REVIEW ARTICLE

Role of genetic factors in recurrent miscarriages - A review

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Abstract

Recurrent miscarriage is classically defined as three or more consecutive pregnancy losses in about 1–5% of couples trying to conceive. However, several researchers have amended this to two or more because of the recent increase in childless miscarriages. Recurrent miscarriage is a clinical challenge for clinicians because there are many possible causes, and diagnostic testing is expensive and time-consuming. Established causes of recurrent miscarriage are antiphospholipid antibodies, uterine anomalies, and abnormal chromosomes in either partner, particularly translocations. Uterine anatomical abnormalities, endocrine abnormalities, infections, immunologic factors, environmental factors, metabolic or hormonal disorders, sperm quality, and maternal and paternal age have each been linked. Among them, the genetic factor plays a significant role in recurrent miscarriage. Approximately 70% of miscarriage conceptions with sporadic spontaneous miscarriage reveal some chromosome abnormality. Specifically, recurrent miscarriage can be caused by a structural or numerical defect in the parents' or fetus' chromosomes. Recurrent miscarriage has been linked to several genes, including those involved in oxidative stress, angiogenesis, clotting, and inflammation. Despite several well-known etiologic factors, the etiology of recurrent miscarriage is unknown in over half of all instances. The current review aims to analyse the role of the genetic basis of recurrent miscarriages. *(Afr J Reprod Health 2022; 26[10]: 72-82).*

Keywords: Recurrent miscarriage, chromosomes, embryos, genetic factors, angiogenesis

Résumé

Les fausses couches récurrentes sont classiquement définies comme trois pertes de grossesse consécutives ou plus chez environ 1 à 5% des couples essayant de concevoir. Cependant, plusieurs chercheurs ont modifié cela à deux ou plus en raison de l'augmentation récente des fausses couches sans enfant. Les fausses couches à répétition sont un défi clinique pour les cliniciens car il existe de nombreuses causes possibles et les tests de diagnostic sont coûteux et prennent du temps. Les causes établies de fausse couche récurrente sont les anticorps antiphospholipides, les anomalies utérines et les chromosomes anormaux chez l'un ou l'autre des partenaires, en particulier les translocations. Les anomalies anatomiques utérines, les anomalies endocriniennes, les infections, les facteurs immunologiques, les facteurs environnementaux, les troubles métaboliques ou hormonaux, la qualité du sperme et l'âge maternel et paternel ont chacun été liés. Parmi eux, le facteur génétique joue un rôle important dans les fausses couches à répétition. Environ 70% des conceptions de fausse couche avec fausse couche spontanée sporadique révèlent une anomalie chromosomique. Plus précisément, les fausses couches à répétition peuvent être causées par un défaut structurel ou numérique des chromosomes des parents ou du fœtus. Les fausses couches récurrentes ont été liées à plusieurs gènes, y compris ceux impliqués dans le stress oxydatif, l'angiogenèse, la coagulation et l'inflammation. Malgré plusieurs facteurs étiologiques bien connus, l'étiologie des fausses couches à répétition est inconnue dans plus de la moitié des cas. La présente revue vise à analyser le rôle de la base génétique des fausses couches à répétition. *(Afr J Reprod Health 2022; 26[10]: 72-82).*

Mots-clés: Fausses couches à répétition, chromosomes, embryons, facteurs génétiques, angiogenèse

Introduction

Recurrent miscarriage is classically defined as three or more consecutive pregnancy losses. However, several researchers have now amended this to two or more because of the recent increase in childless miscarriages[1]. Recurrent miscarriage affects about 1-5% of couples, with at least 50% of those suffering from no evident pathology, with major implications for their relationship and quality of life[2]. The number of miscarriages required to define recurrent miscarriage is a point of contention[3].

