Preimplantation genetic diagnosis (PGD) improves pregnancy outcome for translocation carriers with a history of recurrent losses

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Objective: To determine if preimplantation genetic diagnosis (PGD) for translocation carriers with three or more pregnancy losses reduces loss rates.
Design: Retrospective review of data.
Setting: Preimplantation genetic diagnosis laboratory servicing IVF groups.
Patient(s): Patients (n = 192) undergoing PGD for either a reciprocal translocation or Robertsonian translocation who had three or more previous pregnancy losses.
Intervention(s): Preimplantation genetic diagnosis for translocations.
Main Outcome Measure(s): Pregnancy loss rate, pregnancy success rate defined as delivery of at least one child or an ongoing pregnancy in the third trimester, and length of time to success.
Result(s): Pregnancy loss rate was significantly reduced to 13% post-PGD compared with 88.5% in previous non-PGD pregnancies and to 35% to 64% from naturally conceived pregnancies as reported in the literature. Pregnancy success rate was 87%. Conception occurred after an average of 1.4 cycles or <4 months.
Conclusion(s): Individuals with translocations who have experienced three or more losses benefit from PGD by realizing a significant reduction in loss rate and improvement in rate of success of pregnancy. Length of time to conceive is also dramatically reduced compared with data in the literature for similar populations not undergoing PGD. (Fertil Steril® 2010;94:283–9. ©2010 by American Society for Reproductive Medicine.)

Key Words: Preimplantation genetic diagnosis, reciprocal translocation, Robertsonian translocation, recurrent pregnancy loss, miscarriage

Individuals with translocations are known to have high rates of unbalanced gametes (1–6), have impaired or reduced gametogenesis (2, 6–10), produce high rates of unbalanced embryos (11, 12), and are therefore at risk for infertility and pregnancy loss (13). When conceiving naturally, these individuals experience loss in most pregnancies (11, 12).

The prevailing attitude among various medical specialties is that, because most of the unbalanced pregnancies will miscarry and seldom reach term (14), further interventions other than idiopathic recurrent pregnancy loss (RPL) treatments are unnecessary. However, this attitude does not take into account the pain and suffering caused by RPL. Although the ultimate goal of translocation carriers is to achieve a viable pregnancy free of chromosome abnormalities, reducing the risk of miscarriage is a parallel goal. Previous studies have shown that preimplantation genetic diagnosis (PGD) for translocations reduced loss rates from >90% to <15% (11, 12, 15–19). Additionally, studies of PGD for patients with idiopathic RPL, typically defined as three or more losses, have shown similar rates of reduction (20, 21). However, few PGD studies have looked at the subpopulation of RPL that are carriers of translocations who have had three or more losses. Recently, Otani et al. (19) undertook such a study and found a substantial reduction in miscarriages.

Other studies have focused on natural conception of carriers of structural chromosome aberrations (14, 22, 23) either because of lack of availability of PGD or preference to not prescribe such as an option to conception. Lack of availability of PGD is understandable because of country of origin and governmental restrictions thereof. However, promotion of conceiving naturally despite the availability of PGD may overlook the patient's emotional state (19).

We report on the largest dataset of PGD for chromosomal structural aberrations in Fischer et al. (in preparation). From that dataset we show here the outcome data on a subset of PGD for translocation patients who had three or more losses in previous non-PGD pregnancies.

Instead of using the typical assisted reproductive technology (ART) definition of a successful pregnancy as a sac seen

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after an IVF or other ART cycle, we use the definition of success typical to the RPL literature stated as survival past 24 weeks' gestation. This definition is more useful for this article, as we are comparing outcome to the RPL data in the literature. Additionally, comparison would be difficult with the ART definition of success as it relies on pregnancy per attempt, and one cannot assess how many times the non-ART population attempts to conceive.

MATERIALS AND METHODS

Patients

Through March 2008, PGD for structural chromosome aberrations was performed in 1,060 cycles, the largest dataset reported to date. Samples were sent from 116 IVF centers to our lab for analysis. Testing was performed for couples in which one or both members had a chromosomal structural aberration such as a translocation. A total of 683 patients underwent the 1,060 cycles.

