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Cytogenetic analysis in couples with recurrent pregnancy loss

Serenat Eriş Yalçın [1], Kuyaş Hekimler Öztürk [2], Halil Özbaş [2], Hilmi Baha Oral [3]

[1] Department of Obstetrics and Gynecology, University of Health Sciences Antalya Training and Research Hospital, Antalya, Turkey
[2] Department of Medical Genetics, Süleyman Demirel University School of Medicine, Isparta, Turkey
[3] Department of Obstetrics and Gynecology, Süleyman Demirel University School of Medicine, Isparta, Turkey

ORCID ID of the author(s)
SEY: 0000-0002-6465-325X
KHÖ: 0 0000-0002-7075-8875
HÖ: 0000-0002-7561-1450
HBO: 0000-0003-4544-2833

Corresponding Author
Serenat Eriş Yalçın
Department of Obstetrics and Gynecology, University of Health Sciences Antalya Training and Research Hospital, Varlik Mahallesi, Kazim Karabekir Cd. Muratpasa, Antalya, Turkey
E-mail: serenateris@hotmail.com

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Abstract

Background/Aim: Recurrent pregnancy loss (RPL), described as the loss of two or more pregnancies before 24 weeks of pregnancy, remains a concern for both the couples and the clinicians. Genetic factors tend to be strongly linked to reproductive failure among different etiologies. Our goal was to determine the rates and kinds of chromosomal defects in couples who had repeated pregnancy losses and a history of miscarriage in the first trimester.

Methods: This cross sectional study was conducted at a single tertiary center over a 3-year period. Couples who visited the outpatient clinic due to recurrent pregnancy loss and underwent tests to investigate the etiology were included in the study. Ages, number of abortions and genetic results of the patients were recorded. A total of 253 pairs had been tested for karyotype. Conventional cytogenetic method was used to identify chromosomal aberrations.

Results: Of 506 cases, chromosomal abnormalities were present in 15 (2.9%). Women were more frequently affected than men, with prevalences of 1.9% and 0.98%, respectively. Eight of the 15 cases (53.3%) showed structural deviations and 2 (13.3%) had numerical abnormalities. Additionally, 5 (33.3%) individuals were found to have chromosome variants.

Conclusion: Pregnancy loss is a major adverse life event, and the recurring nature of RPL can intensify the grief experienced. Aside from routine analyses of couples on anatomical, endocrine, and infection factors, these findings suggest that cytogenetic testing is required for an accurate approach to determine the cause of recurrent miscarriages.

Keywords: Chromosomal abnormalities, Cytogenetics, Recurrent pregnancy loss, Translocation

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Introduction

The term "pregnancy loss" refers to the spontaneous termination of a pregnancy before the fetus reaches viability. The phrase encompasses all miscarriages from conception to the 24th week of pregnancy [1]. There has been considerable debate in the literature about the definition of recurrent pregnancy loss (RPL) and, more specifically, to what extent this definition should be expanded or narrowed depending on the number of losses and whether they are consecutive. The number of pregnancy losses needed to meet the requirements for recurrent miscarriage is unknown, but ESHRE guidelines classify RPL as the loss of two or more consecutive pregnancies before 24 weeks of gestation [2]. This definition includes both spontaneous conception and pregnancy losses after ART but excludes ectopic and molar pregnancies and implantation failure. However, some researchers feel that even a spontaneous loss deserves consideration. RPL affects about 15% of births and concerns 1% of the general population [3].

It's difficult to pinpoint the exact cause of RPL because of its multifactorial existence. Despite comprehensive research over a decade to determine the underlying causes, the exact cause of pregnancy loss is only known in around half of the cases. After that, 50% of couples are diagnosed with idiopathic or unexplained RPL [4]. Uterine malformations, thrombophilic disorders, infections, immune dysfunction, multiple endocrine disorders, and parental chromosomal anomalies have all been suggested as contributing factors to pregnancy loss, either alone or in combination. Genetic factors tend to be strongly linked to reproductive failure through a variety of etiologies [5, 6]. Chromosomal etiology is very common in miscarriages, with chromosome abnormalities in the fetus accounting for 29 percent to 60 percent of abortions in the first trimester. A chromosomal abnormality in a partner affects between 3% and 6% of RM pairs, which is ten times higher than in the general population [7]. This chromosomal abnormality has been linked to either a balanced reciprocal translocation carrier parent or a recurrent numerical abnormality that is not normally inherited but can lead to recurrent miscarriages. Furthermore, carriers of chromosomal rearrangements are more likely to produce dysfunctional gametes, which can result in infertility, RPL, and malformations in infants. Balanced reciprocal translocation, Robertsonian translocation, gonosomal mosaic, and inversions are all examples of karyotype changes [8, 9].