Recurrent miscarriage is defined by the World Health Organization (WHO) as three or more consecutive pregnancy losses before the 20th week of pregnancy, whereas it is defined as two pregnancy losses with clinical evidence of pregnancy, according to the American Society for Reproductive Medicine (ASRM)[4]. Recurrent miscarriage should be about 1 in 300 pregnancies, based on the prevalence of sporadic pregnancy loss[5,6]. Several risk factors, such as structural uterine abnormalities and immunological illnesses, have been linked to recurrent pregnancy loss, but it's unknown why they affect some but not all pregnancies. In more than half of the women, no risk factors for miscarriage have been found[7,8].

Although many processes may eventually converge on a single pathway that promotes pregnancy loss, the pathophysiology of recurrent miscarriage varies according to maternal age and gestational age. Chromosomal abnormalities in the conceptus that prevent continued development and the collapse of the fetal-maternal contact, both of which result in cramping, bleeding, and miscarriage, are two common mechanisms. Established causes of recurrent miscarriage are antiphospholipid antibodies, uterine anomalies, and abnormal chromosomes in either partner, particularly translocations[9]. Other characteristics include detectible fetal heart activity pre-loss; normal fetal chromosomal content; advanced maternal age; or couple subfertility. Uterine anatomical abnormalities, endocrine abnormalities, infections, immunologic factors, environmental factors, metabolic or hormonal disorders, sperm quality, and maternal and paternal age have each been linked[10]. It has been reported that immune dysfunction, infection, and psychological stress are associated with recurrent miscarriage, but there is no established treatment methods[11]. Several genes, including those related to oxidative stress, angiogenesis, coagulation, and inflammation, have been associated with recurrent miscarriage. Despite numerous well-known etiologic variables, the cause of recurrent miscarriage remains unexplained in more than half of all cases. Hence, the current review has been aimed at analyzing the role of genetic factors in recurrent miscarriages.

Epidemiology

The study population, how early women recognize their pregnancy, and how different diagnostic criteria are used to diagnose recurrent pregnancy loss all play a role in determining the prevalence of pregnancy loss. The algorithm for the initial evaluation of recurrent miscarriages was shown in Figure 1. Contrary to stillbirth, few nations require healthcare practitioners to record miscarriages in a national registry, so the reported rates of pregnancy loss may be underestimated[12]. Recurrent pregnancy loss is estimated to affect between 1% and 5% of all women who become pregnant, according to data from large-scale studies in Europe and the United States[13,14]. In one meta-analysis, no regional differences in prevalence were discovered; however, societal and cultural views may prevent women from freely discussing their pregnancy losses, resulting in an underestimating of frequency in some areas[8]. Because of the lack of agreement on definitions and classifications, it's impossible to say whether prevalence has changed over time[15,16]. Importantly, despite medical intervention, the majority of women who have recurrent pregnancy loss have a high rate of subsequent live births. Indeed, one prospective cohort study found that 66.7% of women who are sent to a specialty clinic for recurrent pregnancy loss have a live birth within five years of being referred[17]. It is worth noting that this figure includes couples who gave up trying to get pregnant after that, so it could be underestimated. Furthermore, the presence of risk factors that impair fertility, such as uterine abnormalities or ovulation problems, influences the likelihood of a live birth[17]. Many studies have shown that maternal age at conception has an impact on pregnancy loss rates[14,18,19].

In one population-based investigation, the age-related risk of recurrent miscarriages was shown to be J-shaped, with the lowest risk (9.8%) among women aged 25–29 years[14]. Pregnancy loss becomes more likely in women aged 30–35 years and then rises to 33.2% in women aged 40–44 years. According to some research, more than half of all pregnancies in women over the age of 42 are lost[14,18,19]. Age-related pregnancy loss is predominantly caused by chromosome errors (aneuploidy) in the conceptus, with meiosis faults accounting for more than 90% of aneuploidy miscarriages[20–22]. Similarly, the incidence of age-related meiotic errors in pre-implantation embryos and oocytes closely mirrors the J-shaped curve of the age-associated pregnancy loss risk[23,24]. The number of previous recurrent miscarriages,

