ORIGINAL ARTICLE

Reproductive outcomes following preimplantation genetic diagnosis using fluorescence *in situ* hybridization for 52 translocation carrier couples with a history of recurrent pregnancy loss

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Forty-six reciprocal and six Robertsonian translocation carrier couples who experienced recurrent pregnancy loss underwent fluorescence *in situ* hybridization-based preimplantation genetic diagnosis (PGD) for the presence of the two translocated chromosomes. Out of 52 couples, 17 (33%) were undergoing infertility treatment. In total, 239 PGD cycles as oocyte retrieval (OR) were applied. The transferrable rate of negatively diagnosed embryos at the cleavage stage was 26.3%; 71 embryos were transferred as single blastocysts. The clinical pregnancy rate per transfer was 60.6%. We obtained 41 healthy live births with 3 incidences of miscarriage (7.0%). The average cumulative live birth rate was 76.9% during 4.6 OR cycles using a mild ovarian stimulation strategy. The outcomes were classified into four groups based on carrier gender and maternal age (young (<38 years) or advanced). PGD was performed for 52 couples of which the average number of OR cycles was 4.1, 2.1, 6.7 and 4.5 in young female and male carriers and female and male carriers of advanced age; the live birth rate for a primiparity was 77.8, 72.7, 66.7 and 50.0% in those groups. These results suggest that the final live birth rate might be influenced by maternal age regardless of the gender of the carrier.

Journal of Human Genetics (2016) 61, 687–692; doi:10.1038/jhg.2016.39; published online 19 May 2016

INTRODUCTION

Carriers of autosomal reciprocal translocations and Robertsonian translocations often have the opportunity to reproduce, however, these couples are known to be at increased risk for reproductive difficulties such as recurrent pregnancy loss (RPL) or phenotypically abnormal offspring owing to their production of genetically unbalanced gametes. In these carriers, the segmental affinities between the normal and translocated chromosomal regions result in quadrivalent rather than bivalent pairing at meiosis with five potential modes of segregation, and this phenomenon produces unbalanced rearrangements at high incidence.¹

The first establishment of fluorescence *in situ* hybridization (FISH)-based preimplantation genetic diagnosis (PGD) for translocation carriers was reported about 10 years after the first PGD was described by Handyside *et al.*,^{2–5} and this technique has been utilized in clinical trials for patients suffering from reproductive difficulties.^{6–20} Among these 15 studies, the reproductive outcomes of a total of 4700 carriers have been described with the pregnancy rate per embryo transfer at 22–52%, live birth rate per patient at 28–44% (average 34%) and pregnancy loss (number of spontaneous abortions per clinical pregnancy) at 0–18%.^{6–20}

Female carriers who maintain normal ovarian function equivalent to that of females with normal karyotypes are able to sustain controlled ovarian stimulation with gonadotropin.²⁰ Most reports have described that the ovarian response to controlled ovarian stimulation was not impaired by translocation status, suggesting that no differences of the segregation pattern (alternate vs unbalanced) in the PGD results or of the live birth rate between the gender and carrier groups existed.^{12–15,18,20–22} The mechanism governing whether autosomal translocation carriers experience premature ovarian failure or can maintain normal ovarian response has not yet been defined; however, most autosomal translocation carriers retain their fecundity. Therefore, all institutes that practice PGD advocate the application of standard ovarian stimulation to such carriers, representing the controlled ovarian hyperstimulation protocol utilized for regular infertility treatment in each facility. Notably, however, the reproductive outcome of RPL couples utilizing PGD is less efficient than that obtained following natural pregnancy.^{23,24} To potentially improve these outcomes, we established an original *in vitro* fertilization (IVF) protocol featuring dose reduction of gonadotropin for ovarian stimulation, which was able to decrease the incidence of ovarian hyperstimulation syndrome and multiple pregnancies.^{25–27} In

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Received 26 November 2015; revised 28 January 2016; accepted 28 March 2016; published online 19 May 2016

addition, our protocol was expected to improve reproductive outcome even in advanced maternal age couples with ovulation disorder.²⁸

In this paper, we report the clinical outcomes of FISH-based PGD using mild ovarian stimulation and single blastocyst transfer policy for 52 couples, in which one of the partners was an autosomal reciprocal translocation or Robertsonian translocation carrier. We retrospectively evaluated the live birth rate and the chromosomal imbalances of embryos derived from these RPL couples and determined how maternal age affected the reproductive outcome.

