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Original Article

The type and prevalence of chromosomal abnormalities in couples with recurrent first trimester abortions: A Turkish retrospective study

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ABSTRACT

Objective: Chromosomal abnormalities are more common in the first trimester abortions. We aimed to investigate the types and prevalence of chromosomal abnormalities in couples with recurrent first trimester miscarriages in Sivas, Turkey.
Materials and medhods: Three hundred couples (600 individuals) who had a story of recurrent abortion were included in the study. Chromosome analysis was performed after the preparation of lymphocyte culture with the standard method. Karyotype analyses were supported by FISH and aCGH studies.
Results: Total 26 chromosome abnormalities (8.7%) were found in the couples (19 females and 7 males). Fifteen cases (57.7%) were structural anomalies and eleven cases (42.3%) were numerical chromosomal aberrations. We detected 5 balanced translocations (33.3%), 4 Robertsonian translocations (26.7%), 3 inversions (20%), 2 duplications (13.3%) and one deletion (6.7%) among the structural anomalies. Mosaic monosomy X in five cases (45.4%), the combination of mosaic monosomy-trisomy X in three cases (27.3%), the combination of mosaic monosomy-trisomy and tetrasomy X in two cases (18.2%) and mosaic pentasomy X in only one individual (9.1%) were encountered as numerical chromosome aberrations. 19 cases had heterochromatic changes or other chromosomal variations (satellite increments and inv9).
Conclusion: Chromosome analysis in couples with recurrent miscarriage is necessary for possible preimplantation genetic diagnosis. As well as numerical and structural chromosome abnormalities, some chromosomal variations (heterochromatin and satellite increments etc.) may also contribute to recurrent miscarriages. Numerical chromosomal abnormalities are often associated with sex chromosomes and usually seen in females.

Introduction

Recurrent miscarriage (RM) may be defined as two or three and more consecutive pregnancy losses within the first 20 weeks of gestation and include approximately 0.8%–1.4% of pregnancies [1]. There may be many different reasons for this problem, but a possible cause can be identified in only about 50% of women with recurrent miscarriage [2]. The etiology of recurrent abortions may include endocrine or immunological disorders, parental chromosomal anomalies, hypercoagulability and anatomic abnormalities and these consecutive miscarriages are more common in women who are 35 years old and over [3,4]. If the main cause of pregnancy loss cannot be explained; this abortion may be evaluated as idiopathic or unexplained spontaneous abortion.

Chromosome anomalies are important especially in early abortions. The majority of such abnormalities may be due to chromosomal non-disjunction, translocation or another mutation [5]. A balanced or unbalanced karyotype in one of the partners in a couple as a structural chromosomal abnormality (reciprocal or Robertsonian translocations etc.) may result in recurrent miscarriage, or physical and/or mental disorder in next generation [6]. Numerical chromosome anomalies are also a major cause of abortion. Some researchers suggested that the most common chromosomal abnormality observed within first trimester spontaneous abortions may be single trisomies in abortion materials [7]. This problem may be associated with a balanced translocation in parents. The percentage of chromosomal variations was expressed as 5.5% in couples with abortions compared to 0.55% of the general population [8].

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We analyzed the frequency and type of chromosomal abnormalities in recurrent miscarriage in this study.

Materials and methods

Study population

A retrospective study was done in couples with recurrent first trimester abortions detected from 2012 to 2017 in Cumhuriyet University Research Hospital. Recurrent miscarriage may be defined as two or more failed clinical pregnancies that terminate involuntarily before 20 weeks [9]. In this context, three hundred couples (600 individuals) who had two or more consecutive miscarriage were included in the study. The study group consisted of individuals aged 18 years and older with no systemic disease. The types of recurrent miscarriage in the couples such as empty gestational sac, missed abortions, chemical pregnancies and spontaneous abortions were recorded for the study. The couples were screened for multiple thrombophilic gene mutations. Hormone levels of women were measured and ultrasonography and hysterosalpingography were used for the detection of possible abnormalities of the genital tract. Blood tests for immunologic risk factors including antiphospholipid antibodies, antinuclear antibodies and antithyroid antibodies were performed. Ectopic pregnancies, women with anatomic abnormalities, endocrine disorder, autoimmune disease and men with sperm anomalies were excluded from the study. This study was approved by Cumhuriyet University Ethics Committee.