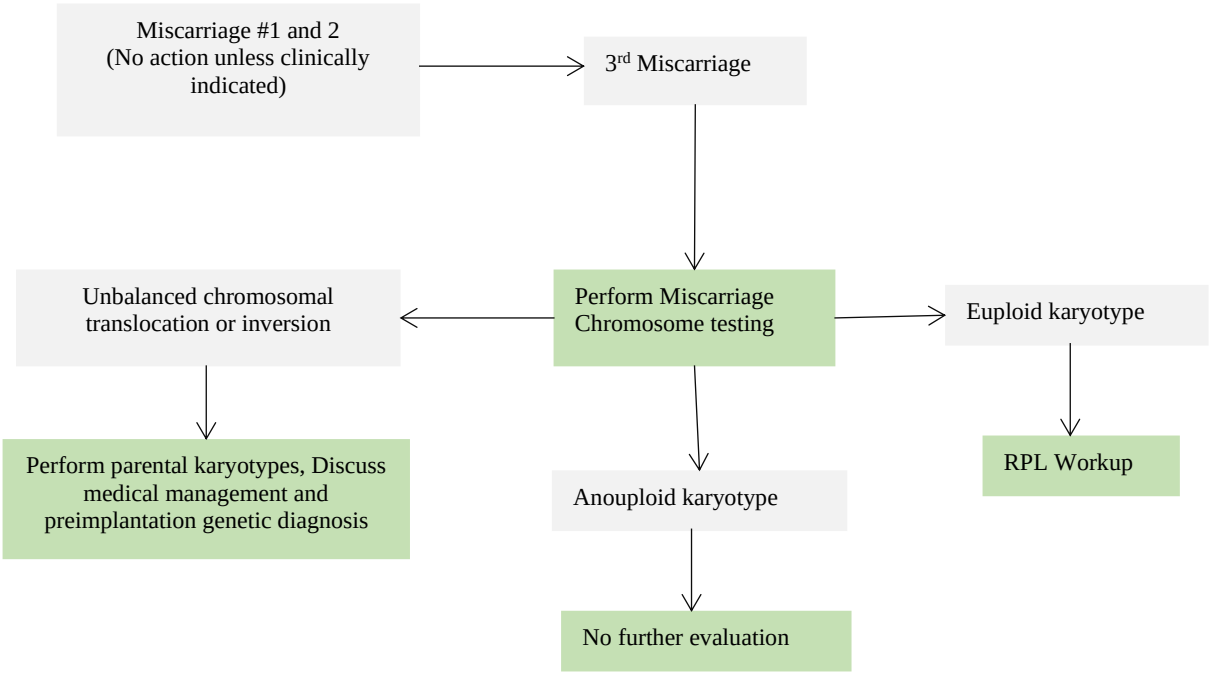

Figure 1: Algorithm for the initial evaluation of early recurrent miscarriages

regardless of the mother's age, is another risk factor for recurrent miscarriages. Indeed, population-based research has revealed that the age-adjusted odds ratio for miscarriage rises steadily after each pregnancy loss[25,26]. and is as high as 63% in women who have experienced six or more losses[27]. Miscarriage risk may also be influenced by genetic factors. The majority of research looking into the role of genetic variables in miscarriage risk used small sample sizes and used different definitions of recurrent pregnancy loss, yielding extremely conflicting results[28,29]. Large genome-wide association studies have found four different risk loci for spontaneous and recurrent pregnancy loss. All four of these risk loci are involved in progesterone production, placentation, and control of gonadotropins. Furthermore, community-based research has discovered that siblings of women with recurrent pregnancy loss have a twice higher rate of miscarriage than the general population[30]. Furthermore, in a comprehensive study, women who had miscarriages were more likely to have a family history of miscarriage; however, partners' family histories were not investigated[31]. Health, environmental, and lifestyle variables are also risk factors for pregnancy loss. Miscarriage is significantly more likely in women who were born small for gestational age[14]. Environmental factors like air pollution and endocrine-disrupting toxins may further increase the risk of miscarriage[32,33]. Smoking and alcohol consumption are other potential risk factors for miscarriage, and alcohol has been linked to poor fetal development[34].

