ORIGINAL ARTICLE: ASSISTED REPRODUCTION

Clinical outcomes in carriers of complex chromosomal rearrangements: a retrospective analysis of comprehensive chromosome screening results in seven cases

Liang Hu, M.D., Ph.D.,[a,b,c,d] Yajing Wei, B.Sc.,[a] Keli Luo, M.D., Ph.D.,[a,b,c] Pingyuan Xie, Ph.D.,[a,c,d] Fei Gong, M.D., Ph.D.,[a,b,c] Bo Xiong, Ph.D.,[c] Yueqiu Tan, Ph.D.,[a,b,c,d] Guangxiu Lu, M.D.,[a,b,c,d] and Ge Lin, M.D., Ph.D.[a,b,c,d]

[a] Institute of Reproduction and Stem Cell Engineering, Central South University; [b] Reproductive and Genetic Hospital of CITIC-XIANGYA; [c] Key Laboratory of Stem Cells and Reproductive Engineering, Ministry of Health; and [d] National Engineering and Research Center of Human Stem Cell, Changsha, People's Republic of China

Objective: To evaluate the clinical outcomes in carriers of complex chromosomal rearrangements (CCRs).
Design: Case series.
Setting: An institute for reproductive and stem cell engineering.
Patient(s): Seven couples with CCRs.
Intervention(s): Assisted reproduction with preimplantation genetic diagnosis (PGD).
Main Outcome Measure(s): PGD results, embryo rating, pregnancy outcomes.
Result(s): In cases 1, 2, 3, 4, 5, and 6, each woman underwent one cycle of PGD. Case 7 underwent two PGD cycles. We obtained 51 blastocysts from seven couples with CCR, of which 47 were eligible for biopsy; only 3 (5.9%) were normal/balanced, and 2 (3.9%) conceptions resulted. One healthy baby girl was born (the other was not yet born at the time of publication). Karyotyping revealed that the healthy baby girl was 46,XX. Although the patient with both a balanced translocation and a CCR (case 7) had 12 embryos available for biopsy, all were chromosomally unbalanced. It is interesting that 22 (57.9%) of the total 38 blastocysts were of high quality for type A CCRs, and 2 (15.4%) of the total 13 blastocysts were of high quality for type B CCR at day 6 after fertilization.
Conclusion(s): The chances of identifying normal/balanced blastocysts in patients with CCR are <6%; the chances of a pregnancy are <4%. Greater complexity CCRs result in fewer transplantable embryos. Moreover, CCRs of greater complexity have a lower rate of high quality blastocysts than CCRs of less complexity. (Fertil Steril® 2017;■:■–■. ©2017 by American Society for Reproductive Medicine.)
Key Words: Complex chromosomal rearrangement, preimplantation genetic diagnosis, comprehensive chromosome screening, next generation sequencing

Discuss: You can discuss this article with its authors and other readers at https://www.fertstertdialog.com/users/16110-fertility-and-sterility/posts/22606-24746

Received July 27, 2017; revised October 5, 2017; accepted November 16, 2017.
L.H. has nothing to disclose. Y.W. has nothing to disclose. P.X. has nothing to disclose. F.G. has nothing to disclose. B.X. has nothing to disclose. Y.T. has nothing to disclose. G.Lu. has nothing to disclose. G.Li. has nothing to disclose.
Supported by the National Nature Science Foundation of China (grant 81372627, to L.H.), the National Basic Research Program of China (grant 2012CB944901, to G.Lu and grant 2016YFC1000206, to G.Li), and the Program for New Century Excellent Talents in University (to G.Li). The funding sources had no influence on the content of this study.
Reprint requests: Ge Lin, M.D., Ph.D., Institute of Reproductive and Stem Cell Engineering, Central South University, Xiangya Road 88#, Changsha 410078, People's Republic of China (E-mail: linggf@hotmail.com).

Fertility and Sterility® Vol. ■, No. ■, ■ 2017 0015-0282/$36.00

https://doi.org/10.1016/j.fertnstert.2017.11.021

Complex chromosomal rearrangements (CCRs) are balanced or unbalanced structural aberrations involving at least three chromosomal breakpoints and chromosome segment exchanges between two or more chromosomes (1–6). They are extremely rare in the entire population (7), with a ~0.5% occurrence in newborns (8). They are classified into categories according to different criteria such as origin, the number of chromosomes, and the number of breakpoints involved (4). There were approximately 250 cases of CCR by the year 2011 (9).