Chromosome analysis

Metaphase chromosome preparations from the peripheral blood cultures were performed on the basis of standard cytogenetic protocol. Peripheral blood samples in heparinized injectors were processed for karyotyping. Phytohaemagglutinin (0.1 mL) was used for the stimulation of cell proliferation and cultured for 72 h at 37 °C, in RPMI 1640 medium. Colchicine was added to the cultures. G-banded karyotyping was performed with trypsin–giemsa banding procedure. Twenty metaphases were scanned for each individual but for suspicious cases (mosaicism etc.) the practice was extended to fifty metaphases.

FISH and aCGH

Further molecular techniques, such as fluorescence in situ hybridization (FISH) and array comparative genomic hybridization (aCGH) were used for detailed analysis. The first stage of the FISH method was the fixation of the sample for the target cells. The fixed cells were incubated (hybridized) in a buffer containing the labeled probe at a specified temperature. The subsequent washing step removed all unbound probe molecules. After the staining with 10 μl DAPI Antifade, the hybridized cells were analyzed by fluorescence microscopy. In aCGH, two genomic DNA samples (control genomic DNA and test genomic DNA) were labeled with different fluorophores and hybridized to a microarray with appropriate blocking reagents. The ratio of the fluorescent intensities of the fluorophores was measured for each gene on the array. This ratio provided a relative measure of the difference in gene copy number between the samples.

SPSS 22.0 program was used for statistical analysis.

Results

A total of 300 couples who had two or more miscarriage were included in the study. Mean maternal age in the group with recurrent abortion was 28.9 ± 5.56 (min: 18-max: 42). When the records were examined, it was found that 48% of the cases were chemical pregnancy, 33% were spontaneous abortion, 12% were missed abortion and 7% of them were blighted ovum. On average, the gravidity, parity and abortion numbers were 4, 1 and 3, respectively (Table 1). Among 300 couples, 26 chromosomal abnormalities (8.7%) were detected in this study. Nineteen of the cases (73.1%) were female and seven (26.9%) of them were male. Fifteen individuals (57.7%) showed structural abnormalities and eleven cases (42.3%) had numerical aberrations. In addition, there were 19 cases who have polymorphic variants or chromosomal variations. Chromosomal variations were predominant among the cases (Fig. 1).

Among structural abnormalities in the group, reciprocal translocations were detected in five cases (33.3%) and Robertsonian translocations were found in four cases (26.7%). Three subjects (20%) had inversion anomaly. One of the inversions was present on the Y chromosome. Duplications were detected in two individuals (13.3%). There was a deletion of long arm of X chromosome in one case (6.7%). Duplications and deletion were detected by aCGH (Cytoscan Optima, Thermo Fisher Scientific, USA).

Eleven numerical chromosome aberrations were found in couples with recurrent abortion and Turner mosaics were the most frequent numerical abnormalities. Five of the numerical anomalies (45.4%) were monosomy X mosaicism and in three cases (27.3%), the combination of monosomy-trisomy X mosaicisms were detected. Two cases (18.2%) had the combination of mosaic monosomy, trisomy and tetrasomy X. Only one of the numerical abnormalities (9.1%) was pentasomy X mosaicism. Mosaic cases were evaluated and confirmed by FISH probes (Vysis, Abbott, USA).