Abnormal chromosomes in either partner

Concerning embryonic factors, the frequency of chromosome translocations in either partner is about 5%. De Braekeleer *et al.*[35] have reported that chromosomal abnormalities like translocations and inversions are positively associated with miscarriages. He analyzed a computerized database covering 22,199 couples generated from the literature on cytogenetic studies and concluded a rate of 4.7% for chromosomal structural rearrangements in couples suffering two or more miscarriages. The first case-control study of 1,284 couples examined whether translocations constitute a risk factor[9]. The previous study indicated a success rate of about 31.9% (15 of 47) at the first pregnancy after ascertainment of the carrier status, which is much less than that with normal chromosomes (71.7%, 849 of 1184), and a cumulative success rate of 68.1% (32 of 47). From these studies, it can be concluded that the prognosis of recurrent miscarriage patients with reciprocal translocations is poor, given that the study was

conducted over seventeen years and included severe cases suffering 10 and 13 miscarriages. At multi-centers in Japan, a total of 2,382 couples with a history of two or more consecutive miscarriages were studied[36]. Of these, 129 (5.4%) had an abnormal karyotype in one partner, excluding inversion 9, 44 in the males, and 85 in the females. Of the 2,253 couples, all had a normal karyotype in both partners. In the affected cases, 85 (3.6%) had translocations, with 13 being Robertsonian translocations. Twenty-nine of the 46 cases (63.0%) who became pregnant with reciprocal translocations in either partner experienced a live birth with natural conception. In contrast, 950 of 1,207 cases (78.7%) with normal chromosomes had successful live births, the difference being significant (P = 0.019). No infant with an unbalanced translocation was found in 29 cases of successful pregnancy following a recurrent miscarriage. In addition, intervention methods such as anticoagulants and supportive psychotherapy have improved the success rates for patients with and without translocations[36].

Fraussen *et al*.[37] compared reproductive outcomes in couples carrying a structural chromosome abnormality, and noncarrier couples were referred for chromosome analysis after two or more miscarriages. They reported that couples whose carrier status was ascertained after recurrent miscarriages have a low risk of viable offspring with unbalanced chromosomal abnormalities and that the chances of having a healthy child are as high as non-carrier couples despite a higher risk of miscarriage. In 92.9 percent of the paternal population, structural errors were found, and they were all balanced translocations. The most prevalent type of chromosomal aberration in a recurrent miscarriage patient is balanced translocation, and counselling the parents can assist improve the pregnancy result. It is possible to provide awareness on the translocation associated-miscarriages in the susceptible couples and reduce their risk by undergoing a cytogenetic analysis[38,39]. There was no significant difference in the number of live births between carriers and non-carriers when chromosomal abnormality carriers were compared[40]. According to this study, recurrent miscarriage can be caused by a variety of factors other than chromosomal abnormalities. By employing high-resolution bands and three-color FISH, a balanced complex chromosomal rearrangement involving chromosomes 3, 18, and 21 with four breakpoints was detected in a family[41]. Only 0.1% of the 1415 couples with complicated chromosomal rearrangements had complex chromosomal rearrangements, according to a retrospective study[42]. Complex chromosomal rearrangements, however uncommon, also have a role in recurrent miscarriage.