While significant chromosomal anomalies and balanced chromosomal rearrangements found in couples who experience recurrent pregnancy loss are recognized as valid etiologies, the utility of preimplantation genetic diagnosis (PGD) is debatable. Fischer et al. [10], on the other hand, proposed that PGD would favor pregnant carrier couples with a history of RPL, increasing the likelihood of a healthy pregnancy significantly. While few structural rearrangements occur spontaneously, the majority tend to be hereditary, so couples with more than two pregnancy losses should undergo cytogenetic analysis and receive genetic counseling to rule out the likelihood of structural rearrangement.

The study's aim was to find out the rate and kinds of chromosomal defects in couples with recurrent pregnancy loss and a history of first-trimester miscarriage.

Materials and methods

This was a retrospective study at a single tertiary center over a 3-year period. The present study was approved by Süleyman Demirel University Faculty of Medicine Clinical Research Ethics Committee (05.04.2017 -73). Couples who visited to the outpatient clinic due to recurrent pregnancy loss (2 or more pregnancy losses) and underwent testing to investigate the etiology were included in the study. Ages, number of abortions and genetic results of the patients were recorded. A total of 253 pairs had been tested for karyotype. All patient samples were subjected to chromosome analysis using peripheral blood. Ordinary cytogenetic procedures were used to prepare metaphase chromosome preparations from peripheral blood cultures. RHG banding was used to conduct cytogenetic research. All patients had 20 metaphases examined, but anomalies and mosaic states required the study to be expanded to 50 metaphases. The chromosomal anomalies have been identified according to the International Human Cytogenetic Nomenclature System (ISCN 2009).

Results

The study included 253 couples (506 cases) with a history of recurrent miscarriage. The female and male partners' median ages were 33.94 (0.70) years and 35.61 (3.94) years, respectively. The number of recurrent abortions per pair ranged from 2 to 7 (Table 1). Chromosomal abnormalities were found in 15 cases (2.9 percent). Women were affected more frequently than men, with prevalence rates of 1.9 percent and 0.98 percent, respectively. Eight of the fifteen cases (53.3 percent) had structural deviations, and two (13.3 percent) had numerical deviations. Additionally, 5 (33.3 percent) individuals were found to have chromosome variants. Among the structural abnormalities that make up the largest group of chromosome anomalies, reciprocal translocations including chromosomes 1, 2, 7, 11, and 21 were observed in 3 cases. In one case, robertsonian translocation including chromosomes 13, 14 was observed. Inversion in chromosome 8 was observed in 3 cases. One of the 2 cases with numerical anomaly had mosaic with monosomy 45 X and the other had 47 XXX karyotype. In addition to these main chromosomal anomalies, pericentric inversion of chromosome 9 was observed in 4 cases. Chromosomal anomalies detected in patients are summarized in Table 2.

Table 1: Demographic data of patients

The median age of the female partner (yr)	33.94(0.70)
The median age of the male partner (yr)	35.61(3.94)
The mean body mass index for females	21.7(1.4)
The mean body mass index for males	19(1.2)
The mean number of abortions	2.3(0.8) (2 to 7 abortions/pairs)
The percentage of consanguineous marriages among couples	22.5%

Table 2: Cytogenetic findings of patients

Cytogenetic findings	Number of miscarriages	Maternal/paternal age
46 XY t(1,2) (p36,p23)	7	34
46 XX t(7,21) (p22,q22)	3	28
46 XX t(1,11) (p3,q13)	3	26
45 XX rob (13,14) (q10,q10)	4	36
47 XX+mar	2	33
46 XX inv (8) (q22q24.3)	3	24
46 XX inv (8) (p23q13)	5	29
46 XY inv (8) (p23q13)	5	37
47 XXX	3	27
Mos 45 X (5)/46, XX (25)	2	25
46 XX inv (9) (p11q13)	4	31
46 XX inv (9) (p11q13)	2	28
46 XY inv (9) (p11q13)	3	37
46 XY inv (9) (p11q13)	2	29
46 XX 13ps+	2	30

Of the 253 couples, 57 (22.5%) were consanguineous marriages. One pair with chromosomal anomaly was consanguineous marriage, and inv 8 (p23q13) karyotype was observed in this couple.