We detected 19 chromosomal variations (heterochromatin and satellite increments and inv9) in our study group. Most frequent polymorphic variant was inv9 with seven cases. Four individuals had 9qh + and the other heterochromatic variations were 1qh+, 16qh + and 15cenh + respectively. Five cases had satellit increments including 13, 14 or 15. chromosomes.

Discussion

Spontaneous miscarriage is the most common complication in pregnancy. A significant proportion of clinically defined pregnancies (about 15%) result in miscarriage [10]. Spontaneous abortion is often associated with chromosome anomalies. Approximately 40% of fetuses with an abnormal karyotype may inherit this anomaly from one of their parents [11] and parental chromosomal abnormalities can cause recurrent miscarriages. In this context, karyotype analysis of couples is important to predict the possible course of pregnancies. On average, 4–8% of couples with recurrent miscarriage have the problem of chromosomal abnormality [12]. In the study of Dutta et al., the rate of chromosome anomalies in couples with recurrent abortions was 6.7% [13]. De la Fuente-Cortés et al. detected this ratio as 7.6% [14]. Tsui et al. reported the highest percentage (9.9%) in this subject [15]. A total of 300 couples were studied and 26 chromosome abnormalities (8.7%) were found in our study. Female to male ratio of chromosome aberrations was about 3/1 in our group. Goddijn et al. found this ratio as 2/1 in

Table 1
The characteristics of couples with recurrent miscarriage.

Characteristics	Female	Male
Age	28.9 ± 5.56	29.2 ± 6.31
BMI	24,8 ± 2.36	24.3 ± 2.31
Tobacco use	34 (11.3%)	92 (30.6%)
Gravidity	4 (2-7)	–
Parity	1 (0-3)	–
Abortion	3 (2-5)	–

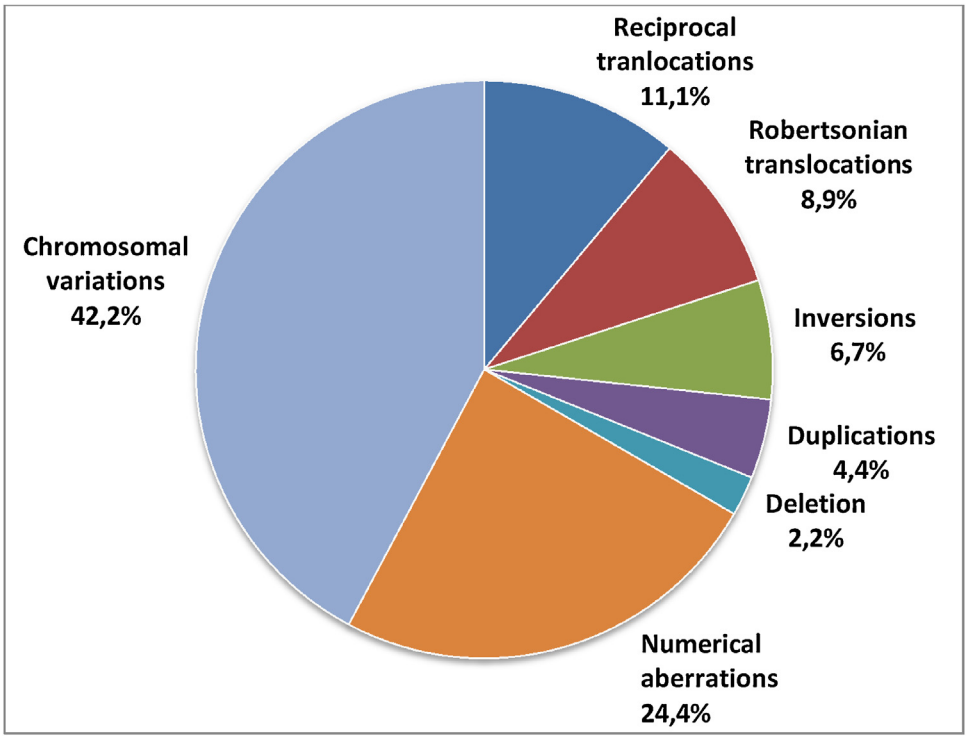

Fig. 1. The proportions of numerical and structural chromosome abnormalities and chromosomal variations.

another study related to abortions [16]. In a research by Turki et al., 77% of chromosome anomaly carriers in couples with recurrent miscarriage were female [17].