Abnormal embryonic karyotypes

Approximately 70% of miscarriage conceptions with sporadic spontaneous miscarriage reveal some chromosome abnormality. However, many spontaneous miscarriages with embryonic abnormalities occur by chance, which is not the case with recurrent miscarriages. On the other hand, it has been recognized that an abnormal embryonic karyotype may cause recurrent cases[26]. A retrospective analysis to examine the frequency of chromosomal abnormalities in products of conception from patients with recurrent miscarriages about the number of previous miscarriages of 1309 pregnancies with a history of 2–20 consecutive first-trimester miscarriages in Nagoya City University Medical Hospital shows that the frequencies of abnormal and normal embryonic karyotypes for each previous miscarriage were studied. The subsequent pregnancy outcome of patients whose previous miscarriages were karyotyped was investigated, as well as the predictive value of previous miscarriage karyotyping for subsequent miscarriages. Chromosomal analysis was performed on products of conception using a standard G-banding technique. As a result, the number of previous spontaneous miscarriages increased the miscarriage rate.

The frequency of abnormal embryonic karyotypes significantly decreased, and that of normal embryonic karyotypes increased dramatically with the number of previous miscarriages. Of the 1309 women, 458 (35.0%) miscarried, and 234 of the miscarriage conceptuses (51.1%) could be karyotyped. Among the 234 recurrent miscarriages, 114 (48.7%) had regular and 120 (51.3%) had abnormal chromosomes. Of the 114 sporadic miscarriages, 27 (23.7%) analyzed had a normal karyotype. The incidence of karyotype normality in recurrent barters was significantly higher than in controls. The incidence

of trisomy in sporadic miscarriages was considerably higher than in recurrent miscarriages. Forty-four of 71 patients with normal karyotypes miscarried later, and 23 of 60 patients with abnormal karyotypes miscarried later. Patients with a previous normal embryonic karyotype miscarry more frequently than those with an abnormal karyotype.

Therefore, the frequency of normal embryonic karyotypes significantly increases with the number of prior miscarriages, and a normal karyotype in a previous pregnancy may predict subsequent miscarriage. When the embryonic karyotype is normal after treatment of miscarriages, one should reconsider whether the therapy was appropriate and whether there are other causes of miscarriages in individuals experiencing six or more unexplained miscarriages[26]. The frequency of average karyotypes increasing suggests that the therapeutic approaches accepted worldwide are not sufficiently efficacious or that other causes of miscarriage, such as genetic abnormalities, are responsible. The treatment success rate for recurrent miscarriages may be estimated at 80% because the miscarriage rate caused by abnormal embryonic karyotypes is approximately 18%. Using in vivo ultrasound bio-microscopy, Laissue *et al*. identified quantitative trait loci (QTL) associated with diverse embryonic lethality phenotypes and subsequent embryonic resorption in 39 inter-specific recombinant congenic mouse strains.[43] They suggested that the short chromosomal intervals between the phenotypes would facilitate the study of a restricted number of candidate genes, potentially dysregulated in patients affected by recurrent miscarriage. Recurrent miscarriages can be caused by structural and numerical abnormalities in the chromosomes of both the parents and the fetus. As a result of nondisjunction, almost half of all miscarriage fetuses have chromosomal abnormalities, making cytogenetic screening critical in detecting spontaneous miscarriages. In a study of 151 recurrent miscarriage patients, 7.3 percent of the women exhibited chromosomal abnormalities, the most common of which was X chromosome mosaicism, followed by Robertsonian translocations and reciprocal translocations. X chromosome mosaicism and inversions were found in 2.1% of the fathers. The miscarriage fetuses were mostly trisomy, polyploidy, and monosomy[44,45].