For our study, we divided CCR into two types (A and B) according to the chromosomal structure of the recombination (10): type A, three-way rearrangements in which each of three chromosomes have one breakpoint and exchange the distal segment; and type B, double two-way translocations, in which two groups of balanced translocations are involved in the chromosomes of the same carrier (3, 10–13). However, the precise mechanism of CCR is unclear.

Carriers of CCRs have a high risk of phenotypic anomalies or reproductive failure such as chromosomally abnormal fetuses, infertility, or repeated miscarriages because of cryptic imbalances of the genome, gene disruption at the breakpoints, or position effect (14, 15). In terms of meiosis, a CCR can produce a large number of gametes with unbalanced chromosomes (16, 17). The majority of the balanced CCRs are carried by females (7). Most female carriers of CCRs demonstrate recurrent miscarriage or babies with an abnormal phenotype (18); male carriers of CCRs are generally infertile due to disturbances in spermatogenesis (19, 20). The application of preimplantation genetic diagnosis (PGD) for CCR carriers can potentially reduce the risk of spontaneous abortions and produce chromosomally balanced fetuses by eliminating the embryos with unbalanced chromosomes (21).

Presently, with the application of molecular biological methods such as fluorescence in situ hybridization (FISH) and array comparative genomic hybridization (array-CGH), the detection rate of CCR has increased. Madan et al. (22) analyzed CCR in 103 adults referred for reproductive problems. Scriven et al. (12) conducted an analysis of the outcome of meiotic segregation of three-way translocations in cleavage-stage embryos and determined the accuracy and limitations of PGD using FISH. However, few reports provide a systemic evaluation of PGD results and embryo quality in the two types of CCRs included in this study, so our report on a series of cases should help fill the gap in this important area.

This case series focuses on the PGD comprehensive chromosome screening results and the embryo quality of two different types of CCRs. We collected information from seven couples with one partner a CCR carrier, and we analyzed the quality of their embryos as well as the next generation sequencing (NGS)–based PGD results. Our analysis provides useful information for reproductive and genetic counseling of two different types of CCR carriers, especially for double two-way CCR carriers.

MATERIALS AND METHODS

Patients

Our case series included seven couples who underwent assisted reproduction with PGD at the Reproductive and Genetic Hospital of CITIC-Xiangya between January 1, 2012, and January 31, 2017. One partner in each couple was a CCR carrier; the study included five female carriers and two male carriers. The seven couples signed informed consent forms after undergoing adequate counseling. This case series was approved by the institutional review board, and our study was conducted in accordance with the ethical principles of the Helsinki Declaration.

For convenience, we numbered the seven couples as cases 1 to 7. The maternal age in all seven cases was <35 years. Four of the seven women (cases 1, 2, 3 and 6) had a history of abnormal pregnancy and fertility, including spontaneous abortions, induced abortions, and conceiving a child with an abnormal karyotype. The woman in case 4 had urinary calculi. The women in cases 5 and 7 had undergone a diagnostic laparoscopy before presenting to our hospital for reproductive help. The men in cases 1, 4, and 8 had liver disease.

Cytogenetic Analysis

Cytogenetic analysis was performed on cultured peripheral blood lymphocytes from the seven couples and on the 1-year-old daughter of the couple in case 3 using the standard G-banding technique. In each case, 20 metaphase GTG lymphocytes were examined with complete analysis of 7 to 8 karyotypes. The karyotypes in this report were described according to ISCN 2013.

Fluorescence In Situ Hybridization

The FISH analyses were performed for the female partner in case 3 and her 1-year-old daughter to confirm their G-banding karyotypes. The procedure was performed on metaphase spreads in accordance with the manufacturer's instructions. We used six commercial probes (Abbott Molecular) including CEP2, CEP4, CEP12, Tel 2p, Tel 4q, and Tel 12p (CEP 2 Spectrum Orange, CEP 4 Spectrum Aqua, CEP 12 Spectrum Green, TelVysion 2p Spectrum Green, TelVysion 4q Spectrum Orange, and TelVysion 12p Spectrum Green). We performed FISH in two steps: first, we used 2p, 4q, and CEP4 probes followed by the 12p, 4q, CEP2, CEP4, and CEP12 probes.

NGS-based PGD Procedure

The seven female patients, one from each case, were treated with different protocols for ovulation stimulation according to their individual situations using human chorionic gonadotropin, gonadotropin-releasing hormone, and recombinant follicle-stimulating hormone. Oocyte retrieval was performed by a skilled doctor, and the collected oocytes were fertilized by intracytoplasmic sperm injection.

Fertilized oocytes were carefully cultured using G-1 medium (Vitrolife) at 37°C in a CO_2 incubator. On the third day