Chromosomal abnormalities and reproductive outcome in Malaysian couples with miscarriages

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ABSTRACT

Introduction: This study was done to determine the prevalence of chromosomal abnormalities and the subsequent reproductive outcome in couples who had two or more miscarriages.

Methods: 56 couples with a history of at least two previous miscarriages were evaluated for prevalence and types of chromosomal abnormalities from their karyotype records. The study was a retrospective one, and subsequent reproductive outcome after a period of 12–24 months from the time of karyotyping was obtained by telephone interviews and scrutiny of the case records. The comparison of reproductive outcome was done by chi-square statistics.

Results: Five couples (8.9 percent) had a chromosomal abnormality in one partner. Three cases of reciprocal translocations t(5;11), t(9;14), dup(9q); one Robertsonian D/D translocation 13/14; and one mosaic Down syndrome male karyotype were found. Among the 32 couples available for follow-up, there was a lower incidence of subsequent live healthy births among chromosomally-normal couples (35.7 percent) compared to chromosomally-abnormal ones (25 percent). However, the difference was not statistically significant (p-value is 1.0). There was a lower incidence of subsequent abortions in chromosomally-normal couples (42.8 percent) compared to chromosomally-abnormal ones (50 percent), but the difference was also not statistically significant (p-value is 1.0).

Conclusion: Chromosomal abnormalities were seen in 8.9 percent of the couples, and translocations were the commonest abnormality found. The frequencies of subsequent live healthy births and subsequent abortions showed no significant difference between couples having normal karyotypes and those having chromosomal abnormality in one partner.

Keywords: chromosomal abnormalities, karyotype, miscarriages, reproductive outcome, translocations

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INTRODUCTION

Miscarriage or spontaneous pregnancy loss is a common clinical problem and occurs in 10%–15% of clinically-recognised pregnancies.[1] Chromosomal abnormalities in the embryo are the commonest cause for miscarriages.[2] The prevalence of chromosomal abnormalities in couples having recurrent pregnancy loss has been found to be higher than that in the general population (0.3%–0.4%).[3,4] When a chromosomal anomaly is found in one of the partners and is precisely identified, a more exact prognosis for future pregnancies can be given, and antenatal diagnosis may be offered in suitable cases.[4] No data is available on the prevalence of chromosomal abnormalities in Malaysian couples with recurrent miscarriages. Recurrent pregnancy loss is traditionally defined as three or more consecutive pregnancy losses before 20 weeks of gestation or of foetuses less than 500 g in weight.[5] However, the present study was done in couples who had two or more spontaneous miscarriages. The objectives of the present study were to determine the prevalence and type of chromosomal abnormalities in couples experiencing two or more miscarriages and to observe the reproductive outcome in terms of live healthy births, abnormal pregnancies or no pregnancy in these couples after a period of follow-up.

METHODS

56 couples with two or more spontaneous miscarriages were referred to the Department of Human Genetics for chromosomal analysis of their karyotype from their chromosomal records. These couples were referred by their treating physicians from different hospitals, including Hospital Universiti Sains Malaysia, Hospital Kota Bharu, and other hospitals in Kelantan and nearby states in

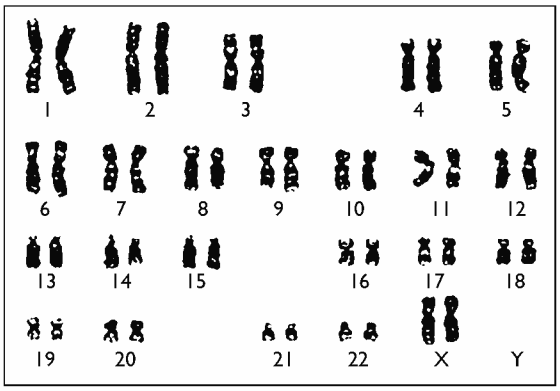

Fig. 1 Karyotype of a 29-year-old female patient shows a 46, XX, der(9) t(9;14)(q34;q31) pattern.