Although a lot of studies claim that structural aberrations are more common than numerical abnormalities [12,18], Farahmand et al. declared numerical anomalies as the most common chromosomal aberration in couples with recurrent abortions [19]. We have seen a bit dominancy of structural abnormalities in our study. Fifteen cases (57.7%) had structural and eleven cases (42.3%) had numerical aberrations.

Cytogenetic analyses revealed the higher prevalence of chromosomal translocations in couples with recurrent miscarriage and the analysis of each translocation case is necessary to assess the risk of further miscarriage [20]. El Hachem et al. suggested that the most common parental abnormalities are balanced translocations in recurrent pregnancy loss [21]. Unbalanced translocations account for about 1% of spontaneous miscarriages. The couples with balanced translocations may produce an unbalanced gamete again and again, resulting in recurrent miscarriage [22]. We observed 5 balanced translocations, 4 Robertsonian translocations, 3 inversions, 2 duplications and one deletion among structural anomalies of our couples (Table 2).

Structural abnormalities of Y chromosome may be important in terms of the miscarriage in couples. Inversion of Y chromosome has been reported in association with spontaneous abortions [23]. We found an inv (Y)(p11.32;q11.23) in the normal male partner of a couple with recurrent miscarriage. His father and brother also had same chromosomal abnormality.

Numerical aberrations usually present as the sex chromosomal aneuploidy in the couples with recurrent miscarriage. The presence of Mosaic Turner syndrome is common in female individuals with spontaneous abortion. Numerical chromosome abnormalities in our couples consisted of sex chromosome aneuploidies. Five of the numerical anomalies were mosaic X monosomies (Turner), three of them were the combinations of mosaic monosomy and trisomy X, two cases had the combination of mosaic monosomy, trisomy and tetrasomy X and only one case was mosaic pentasomy X in current research (Table 3).

About 16% of the karyotypic abnormalities in Turner's syndrome comprise of mosaicism. The risk of spontaneous pregnancy loss is high in mosaic Turner cases and also females with a distal Xp deletion [24]. Mosaic cases should be monitored closely due to the abortion risk in case of pregnancy. In a study by Doğer et al., 35 of 52 pregnancies of mosaic Turner cases (67.3%) resulted in spontaneous abortion [25].

Several changes that could not be accepted as chromosomal abnormalities should be analysed in relation to recurrent abortions. Chromosomal variations in couples such as heterochromatic polymorphisms, satellite increments have been implicated in mitotic instability and aneuploidy risk [26]. Some researchers

Table 2
Structural chromosome aberrations.

Chromosome aberrations	
46,XY,t(2;3)(p13;q27)	45,XX,rob(13;14)(q10;q10)
46,XX,t(3;12)(p23;p13)	45,XY,rob(13;14)(q10;q10)
46,XX,t(12;17)(p11;q13)	46,XY,inv(1)(q25;p36)
46,XX,t(3;8)(q26;p21)	46,XY,inv(17)(q23;q24)
46,XY,t(2;5)(p12;p23)	46,XY,inv(Y)(p11.32;q11.23)
45,XX,rob(13;14)(q10;q10)	46,XX 11q25 × 3
45,XX,rob(13;14)(q10;q10)	46,XY Xq13 × 2
46,XX,del(X)(q26)	

Table 3
Numerical chromosome aberrations.