Genetic abnormalities

Recurrent miscarriage has been suggested to be caused by mutations in encoding genes for various factors. Recurrent miscarriage may be caused by oxidative stress, thrombophilic factors, and immunologic factors such as Human Leukocyte Antigen and cytokine gene alterations. Recurrent miscarriage has been linked to polymorphisms in the androgen receptor (AR), estrogen receptor (ER), and progesterone receptor (PR). Inflammatory cytokine cascades have been implicated in the pathogenesis of recurrent miscarriage[46]. Polymorphisms in cytokine genes may affect the risk of recurrent miscarriage, but genetic association studies are often limited by small sample sizes. A meta-analysis of all available studies can increase the precision of these estimates. However, there were no significant association between recurrent miscarriage and tumor necrosis factor (-308A, or –238A), interferon-gamma (+874T), interleukin (IL)-1beta (-511T), IL-6 (-174G), or IL-10 (-1082A, or -819T, or -592A). Although significant associations were found with IL-1beta (- 31T) (odds ratio (OR) 2.12 (95% confidence interval (CI) 1.04 to 4.33)) and IL-6 (-634G) (OR 0.22 (95% CI 0.09 to 0.57)), the findings are not consistent with more than modest associations between these candidate cytokine polymorphisms and recurrent miscarriage[46,47]. Levrant *et al*. reported that IL-1 receptor antagonist gene polymorphisms were not risk factors for recurrent pregnancy loss[48]. Meta-analyses can get more accurate estimates of effect sizes by adding data from future association studies.

Faridi *et al*.[49] reported a higher prevalence of activated killer immunoglobulin-like receptors (KIR) genes; the BB genotypes were seen in women with recurrent miscarriage than in controls. The results indicated that the balance between inhibitory and activating receptor-mediated signals in natural killer (NK) cells is inclined toward a more activating state that may contribute to pregnancy loss. Several studies have shown that both hereditary and acquired thrombophilia increase the risk of adverse pregnancy outcomes, including miscarriages[50]. Increased risk has been found for carriers of known predisposing mutations. Although several findings have analyzed the association of polymorphisms of thrombophilia with recurrent miscarriage, most of them have been

unable to confirm these results, and the role of each of these mutations in recurrent miscarriage remains uncertain[51,52]. Goodman *et al.* reported in *Apoprotein E* (*Apo E2, Apo E3, Apo E4*) polymorphisms that women experiencing recurrent pregnancy loss had a significantly higher prevalence of Apo E3/4, E4/4 genotypes (21.7%) compared with control women (5.4%)[53]. They suggested that the *Apo E4* polymorphism may contribute to the thrombophilic risk factors of recurrent miscarriage. Bogdanova *et al.*[54] sought to verify whether variation in the gene's promoter encoding placental anticoagulant protein annexin A5 (ANXA5) represents a risk factor for recurrent pregnancy loss. Then they found a significant association[54]. It should facilitate the development of improved prognostic algorithms for recurrent pregnancy loss, involving a more precise assessment of individual disease risks and provide a guide to offering adequate therapies where relevant. In human chorionic gonadotropin–subunit genes (CGI), Rull *et al*. found that two single nucleotide polymorphisms in intron 2 of both CGB5 and CGB8 and four CGB5 promoter variants were linked to a significant protective effect in human chorionic gonadotropin–subunit genes (CGI)[55].

The carriers of minor alleles had a reduced risk of recurrent miscarriage. The findings encourage studying the functional effects of the identified variants on *CGB* expression and human chorionic gonadotropin hormone activity to elucidate further the role of *CGB* variation in recurrent miscarriage[55]. As for mitochondrial DNA (mtDNA), the recent report suggests that no apparent increased frequency of heteroplasmic mtDNA variations or amounts of aberrant mtDNA were detected in the recurrent miscarriage group, making it an unlikely cause of miscarriage[56]. Allelic variants of the detoxification genes that have impaired biotransformation functions may increase susceptibility to reproductive toxicity, leading to endometriosis, recurrent miscarriage, and poor pregnancy outcomes. Parveen *et al.* investigated *CYP1A1, CYP2D6, GSTT1, GSTP1,* and *GSTM1,* which are involved in the phase I and phase II detoxification systems, for their role in the etiology of unexplained recurrent miscarriage[57]. They observed significant protective effects of phase I wild-type genotypes and an association of the *GSTT1* null genotype with recurrent miscarriage, highlighting the importance of the balance of phase I or phase II detoxification systems in the etiology of recurrent miscarriage. Several studies have shown that both hereditary and acquired thrombophilia increase the risk of adverse pregnancy outcomes, including miscarriages[50]. Increased risk has been found for carriers of known predisposing mutations. Although several findings have analyzed the association of polymorphisms of thrombophilia with recurrent miscarriage, most of them have been unable to confirm these results, and the role of each of these mutations in recurrent miscarriage remains uncertain[51,52].