Discussion

The inefficiency of human reproduction is evidenced by the fact that a large percentage of all pregnancies do not succeed in a live birth. Miscarriage occurs in approximately 15-20% of all clinically recognized pregnancies, and total pregnancy loss is predicted to be 30% -50% [11]. The cause of most miscarriages before 12 weeks of pregnancy can be attributed to fetal aneuploidy.

For both the patient and the clinician, recurrent abortion remains a daunting process. In the first and second trimesters, chromosomal defects are the most common cause of spontaneous abortions, with a prevalence of approximately 70% during the first 6 weeks, 50% before 10 weeks, and 5% after 12 weeks [12]. The majority of fetal chromosome defects are de novo, according to numerous cytogenetic studies, and parental karyotypes appear normal. Various research, on the other hand, have been performed to assess the prevalence of chromosomal defects among couples who have had repeated miscarriages. This prevalence varies between 2.7 and 13.9 percent [13, 14]. These discrepancies may be due to variations in sample size and requirements.

The products of conception in translocation carrier pairs may have a regular karyotype, a balanced structural chromosome abnormality, or an unbalanced structural chromosome abnormality. The final scenario will result in a fetus being miscarried, a child being stillborn, or a child being born with serious congenital defects and significant mental disabilities [15].

The inversion of chromosome number 9 occurs with a high frequency of structural heteromorphism, a natural variation that is inherited by the family as a mendelian trait. Despite widespread disagreement, most cytogeneticists conclude that this variation is a harmless chromosomal polymorphism of the standard human karyotype [16]. The incidence is projected to be 1-3 percent of the general population, with Asians having the lowest rate with 0.25 percent. Among various species, inv (9) (p11q12) and inv (9) (p11q13) are the most common [17]. It's debatable whether heteromorphism will cause disease. Inv (9) has been linked to infertility and multiple abortions, according to Ueharas et al. [18]. Rodriquez et al. [19] believed Yq + was not linked to birth failure, while Genest et al. [20] assumed Yq + was linked to repeated miscarriages. In addition to habitual abortion, chromosome 9 inversions have also been associated with other diseases such as schizophrenia, bipolar disorders, mental retardation, hermaphroditism, obstetric infertility, and undescended testis [21, 22]. Infertility, repeated miscarriages, hydatidiform molar pregnancies, azoospermia, congenital abnormalities, growth retardation, and its association with irregular phenotype have all been recorded [23,24]. Garcia-Peiró et al. [25] investigated the sperm DNA integrity of a male patient with infertility and inv (9) karyotype and discovered high sperm DNA fragmentation, considerable meiotic changes, abnormal aneuploidy, and abnormal seminogram parameters, all of which can cause chromosomal imbalance in the generation. The higher incidence of Down syndrome and other abnormalities in the lineage of these carriers have been documented [26]. Although polymorphic variants containing pericentric inversions of chromosomes 9 and Y are said to be common in the general population, chromosomal inversions and polymorphic variants in recurrent pregnancy loss must be considered to determine future risk and provide better genetic counseling.

The most common chromosomal abnormalities found in this study, as recorded in other studies, are structural chromosomal abnormalities. According to the literature, chromosomal structural disorders affect 0.7 percent of the normal population, 2.2 percent of which had a miscarriage once, 4.8 percent of which had a miscarriage twice, and 5 percent of which had miscarriages three times [6, 9]. Translocations (reciprocal translocations, Robertsonian translocations, inversions, deletions, and duplications) are the most common structural chromosomal abnormalities in recurrent miscarriages [1, 3]. Balanced translocation prevalence among couples ranges from recurrent miscarriage to 0-31% in different studies. When one partner of a couple has a balanced chromosome translocation, the chances of miscarriage nearly double. A balanced translocation in a partner is found in 3-5 percent of couples who have recurrent miscarriages [6, 9, 15].