Chromosome aberrations	
45,X[1]/46,XX[53]	45,X[3]/47,XXX[1]/46,XX[70]
45,X[2]/46,XX[53]	45,X[2]/47,XXX[1]/46,XX[55]
45,X[4]/46,XX[54]	45,X[1]/47,XXX[3]/46,XX[54]
45,X[3]/46,XX[55]	45,X[2]/47,XXX[1]/48XXXX[1]/46,XX[79]
45,X[3]/46,XX[84]	45,X[2]/47,XXX[4]/48,XXXX[1]/46,XX[60]
49,XXXXX[2]/46,XX[47]	

claimed that classical 9qh+/12qh + variants may contribute to recurrent miscarriages [27]. Heterochromatic changes occur at a relatively high frequency as a polymorphism in the population. On the other hand, chromosome analysis of some cases with recurrent miscarriage may show variations in the satellite regions of acrocentric chromosomes [28]. Although the presence of satellite increases in some recurrent miscarriages, the association of satellite increments with recurrent abortions is still unknown. In this study, we determined 19 chromosomal variations in couples with recurrent abortions (Table 4). Seven individuals had heterochromatic polymorphisms and five individuals had large satellite (satellite increase). Four of seven heterochromatic variations were 9qh+. The other samples were 1qh+, 15cenh + and 16qh+. Three of the satellite increases belonged to chromosome 15. The other two samples separately contained satellite increments of chromosomes 13 and 14. On the other hand, we found inv(9) in seven cases (2.3% of all couples). Pericentric inversions occur with a frequency of 1–2% in the general population. The risk of a chromosomal instability in the offspring of couples with pericentric inversions is between 5–10% [29]. Inversion of chromosome 9 is one of the most common structural variations seen in karyotype analysis [30]. One percent of people have such inversions in the heterochromatic region of chromosome 9, which can be evaluated to be a population variant [28]. It is a question mark whether these variants are associated with abortions. Although Tsui et al. said that pericentric inversion of chromosome 9 has not a role in recurrent abortion [15], in a study by Purandare et al., chromosome 9 showed the variations which included qh + and inv(9) and they claimed that these cases with chromosome 9 abnormalities had high frequency of first trimester miscarriages [31].

If there are chromosomal abnormalities in couples with RM, preimplantation genetic diagnosis (PGD) may have a beneficial effect on pregnancy outcomes. Suitable selection can allow transfer of embryos with normal karyotype. This procedure is expected to reduce spontaneous abortions, and increase live-birth rates [32]. We provide genetic counseling to couples with recurrent miscarriages who have chromosomal abnormalities and refer this patients to PGD in our center.

In conclusion, structural and numerical abnormalities such as translocations, deletions, duplications, trisomies and monosomies can be considered as the risk factors in terms of recurrent abortions. We believe that the heterochromatic variants and satellite increments may be present in the etiology of miscarriage. This claim is an important approach in our study and it is worthy of discussion in relevant centers. Further studies are needed to determine the possible association of some chromosomal variations with recurrent abortions in different populations. The limitations of our study were the relatively low number of subjects and the lack of comparison of couples with a control group. PGD has recently gained importance in couples with chromosomal abnormalities.

Table 4
Polymorphic variants and other chromosomal variations.

Chromosomal variations	
46,XY,9qh+	46,XX,15ps+
46,XY,9qh+	46,XY,15ps+
46,XX,9qh+	46,XY,inv(9)(p12;q13)
46,XX,9qh+	46,XX,inv(9)(p12;q13)
46,XX,1qh+	46,XX,inv(9)(p12;q13)
46,XX,15cenh+	46,XX,inv(9)(p12;q13)
46,XX,16qh+	46,XX,inv(9)(p12;q13)
46,XX,13ps+	46,XX,inv(9)(p12;q13)
46,XY,14ps+	46,XX,inv(9)(p12;q13)
46,XX,15ps+	

Conflict of interest

The authors declare that there is no conflict of interest regarding the publication of this article.

