 45X | 1 | 2.4 | 0 | 0.0 | 1 | 1.1 |
| 45X mosaicism | 3 | 7.1 | 1 | 2.2 | 4 | 4.6 |
| 47XXY | 3 | 7.1 | 1 | 2.2 | 4 | 4.6 |
| 47XYY | 0 | 0.0 | 1 | 2.2 | 1 | 1.1 |
| Mosaicism | 2 | 4.8 | 0 | 0.0 | 2 | 2.3 |
| Subtotal* | 9 | 21.4 | 3 | 6.7 | 12 | 13.8 |
| Total | 42 | | 45 | | 87 | |

Chi-square test: *$p = 0.046$. $n$ = sample size.

## 5. Conclusion

In this 6-year retrospective analysis, we demonstrated regional differences in second-trimester amniocyte karyotyping results and established a database of common chromosomal anomalies that could be useful for genetic counseling, especially for women in eastern Taiwan.

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