The male to female ratio of chromosomal rearrangement in our sample was 1: 2. Female predominance appears to be because chromosome abnormalities in fertile females may be linked to male infertility. Testart et al. [27] reported a higher frequency of translocation and inversion (3.6: 1) in men compared to women in a study conducted in couples who received intracytoplasmic sperm injection treatment. Due to poor mobility recorded in spermatozoa with a high prevalence of structural chromosome abnormalities, male reciprocal translocation carriers may expect a lower fertility rate.

Many Middle Eastern and Arab cultures and communities, including our own, practice consanguineous marriage. Because of the development of autosomal recessive gene mutations inherited from a shared ancestor, offspring of consanguineous relationships may be at a higher risk for genetic disorders. The more closely the parents are related biologically, the more likely their children would inherit similar copies of one or more harmful recessive genes. For example, first cousins are thought to share 12.5 percent (1/8) of their genes. As a result, their descendants would be homozygous (or, more specifically, autozygous) at 6.25 percent (1/16) of their gene loci on average (that is, they will receive identical copies of the gene from each parent at these sites in their genome). As determined in various studies, the rate of abortion among related couples is significantly higher compared to unrelated couples [28, 29]. It may not be possible to find the gene locus associated with poor obstetric outcome with standard karyotype analysis. As advanced genetic analysis becomes widespread, it may be possible to obtain PGD support by revealing the genetic defects in these couples.

Finally, our findings support previous research [3-6] that found an increase in the number of balanced chromosomal translocations in couples who have had two or more miscarriages compared to the general population. Couples with RPL are less likely to have numerical chromosomal anomalies. Sex

chromosome aneuploidy is the most common type of these anomalies, which occur infrequently.

Limitations

The limitations of our study include its retrospective nature, lack of genetic analysis results of the conception materials, and the fact that it does not include advanced examinations other than standard karyotype analysis.

Conclusion

Pregnancy loss is a major adverse life event. Our findings suggest that parental chromosomal disorders play a significant role and karyotype analysis is a clinically useful test in cases of recurrent pregnancy loss. Multidisciplinary approaches involving an obstetrician and a clinical geneticist will aid in the achievement of positive outcomes in these patients.

This paper has been checked for language accuracy by JOSAM editors.
The National Library of Medicine (NLM) citation style guide has been used in this paper.