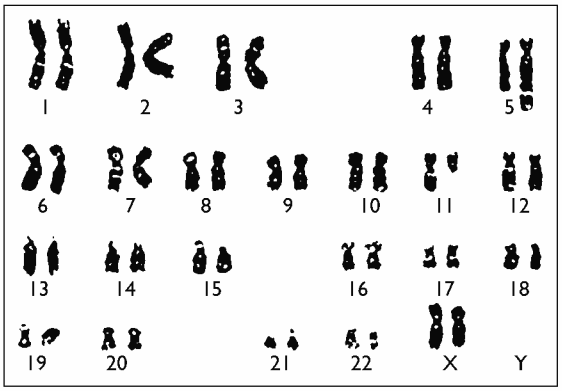

Fig. 2 Karyotype of a 25-year-old female patient shows a 46, XX, der(5)t(5 ;11)(q35;q13–25) pattern.

Malaysia, during the years 2005–2006. The present study was a retrospective one. The number of couples having miscarriages and the type of chromosomal abnormalities were obtained from the records and the chromosomal abnormalities were reconfirmed. After a period of 12–24 months from the time of their chromosomal analysis, the couples were asked about their reproductive outcome through telephonic interviews. Of the 56 couples in the study, the history of the reproductive outcome of only 32 couples could be determined. Case records scrutiny was also done to supplement the history. 24 couples could not be followed up due to several reasons, including a change of telephone numbers or residence due to transfers, a lack of follow-up case records or a lack of consent for giving the required information. Reproductive outcomes, like normal live births, miscarriages, ectopic pregnancies, still-births, any other abnormal pregnancies or no subsequent pregnancy, were noted.

Analysis of the results included the determination of the prevalence and description of chromosomal abnormalities, and the median age of the mothers. The association of chromosomal abnormalities with subsequent reproductive outcome was calculated by chi-square analysis using the Statistical Package for Social Sciences version 15.0 (SPSS Inc, Chicago, IL, USA). For chromosomal analysis, the standard procedure was used. Peripheral blood lymphocytes of the subjects were cultured along with phytohaemagglutinin and antibiotics, at 37°C for 72 hours. The metaphases were arrested by adding colcemid at the 70th hour of the culture, and the cultures were harvested using standard cytogenetic procedures. Karyotype analyses from G-banded metaphases were carried out according to the International System for Human Cytogenetics Nomenclature (ISCN) 2005. For all the cases, at least 20 metaphases were analysed.

RESULTS

A total of five couples (8.9%) with recurrent miscarriages had chromosomal abnormalities in one partner. Structural abnormalities were found in four cases, and one case had a numerical abnormality with mosaicism. Three cases of balanced reciprocal translocations (60%), one Robertsonian D/D translocation (20%) and one case of mosaic Down syndrome male (20%) were found. Out of the five abnormalities, three occurred in females (60%), while two occurred in males (40%). Case 1, a female patient aged 29 years, showed a karyotype pattern of 46,XX,der (9)t(9;14)(q34;q31) (Fig. 1). This was a case of balanced translocation. Here, a derivative chromosome 9 had resulted from a translocation of a segment of chromosome 14q31–32 to chromosome 9q34, resulting in a derivative chromosome 9. This couple had six miscarriages and one molar pregnancy, and no live birth was reported on follow-up.

The second case of chromosomal abnormality was seen in a 25-year-old female patient, who had a balanced translocation between chromosomes 5 and 11 with the karyotype pattern of 46,XX,der(5)t(5;11)(q35;q13–25) (Fig. 2). In this case, a segment q13–25 of chromosome 11 had been translocated to segment q35 of chromosome 5, resulting in a derivative chromosome 5. This woman had one healthy, chromosomally-normal baby after three miscarriages, as reported on follow-up. The third abnormal karyotype was detected in a 30-year-old male, who showed a karyotype pattern of 45,XY,der(13;14)(q 10;q10). This was a Robertsonian translocation, which originated through centric fusion of the long arms of chromosomes 13 and 14, with simultaneous loss of both short arms, resulting in a derivative (13;14). This couple had three miscarriages before the referral for karyotype analysis, but no pregnancy was reported on follow-up at 12 months.

A fourth abnormality was observed in a 35-year-old male, who had a mosaic karyotype pattern of 46,XY (80%) / 47,XY,+21 (20%), where 20% of the cells showed trisomy 21. However, this male patient had no apparent

Table I. Reproductive outcome of couples obtained via interviews and case file scrutiny after karyotyping.