Among recurrent miscarriage patients, 30–40% are so-called "unexplained fetal losses," where no reason can be provided by routine gynecological, endocrine, or cytogenetic tests. About 70% of these cases in Europe and the USA are due to thrombotic episodes. The importance of hereditary thrombophilic factors for recurrent miscarriage is well recognized, and a statistical meta-analysis[58] compiled from 31 association reports convincingly demonstrates the roles of Factor V Leiden and Factor II prothrombin mutation (PTm) as hereditary recurrent miscarriage factors for Caucasians. Recent findings show that mutations in SYCP3, a gene encoding an essential component of the synaptonemal complex central to the interaction of homologous chromosomes, are associated with recurrent miscarriage[59]. The findings indicate that SYCP3 mutations are likely to produce an abnormal synaptonemal complex in a dominant-negative manner, as well as contribute to abnormal chromosomal behavior that may result in recurrent miscarriage. Combined with the fact that similar mutations have been identified in two males with azoospermia, data suggest that sexual dimorphism in response to meiotic disruption occurs in humans. Mice deficient in meiotic genes often show different phenotypes between males and females. It has been speculated that the checkpoint systems that mediate the completion of synapsis in the meiotic prophase differ between males and females[60]. SYCP3 deficient mice show complete meiotic arrest, leading to infertility in males, whereas in females, this leads to aneuploidy in the oocytes, which resembles a recurrent miscarriage[61,62]. Similar phenotypic discordance has also been reported in mice with an SMC1B deficiency or in those with hypomorphic mutations in the Rad51c gene[63–65]. According to the SYCP3

data, humans have a similar sexual-dimorphism phenomenon[59].

Preimplantation genetic diagnosis (PGD)

Preimplantation genetic diagnosis (PGD) for people suffering recurrent miscarriages is increasingly being performed worldwide[66]. The live-birth rates with PGD per in vitro fertilization (IVF) in reciprocal translocation carriers (23.7%) are comparable to or somewhat lower than those (63.0%) with a subsequent first natural conception[67]. However, it is challenging to compare IVF-PGD and natural conception in translocation carriers because information on the live-birth rate in the subsequent first pregnancy and time-based, not cycle-based, cumulative pregnancies after IVF-PGD or natural birth is very limited. Chun et al. described the details of 43 reciprocal and six Robertsonian translocation carriers separately, though it was unclear whether the patients consisted of only recurrent miscarriages[68]. Studies have reported that 14 of the 43 (32.6%) patients with reciprocal translocations succeeded in having a baby after 59 started cycles (mean age, 31.5 ± 4.0). In comparison, one of six (16.7%) patients with Robertsonian translocations had a baby after 11 cycles (mean age, 30.8 ± 3.5)[66]. The success rate with first-cycle PGD (32.6%) was comparable to the natural pregnancy rate (31.9%) reported by us for patients with reciprocal translocations[9].