References

1. Andersen AN, Wohlfahrt J, Christens P, Olsen J, Melbye M. Maternal age and fetal loss: Population based register linkage study. BMJ. 2000;320:1708-12.
2. Bender Atik R, Christiansen OB, Elson J, Kolte AM, Lewis S, Middeldorp S, et al. ESHRE guideline: recurrent pregnancy loss. ESHRE Guideline Group on RPL, Hum Reprod Open. 2018;2018(2):hoy004.
3. Rajasekhar M, Gopinath P. M, Sreelakshmi K, Satyamoorthy K. A Cytogenetic Study of Couples with Miscarriages: An Experience from Manipal Referral Centre. Int J Hum Genet. 2013;13(2):93-7.
4. Ljunger E, Cnattingius S, Lundin C, Annerén G. Chromosomal anomalies in first-trimester miscarriages. Acta Obstet Gynecol Scand. 2005;84(11):1103-7.
5. Elghezal H, Hidar S, Mougou S, Khairi H, Saad A. Prevalence of chromosomal abnormalities in couples with recurrent miscarriage. Fertil Steril. 2007;88:721–3.
6. Flynn H, Yan J, Saravelos SH, Li TC. Comparison of reproductive outcome, including the pattern of loss, between couples with chromosomal abnormalities and those with unexplained repeated miscarriages. Journal of Obstetrics and Gynaecology Research. 2013;40(1):109-16.
7. Branch DW, Gibson M, Silver RM. Recurrent miscarriage. New England Journal of Medicine. 2010;363(18):1740-7.
8. Toth B, Jeschke U, Rogenhofer N, Scholz C, Würfel W, Thaler CJ, et al. Recurrent miscarriage: current concepts in diagnosis and treatment. Journal of Reproductive Immunology. 2010;85(1):25-32.
9. Goud TM, Mohammed Al Harassi S, Khalfan Al Salmani K, Mohammed Al Busaidy S, Rajab A. Cytogenetic studies in couples with recurrent miscarriage in the Sultanate of Oman. Reprod Biomed Online. 2009;18(3):424-9.
10. Fischer J, Colls P, Escudero T, Munné S. Preimplantation genetic diagnosis (PGD) improves pregnancy outcome for translocation carriers with a history of recurrent losses. Fertil Steril. 2010;94(1):283-9.
11. Meshkat Z, Khadem Ghaebi N, Khajedaluee M, Aghili Z, Rostami S. Prevalence of chromosomal aberrations in couples with recurrent miscarriages in the city of Mashhad, Iran: a cross-sectional study. Arch Med Lab Sci. 2016;2(1).
12. Franssen MT, Korevaar JC, van der Veen F, Leschot NJ, Bossuyt PM, Goddijn M. Reproductive outcome after chromosome analysis in couples with two or more miscarriages: index [corrected]-control study. BMJ. 2006;332(7544):759–63.
13. Stephenson MD, Sierra S. Reproductive outcomes in recurrent pregnancy loss associated with a parental carrier of a structural chromosome rearrangement. Human Reproduction. 2006;21(4):1076-82.
14. Mozdarani H, Meybodi AM, Zari-Moradi S. A cytogenetic study of couples with recurrent spontaneous abortions and infertile patients with recurrent IVF/ICSI failure. Indian Journal of Human Genetics. 2008;14(1):1.
15. Campana M, Serra A, Keri G. Role of chromosome aberrations in recurrent abortion: a study of 269 balanced translocations. Am J Med Genet. 1986;24:341-65.
16. Gardner RJM, Sutherland GR 1996 Chromosome abnormalities and genetic counselling. 2nd edn. Oxford University Press, New York.
17. Lee KS, Lee JA, Song HH, Byun J, Ahn JS, Zang DY, et al. Solitary dural extramedullary plasmacytoma with inv (9) (p13q21). Am J Clin Oncol. 2004;27:638-9.
18. Uehara S, Akai Y, Takeyama Y, Takabayashi T, Okamura K, Yajima A. Pericentric inversion of chromosome 9 in prenatal diagnosis and infertility. Tohoku Journal of Experimental Medicine. 1992;166:417–27.
19. Rodriguez-Gómez MT, Martín-Sempere MJ, Abrisqueta J. C-band length variability and reproductive wastage. Human Genetics. 1987;75(1):56-61.
20. Genest P. Chromosome variants and abnormalities detected in 51 married couples with repeated spontaneous abortions. Clinical Genetics. 1979;16(6):387-9.
21. Nanko S, Kunugi H, Sasaki T, Fukuda R, Kawate T, Kazamatsuri H. Pericentric region of chromosome 9 is a possible candidate region for linkage study of schizophrenia. Biol Psychiatry. 1993;33:655-8.
22. Mc Candless F, Jones I, Harper K, Craddock N. Intrafamilial association of pericentric inversion of chromosome 9, inv (9) (p11-q21), and rapid cycling bipolar disorder. Psychiatr Genet. 1998;8:259-62.
23. Scarinci R, Anichini C, Vivarelli R, Berardi R, Pucci L, Rosaia L, et al. Correlation of the clinical phenotype with a pericentric inversion of chromosome 9. Boll Soc Ital Biol Sper. 1992;68:175-81
24. Kim JW, Lee JY, Hwang JW, Hong KE. Behavioral and developmental characteristics of children with inversion of chromosome 9 in Korea: A preliminary study. Child Psychiatry Hum Dev 2005;35:347-57.
25. García-Peiró A, Oliver-Bonet M, Navarro J, Abad C, Guitart M, Amengual MJ, et al. Dynamics of sperm DNA fragmentation in patients carrying structurally rearranged chromosomes. Int J Androl. 2011;34:e546–53.
26. Serra A, Brahe C, Millington-Ward A, Neri G, Tedeschi B, Tassone F, et al. Pericentric inversion of chromosome 9: prevalence in 300 Down syndrome families and molecular studies of nondisjunction. Am J Med Genet Suppl. 1990;7:162-8.
27. Testart J, Gautier E, Brami C, Rolet F, Sedbon E, Thebault A. Intracytoplasmic sperm injection in infertile patients with structural chromosome abnormalities. Hum Reprod. 1996; 11(12):2609-12.
28. Donbak L. Consanguinity in Kahramanmaras city, turkey, and its medical impact. Saudi Med J. 2004;25(12):1991-4.
29. Saad FA, Jauniaux E. Recurrent early pregnancy loss and consanguinity. Reprod Biomed Online. 2002;5(2):167-70.