Events	No. (%) of couples with normal karyotype	No. (%) of couples with abnormal karyotype
Live birth	10 (35.7)	1 (25.0)
Abortion	12 (42.9)	2 (50.0)
Ectopic pregnancy	2 (7.1)	0
Molar pregnancy	1 (3.6)	0
Stillbirth	2 (7.1)	0
Aneuploid birth (trisomy 13 with early neonatal death)	1 (3.6)	0
No pregnancy	0	1 (25.0)
Total	28 (100)	4 (100)

features of Down syndrome as per case records. This couple could not be traced for follow-up, and no details of subsequent reproductive outcomes were available. A fifth abnormality detected recently, was a woman aged 26 years, with a 46,XX,dup(9)(q13) karyotype pattern. She had two miscarriages and no live births on follow-up at 12 months.

Of the 32 couples who were available for follow-up, 11 cases had live, healthy babies. Thus, the overall incidence of live healthy births in the couples (comprising 28 chromosomally-normal couples and four carrying chromosomal abnormalities) was 34.4%. The median age of women in the study was 33 years. Among the 28 chromosomally-normal couples, ten (35.7%) had subsequent live healthy births, 12 (42.9%) had subsequent abortions, two (7.1%) had ectopic pregnancies, one (3.6%) had a molar pregnancy, one (3.6%) had a baby with trisomy 13 with early neonatal death and two (7.1%) had still-births. Among the four chromosomally-abnormal couples available for follow-up, one (25.0%) had a live healthy baby, two couples (50%) had subsequent abortions, and one couple had no subsequent pregnancy. There were no subsequent ectopic pregnancies, molar pregnancies or aneuploid births in the group of chromosomally-abnormal couples (Table I). The mosaic Down syndrome case was excluded from the evaluation as there was no available follow-up. The incidence of successful healthy pregnancies was 35.7% in the chromosomally-normal couples, while it was 25.0% in couples where there was translocation in one partner. Though the chances of a successful reproductive outcome were found to be higher in chromosomally-normal couples than those carrying translocations (odds ratio [OR] 1.667; 95% confidence interval [CI] 0.152–18.217), the difference was statistically not significant ($p = 1.0$). The chances of a subsequent abortion were lower among chromosomally-normal couples compared to those carrying translocations (OR 0.750; 95% CI 0.092–6.112), but the difference was also not statistically significant, using Fisher's exact test ($p = 1.0$).

DISCUSSION

In the present study, chromosomal abnormalities were seen in 8.9% of the couples having two or more pregnancy losses. A review of the literature showed that 4.7%–12.5% of couples with at least two spontaneous abortions carry chromosomal abnormalities in one partner.[6-9] Though high, the prevalence of chromosomal abnormalities in the present study was found to be within the limits reported in other study populations. The frequency of chromosomal abnormalities detected in couples with miscarriages depends on how strictly the candidates had been chosen for chromosomal analysis. Indeed, the selection of couples for chromosomal study is an important issue in some parts of Malaysia, where the population is apprehensive about genetic studies, and most cases agree to the tests only after other causes of recurrent miscarriages have been ruled out. Selective referral of cases was possibly a reason for the small sample size and the reasonably high prevalence of chromosomal abnormalities detected in the present study.

In the present study, the chromosomal abnormalities were t(5;11), t(9;14), dup(9q) and Robertsonian D/D translocation 13/14, making balanced reciprocal translocations the commonest abnormality (75%). In a previous study of 1,555 couples with recurrent first trimester miscarriages, balanced reciprocal translocations were the commonest abnormality found.[10] In another survey of 500 couples with recurrent miscarriages, translocations were seen in 44%, inversions in 8% and mosaicism in 48% (majority X chromosome) of the cases.[11] We found a higher involvement of chromosome 9 (seen in two cases) rather than inversions or X chromosome involvement in the present series. Diedrich et al also found a higher involvement of maternal X chromosome mosaicism, and reported that translocations of some chromosomes such as 1, 7 or 22 led to abortions, while those involving chromosome 5, 9, 14 or 21 led to the birth of handicapped children.[12] However, it was seen that the involvement of chromosome 5, 9, 14 or 21 only led to miscarriages in our series. In another study of

chromosomal abnormalities in 122 couples with three or more miscarriages, the abnormalities included one case of Robertsonian translocation 13q/14q (also seen in the present study), other translocations (2; 17), (5; 9), (11; 22), (17; 22) and two cases of X chromosome abnormalities.[13] The mosaic Down syndrome karyotype [46,XY (80%) / 47,XY,+21 (20%)], which was found in our series, has possibly not been described earlier.