Regarding the Robertsonian cases, the success rate by natural pregnancy (63.6%) is much higher than that (16.7%) with first-cycle PGD. In the case of Robertsonian translocations, PGD may not be necessary because natural success rates are relatively reasonable. Platteau *et al*. determined the aneuploidy rate in embryos of women with unexplained recurrent miscarriages and evaluated whether preimplantation genetic diagnosis for aneuploidy screening could be a feasible approach to improve the possibility of successful pregnancy in these couples[69]. They reported that the aneuploidy rate was 43.85% and 66.95% in the younger and older groups. There was no therapeutic evidence to prescribe IVF with or without a preimplantation genetic diagnosis for aneuploidy screening for this heterogeneous group of patients. Mastenbroek *et al.* conducted a multi-centre, randomized, double-blind, controlled trial comparing three cycles of IVF with and without preimplantation genetic screening in 408 women aged 35 through 41 years of age[70]. The data suggested that the ongoing-pregnancy rate was significantly lower in the women assigned to preimplantation genetic screening and that the women assigned to preimplantation genetic screening also had a considerably lower live birth rate. Then, preimplantation genetic screening did not increase the number of ongoing pregnancies and live births after IVF in women who were older than 35 but it significantly reduced the rates.

Until now, research has concentrated on genetic and epigenetic variants related mostly to immune response and inflammatory mediators, yielding a substantial association between recurrent miscarriage and immunological processes. Unknown causes of miscarriage may thus be caused by an immunological imbalance mediated by T-helper Th1/Th2/Th17 cytokines and regulatory T cells[71]. Overall, PGD cannot improve the success rate at the first oocyte retrieval in recurrent miscarriages with reciprocal translocations and in the natural course of events. Patients should receive accurate information regarding advantages and disadvantages. After receiving the report, couples can decide with the clinician whether to perform PGD. PGD is still a relatively new technique, and the impact of removing one or two blastomeres at the eight-cell stage on adulthood has still never been sufficiently evaluated. Such an approach should be proposed for well-selected cases only. We must identify those with difficulty in achieving successful delivery amongst recurrent miscarriages with translocations in the future.

Conclusion

Genetic variables appear to play a complex role in the efficiency of human reproduction. Classically, high rates of chromosomal errors have been among the leading etiologies for fetal loss, and more recent studies have begun to highlight the critical role that specific single gene defects may play in pregnancy maintenance. Overall, the prognosis for a patient with RPL is good, and most women with a history of RPL are less likely to miscarry in a subsequent pregnancy than to deliver a live birth. It is only after many sequential losses that this ratio reverses. To help aid couples struggling with RPL, limited and focused genetic testing is recommended as part of the diagnostic approach. PGD may indicate a small

proportion of couples with defined translocations or select single-gene disorders. Although great strides have been made to increase the accuracy and practicality of PGS for couples with RPL, such investigations are not presently indicated outside of clinical studies. Still, they hold much promise for future incorporation into the treatment of couples with RPL. Most causes of recurrent miscarriage may include abnormal chromosomes in either partner, particularly translocations, antiphospholipid antibodies, and uterine anomalies. As for embryonic factors, the frequency of normal embryonic karyotypes significantly increases with the number of previous miscarriages, and a normal karyotype in an earlier pregnancy predicts subsequent miscarriage. Previous attempts to describe genetic factors using the candidate gene approach have been relatively unsuccessful due to mammalian reproduction's physiological, cellular, and genetic complexity. Future studies in mammalian animal models will likely accelerate our understanding of the molecular mechanisms involved in recurrent pregnancy loss. They will provide additional candidate genes to screen in cases of repeated miscarriage and embryos that have genetic factors.

Future directions

In the wake of the completion of the human genome project, future preimplantation genetic testing is predicted to include affordable sequencing of individual embryonic cells for comprehensive chromosomal and single-gene disorder analysis[72]. Two studies have investigated using next-generation sequencing (NGS) for preimplantation embryo assessment. One study investigated the use of NGS to identify aneuploidy and chromosomal rearrangements, while the other used NGS in the PGD of monogenic diseases[73,74]. Both studies reported similar overall costs and increased diagnostic accuracy compared to current methods. However, additional studies with large sample sizes are needed before NGS-based preimplantation testing can be implemented in routine practice. NGS also provides a level of genetic detail that may identify congenital abnormalities that, while present, may not have been assessed for clinical relevance.

Conflicts of interest

The authors declare no conflict of interest.

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