Previous pregnancy information was known for 807 of the 1,060 cycles (76%). Of these, 192 patients undergoing PGD for either a reciprocal translocation or Robertsonian translocation had three or more previous pregnancy losses. These 192 patients were selected as the population of interest for this article. For these RPL patients, the average maternal age was 34 and the average number of previous losses was 3.8 (range: 3–7).

Preimplantation Genetic Diagnosis

Previous to IVF and PGD, patients underwent evaluations used for IVF and RPL patients such as history and physical exam, saline sonogram, hysterosalpingogram, TSH, prolactin, thyroid peroxidase antibodies, antithyroglobulin antibodies, lupus anticoagulant, anticardiolipin antibodies, antiphosphatidyl serine, beta 2 glycoprotein antibodies, fasting blood sugar, complete metabolic panel, complete blood count, hemoglobin electrophoresis, indirect Coombs, homocysteine, lupus anticoagulant, anticardiolipin, antiphosphatidyl serine, antithrombin III, Protein C, Protein S total and free, factor V and II mutation analysis, and beta 2 glycoprotein antibody testing. Chromosome analyses were performed on the spouse if not previously done.

Preimplantation genetic diagnosis via fluorescence in situ hybridization (FISH) was performed on either polar bodies or blastomeres obtained from day 3 embryos. Embryo transfer occurred on day 4 or 5 of development as determined by the timing of the reporting of the PGD results or per preference of the physician at the IVF group. The vast majority of cycles were performed by embryo biopsy. For embryo biopsy cycles, one cell per embryo at day 3 was biopsied and fixed on a glass slide using the modified Carnoy method as previously described (24). The single cell analysis was conducted following strict assessment criteria described by Munné (25). For PB biopsy cases, the same procedure described in Munné et al. (11) was followed without modification.

For reciprocal translocation cases, preliminary testing of probes was completed in blood of the carriers of the aberration to confirm the chosen probes correctly identified the finding.

Testing of polar bodies was performed via chromosome painting per methods described previously (11). Testing of blastomeres was performed via FISH on interphase chromosomes as previously described (11).

Reciprocal translocation testing on blastomeres is completed by use of commercially available probes (Vysis) to identify translocated and nontranslocated portions of the chromosomes. All of the probe strategies allowed the discrimination of normal or balanced from unbalanced cells, but not of normal from balanced. Because translocation patients produce a large number of abnormal embryos (25), it would be uncommon to have only normal embryos for replacement. Most patients are comfortable with this approach.

A minimum of three probes was used for reciprocal translocations. Typically, a combination of two probes placed distal to the breakpoints and one or two placed proximal to such are used. Robertsonian translocation testing on blastomeres is performed using enumerator probes for the chromosomes involved, and especially if the patient was 35 or older, other chromosomes such as X, Y, 8, 13, 14, 15, 16, 17, 18, 20, 21, or 22 were analyzed. The supplier's protocol with slight modifications (26) was followed for the first hybridization step. After the analysis of the first round of hybridization was completed and the slides were washed for 30 seconds, a second hybridization panel was applied. This step was repeated whenever a third round of hybridization was necessary.

The tests used in this study did not detect uniparental disomy arising from balanced Robertsonian translocations. For this and other reasons, prenatal diagnosis was recommended after PGD. Robertsonian translocation carriers were informed that if prenatal diagnosis indicated a balanced translocation, UPD testing could be performed. Robertsonian translocation patients were informed that their risk of UPD was 1% or less if the balanced translocation was present.

Informed consent for PGD was obtained from all patients, and outcome data was collected as a part of quality control procedures. Follow-up continued through pregnancy to postdelivery to obtain any prenatal and postnatal diagnoses.

This review was determined to be exempt from institutional review board (IRB) approval. According to the Western IRB in Olympia, Washington, under common rule 45 CFR 46.101(b)(4), exemptions include "research, involving the collection or study of existing data, documents, records, pathologic specimens, if these sources are publicly available or if the information is recorded by the investigator in such manner that subjects cannot be identified, directly or though identifiers linked to subjects."

Preimplantation genetic diagnosis outcome was compared with published data in the literature. These datasets are summarized in Table 1.