MATERIALS AND METHODS

Patients

There are strict guidelines for the performance of PGD in Japan established by the Japan Society of Obstetrics and Gynecology (JSOG). These state that patients should be given genetic counseling by a certified specialist in Medical Genetics or a genetic counselor at a clinical research center as well as from a third party institution as a second opinion, and that written consent should be required in the application. For situations involving habitual spontaneous abortion or high-risk pregnancies with serious genetic disorders, all individual cases should be evaluated by consecutive ethical committees at the supervising clinical research center and by JSOG. All couples who underwent PGD in this study were required to satisfy the following two conditions: (i) one partner was a carrier of a chromosomal abnormality; and (ii) a history of RPL, defined as two or more previous miscarriages that were attributed to either partner's chromosomal abnormality, and obstetric history, or serious genetic disorders with a prospectively high-risk pregnancy. Genetic counseling was administered to 90 PGD candidate couples who had an abnormal karyotype in either partner and written consent was obtained for this PGD clinical study with provision of an explanation of the benefits and drawbacks by our certified specialist in Medical Genetics and genetic counselor. All 90 cases were approved by the Institutional Review Board of the Kato Ladies Clinic on the first review; out of 90 couples, 88 were eventually accredited for PGD by the JSOG ethics committee during the second stage of review, and 2 cases were rejected as they were considered to be outside of the indication.

This clinical study involved the provision of PGD for the 52 accredited couples who have a reciprocal translocation ($n=46$) or Robertsonian translocation ($n=6$) in either partner, between January 2007 and December 2013 at the Kato Ladies Clinic. Of the 52 couples in the RPL group, 17 infertile patients (6 oligozoospermia, 11 fallopian tubal factor or premature ovarian insufficiency) were included. The maternal age of the 33 female carriers and 19 normal karyotype females whose husbands were carriers was 36.2 and 37.2 years, respectively; all of the 52 couples had previously experienced two or more miscarriages (range: 2–7) before 20 weeks. Only three of the couples had experience of delivery representing one case of a healthy child, one case of stillbirth and one case of neonatal death with structural imbalance (Table 1). PGD was performed for the 52 couples with RPL, which included 33% (17/52) infertile patients.

Minimal stimulation and natural cycle IVF protocol

The controlled ovarian stimulation protocol is commonly used for infertility treatments to obtain as many oocytes as possible, and is also utilized for PGD protocols; however, we have established alternate unstimulated and mild ovarian stimulation cycles for the assisted reproductive technology protocol that we apply in daily clinical practice. In this study, the unstimulated and minimal stimulation IVF protocols were performed as described previously.^{25–27} In brief, a clomiphene citrate-based minimal stimulation protocol was used in the majority of cycles (87.5%), whereas unstimulated natural (or using clomiphene citrate without gonadotropin) IVF was performed for the remaining cycles (12.5%). Clomiphene citrate (50–100 mg per day) was administered orally with an extended regimen from cycle day 3 until the day before inducing final oocyte maturation. Human menopausal gonadotropin or recombinant follicle-stimulating hormone was provided in the form of injections (50–150 IU/every other day) or human menopausal gonadotropin nasal spray for the clomiphene citrate-based stimulation protocols to obtain several matured follicles. Monitoring involving ultrasound scans and hormonal profiles (estradiol, luteinizing hormone and progesterone) was usually started on day 8 and continued every other day until the triggering day. Ovulation triggering was performed with a gonadotropin-releasing hormone agonist, busereline (Suprecur, 600mg, Sanofi K.K. Co., Ltd, Tokyo, Japan) administered in nasal spray form. In the natural cycle IVF protocol, the only pharmaceutical intervention consisted of inducing the final oocyte maturation with a gonadotropin-releasing hormone agonist. Monitoring as described above was started in the morning of day 10 and/or 12 according to the patient's menstruation cycle length. When the leading follicle reached 18 mm with a concomitant estradiol level >250 pg ml⁻¹, oocyte retrieval (OR) was scheduled. OR was usually conducted 32–35 h after triggering, but it was also performed 20–30 h after triggering for some patients when the start of the LH surge was detected.

OR, IVF and blastocyst culture

OR was practiced without anesthesia using an extra-thin 21 G needle (Kitazato BioPharma Co., Ltd., Fuji City, Japan). Conventional insemination was performed approximately 3 h after OR and intracytoplasmic sperm injection was performed as the insemination method in the presence of moderate or severe male factor infertility. Normally fertilized zygotes with two pro-nuclei, which were assessed 16–20 h after insemination, were cultured individually with Quinn's Advantage Protein Plus cleavage medium (Origio, Malov, Denmark) from day 1 to 3. All biopsied embryos at the cleavage stage were transferred to Quinn's Advantage Protein Plus blastocyst medium from day 4 to 6.