References

[1] Cao X, Cui Y, Zhang X, Lou J, Zhou J, Wei R. The correlation of sperm morphology with unexplained recurrent spontaneous abortion: a systematic review and meta-analysis. Oncotarget 2017;8(33):55646–56.
[2] Sharif K, Sharif Y, Watad A, Yavne Y, Lichtbroun B, Bragazzi NL, et al. Vitamin D, autoimmunity and recurrent pregnancy loss: more than an association. Am J Reprod Immunol 2018;80(3), doi:http://dx.doi.org/10.1111/aji.12991 e12991.
[3] Gabbai D, Harlev A, Friger M, Steiner N, Sergienko R, Kreinin A, et al. Pregnancy outcomes among patients with recurrent pregnancy loss and uterine anatomic abnormalities. J Perinat Med 2018;46(7):728–34.
[4] Xiang H, Yan H, Sun B, Feng F, Chen P. Decreased expression of long non-coding RNA SNHG7 cause recurrent spontaneous abortion through suppression proliferation and invasion of trophoblast cells via miR-34a. Am J Transl Res 2019;11(1):463–72.
[5] Gonçalves RO, Santos WV, Sarno M, Cerqueira BA, Gonçalves MS, Costa OL. Chromosomal abnormalities in couples with recurrent first trimester abortions. Rev Bras Ginecol Obstet 2014;36(3):113–7.
[6] De Krom G, Arens YH, Coonen E, et al. Recurrent miscarriage in translocation carriers: no differences in clinical characteristics between couples who accept and couples who decline PGD. Hum Reprod 2015;30(2):484–9.
[7] Yakut S, Toru HS, Çetin Z, Özel D, Şimşek M, Mendilcioğlu İ, et al. Chromosome abnormalities identified in 457 spontaneous abortions and their histopathological findings. Turk Patoloji Derg 2015;31(2):111–8.
[8] Sudhir N, Kaur T, Beri A, Kaur A. Cytogenetic analysis in couples with recurrent miscarriages: a retrospective study from Punjab, North India. J Genet 2016;95 (4):887–94.
[9] Priya PK, Mishra VV, Roy P, Patel H. A study on balanced chromosomal translocations in couples with recurrent pregnancy loss. J Hum Reprod Sci 2018;11(4):337–42.
[10] Niroumanesh S, Mehdipour P, Farajpour A, Darvish S. A cytogenetic study of couples with repeated spontaneous abortions. Ann Saudi Med 2011;31(1):77–9.
[11] 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. J Obstet Gynaecol Res 2014;40(1):109–16.
[12] El-Dahtory FA. Chromosomal abnormalities as a cause of recurrent abortions in Egypt. Indian J Hum Genet 2011;17(2):82–4.
[13] Dutta UR, Rajitha P, Pidugu VK, Dalal AB. Cytogenetic abnormalities in 1162 couples with recurrent miscarriages in southern region of India: report and review. J Assist Reprod Genet 2011;28(2):145–9.
[14] De la Fuente-Cortés BE, Cerda-Flores RM, Dávila-Rodríguez MI, García-Vielma C, De la Rosa Alvarado RM, et al. Chromosomal abnormalities and polymorphic variants in couples with repeated miscarriage in Mexico. Reprod Biomed Online 2009;18(4):543–8.
[15] Tsui KM, Yu WL Lo FM, Lam TS. A cytogenetic study of 514 Chinese couples with recurrent spontaneous abortion. Chin Med J (Engl) 1996;109(8):635–8.
[16] Goddijn M, Joosten JH, Knegt AC, van derVeen F, Franssen MT, et al. Clinical relevance of diagnosing structural chromosome abnormalities in couples with repeated miscarriage. Hum Reprod 2004;19(4):1013–7.
[17] Turki RF, Assidi M, Banni HA, Zahed HA, Karim S, Schulten HJ, et al. Associations of recurrent miscarriages with chromosomal abnormalities, thrombophilia allelic polymorphisms and/or consanguinity in Saudi Arabia. BMC Med Genet 2016;17(Suppl 1):69, doi:http://dx.doi.org/10.1186/s12881-016-0331-1.
[18] Pasińska M, Haus O, Adamczak R, Mucha B, Szymański W, Ferenc A, et al. [The results of cytogenetic investigations in 107 couples with recurrent spontaneous abortions from Pomerania-Kujawy region of Poland]. Wiad Lek 2005;58(11-12):630–5.
[19] Farahmand K, Kalantari H, Fakhri M, Fazeli AS, Moradi SZ, Almadani N, et al. Evaluation of 1100 couples with recurrent pregnancy loss using conventional cytogenetic, PGD, and PGS: hype or hope. Gynecol Endocrinol 2016;32(6):483–7.
[20] Ozawa N, Maruyama T, Nagashima T, Ono M, Arase T, Ishimoto H, et al. Pregnancy outcomes of reciprocal translocation carriers who have a history of repeated pregnancy loss. Fertil Steril 2008;90(4):1301–4.
[21] El Hachem H, Crepaux V, May-Panloup P, Descamps P, et al. Recurrent pregnancy loss: current perspectives. Int J Womens Health 2017;9:331–45.
[22] Kochhar PK, Ghosh P. Reproductive outcome of couples with recurrent miscarriage and balanced chromosomal abnormalities. J Obstet Gynaecol Res 2013;39(1):113–20.
[23] Sheth FJ, Shah UJ, Desai MJ, Sheth JJ. Clinical profile of inversion Y in people of Gujarat, West India. Int J Hum Genet 2011;11(4):245–8.
[24] Malla TM, Pandith AA, Dar FA, Zargar MH, Shah ZA. De novo Xp terminal deletion in a triple X female with recurrent spontaneous abortions: a case report. J Genet 2014;93(3):819–22.
[25] Doğer E, Çakıroğlu Y, Ceylan Y, Ulak E, Özdamar Ö, Çalışkan E. Reproductive and obstetric outcomes in mosaic Turner's Syndrome: a cross-sectional study and review of the literature. Reprod Biol Endocrinol 2015;13:59, doi:http://dx.doi.org/10.1186/s12958-015-0055-7.