In the present study, translocations were commoner in women (60%) compared to men, which was also reported by other authors. According to some authors, as male carriers of translocations have reduced fertility, females having chromosomal abnormalities are a commoner finding in couples with recurrent miscarriages.[14] Age-related poor egg quality possibly leads to higher chances of miscarriages, making spontaneous miscarriages commoner for women above the age of 40 years.[15] The median age of women in the present study series was 33 years, similar to the study by Clifford et al who reported a median age of 34 years in women having repeated miscarriages.[16] However, larger studies are needed to confirm our findings of a younger group of women having repeated miscarriages. In the present study, only 34.4% of the couples had a subsequent successful healthy pregnancy outcome, in contrast to a previous study where the incidence of a successful pregnancy outcome in couples who had miscarriages has been reported to be nearly 70%.[17] Our findings in the present study could be due to stringent case selection with fewer referrals. This possibly reflects reluctance on the part of patients to undergo genetic testing. The low rate of successful reproductive outcome found in the present study possibly does not reflect the actual success rate after miscarriages because of the very stringent case selection.

Some authors have shown that chances of successful reproductive outcome are not significantly lowered by the presence of translocation in one partner.[18,19] Similarly, in the present study, though there were more healthy live births in the chromosomally-normal couples (35.7%) compared to those carrying translocations (25%), the difference in the incidences were not statistically significant ($p = 1.0$; OR 1.667; 95% CI 0.152–18.217). Therefore, in the present study, couples with chromosomal translocation had fairly comparable chances of a successful reproductive outcome vis-à-vis those with no abnormalities, though an OR of 1.667 implied that chromosomally-normal couples had better chances of having a subsequent healthy live birth. In the present study, there were more subsequent abortions among carriers of translocation (50%), compared to chromosomally-normal couples, but the difference in the incidences was not statistically significant ($p = 1.0$). Chances of subsequent abortions were found to be lower among chromosomally-normal couples compared to those carrying translocations (OR 0.750, 95% CI 0.092–6.112). However, abnormal reproductive outcomes like molar and ectopic pregnancies, and stillbirths, were found in the chromosomally-normal couples, while none was found in couples carrying translocation. Sugiura-Ogasawara et al predicted a poorer prognosis in carriers of translocation, with a higher rate of subsequent miscarriages and lower rates of viable pregnancies.[20] Nearly 3% of couples with translocations were found to give birth to a chromosomally-abnormal child.[21] It has been seen that though the foetus sometimes carries the same translocation as the parent, it rarely gives rise to an abnormal phenotype at birth.[17,22] Portnoi et al also found that despite the presence of chromosomal abnormalities in the couples, there were few foetuses with chromosomal abnormalities on follow-up amniocentesis.[23] Interestingly, in the present study, while one chromosomally-normal couple had a trisomy 13 baby with an early neonatal death, there were no known aneuploid births among couples carrying translocations.

As this comparative follow-up study was done retrospectively in a very limited sample and over a short follow-up period, larger studies over longer periods are required to provide useful data for the Malaysian population. Antenatal diagnosis can be offered to detect the foetal karyotypes, and pre-implantation genetic diagnoses with assisted reproductive technology are offered as management for repeated miscarriages in some centres.[24] Even if such interventions are not available in some settings, the follow-up data of such couples with recurrent miscarriages would be useful to show the trend of possible future pregnancy outcomes. In the absence of data on the outcome of pregnancy, couples with balanced translocation can be informed only of the theoretical risk of abnormal pregnancies using hypothetical data.

In conclusion, chromosomal abnormalities are common in couples having recurrent miscarriages, and chromosomal analysis provides an important investigative tool for such couples. In the present study done in a Malaysian population, the prevalence of chromosomal abnormalities in couples having two or more miscarriages was found to be 8.4%, and balanced translocations were the commonest abnormality found. The presence of a translocation in one partner did not make the prognosis significantly worse, compared to couples having normal karyotypes. As the prognosis of couples having recurrent miscarriages may be good, even if one partner is carrying a translocation, the treating physician should encourage the couples, irrespective of their chromosomal status, to attempt for a healthy pregnancy. Follow-up data from

larger studies in Malaysian couples are needed to assist physicians in counselling their patients.

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