[26] Dubey S, Chowdhury MR, Prahlad B, Kumar V, Mathur R, Hamilton S, et al. Cytogenetic causes for recurrent spontaneous abortions – an experience of 742 couples (1484 cases). Indian J Hum Genet 2005;11(2):94–8.
[27] Mierla D, Stoian V. Chromosomal polymorphisms involved in reproductive failure in the Romanian population. Balkan J Med Genet 2012;15(2):23–8.
[28] Sheth FJ, Liehr T, Kumari P, Akinde R, Sheth HJ, Sheth JJ. Chromosomal abnormalities in couples with repeated fetal loss: an Indian retrospective study. Indian J Hum Genet 2013;19(4):415–22.
[29] Rao L, Murthy K, Babu A, Venkata P, Deenadayal M, Singh L. Chromosome inversions and a novel chromosome insertion associated with recurrent miscarriages in South India. Arch Gynecol Obstet 2005;272(4):273–7.
[30] Demirhan O, Pazarbasi A, Suleymanova-Karahan D, Tanriverdi N, Kilinc Y. Correlation of clinical phenotype with a pericentric inversion of chromosome 9 and genetic counseling. Saudi Med J 2008;29(7):946–51.
[31] Purandare H, Fernandes NV, Deshmukh SV. Chavan S. Heterochromatic Variations and Pregnancy Losses in Humans. Int J Hum Genet 2011;11(3):167–75.
[32] Munné S, Chen S, Fischer J, Colls P, Zheng X, Stevens J, et al. Preimplantation genetic diagnosis reduces pregnancy loss in women aged 35 years and older with a history of recurrent miscarriages. Fertil Steril 2005;84(2):331–5.