Zona breaching, embryo biopsy, fixation and FISH

Embryo biopsy was performed on the night of day 2 after OR to the morning of day 3, mechanical zona cutting was performed as previously described by Verlinsky with some modifications.^{29,30} A 2-blastomere biopsy strategy was practiced for 5–10-cell stage embryos using aspiration by a thin pipette (Kitazato BioPharma Co., Ltd., Japan). Blastomeres were fixed using the Carnoy fixative method that was described by Velilla *et al.*^{31–33} with some modifications.

Table 1 Indications for the 52 couples carrying a translocation

	Female carriers 33 couples		Male carriers 19 couples	
Average maternal age $\pm$ s.d. (range)	36.2 $\pm$ 3.9 (27–44)		37.2 $\pm$ 3.6 (28–44)	
No. of chromosomal abnormalities	TL 28	ROB 5	TL 18	ROB 1
^a Male factor infertility (No. with oligozoospermia)	1	0	4	1
Female factor infertility	7	1	3	0
Unknown infertility	20	4	11	0
Average number of previous miscarriages No. $\pm$ s.d. (range)	3.5 $\pm$ 1.4 (2–6)	2.2 $\pm$ 0.4 (2–3)	3.4 $\pm$ 1.5 (2–7)	5
IVF experience (couple)	6	3	9	1
Total birth history	3 ^b	0	0	0

Abbreviations: IVF, *in vitro* fertilization; ROB, robertsonian translocation; TL, reciprocal translocation.

^aNumber of husbands whose sperm concentration was less than 15 million per ml according to the WHO definition male infertility.

^bIncluding one case of still birth and one case of neonatal death with structural imbalance.

We chose to use the subtelomeric probes method that was described by Scriven *et al.*⁵ with some modifications: in the case of two-way translocation, two differentially labeled subtelomeric probes that were distal to the breakpoints on the translocated segments and another two subtelomeric probes that were on the opposite centric segment from the breakpoint were applied for chromosome enumeration. The subtelomeric FISH probes were purchased from GSP Lab., Inc. (Abnova, Taipei, Taiwan) and the recommended procedure was followed with some modifications. In total, four FISH probes were used to analyze the numbers of each of the two segments and two chromosomes of the two-way translocations, and FISH results were assigned as being consistent with alternate, 2:2 adjacent-1, 2:2 adjacent-2, 3:1 or 4:0 disjunction at meiosis I using the FISH scoring criteria described by Scriven *et al.*⁵ and Munne *et al.*⁶

Single vitrified-warmed blastocyst transfer

In all cases, embryos were cultured to blastocysts after being biopsied. Blastocysts that showed negative FISH results and were morphologically normal were vitrified electively for subsequent use in the transfer cycle.

Vitrification using the Cryotop vitrification kit (Kitazato BioPharma Co., Ltd.) has been described previously³⁴ and frozen-thawed embryo transfers were performed in spontaneous natural cycles on day 5 after ovulation was confined. Embryo transfer procedures were performed under vaginal ultrasound guidance using a specially designed soft catheter (Kitazato BioPharma Co., Ltd.) by placing a single embryo in a minimal volume into the mid-uterine cavity.³⁵

RESULTS

Out of 52 couples completing PGD who had a history of RPL, 33 female carriers and 19 females with normal karyotype whose husbands were translocation carriers had average maternal ages of 36.2 and 37.2 years, respectively (Table 1).

Out of 239 OR cycles using unstimulated natural cycles and mild ovarian stimulation, 174 were performed in the female carrier group

and 65 in the male carrier group; the average number of oocytes retrieved was 2.4 ± 2.1 (s.d.; range: 1–16) and 2.9 ± 2.0 (1–10), respectively. No significant differences were identified in factors including the cancellation rate, median number of cumulus oocyte complexes in successful OR cycles, 2 pronucleus (2PNs) oocytes, cleavage embryo formation rates, transferrable rate, pregnancy and live birth rate, and the miscarriage rate between female and male carrier groups (Table 2).

We also compared the reproductive outcome for obtaining primiparity between the four groups within our cohort that consisted of carrier gender and maternal age (young: <38 years; advanced: ≥ 38) as shown in Table 3. A higher cumulative live birth rate was seen in the young maternal age group: 77.8 and 72.7% in young female carriers and young normal karyotype females whose husbands were in the carrier group; in the advanced maternal age group, the rate was 66.7 and 50.0% in the advanced maternal age carrier and advanced maternal age normal karyotype groups. The distribution of the number of OR cycles needed to obtain primiparity is shown in Table 3; the average number of OR cycles in the successful group was 3.8 and 1.6 in the young female carrier and young normal karyotype female group, respectively, and 7.4 and 3.8 in the advanced maternal age carrier and advanced normal karyotype female group, respectively. In general, Robertsonian translocation carrier couples have a high risk of abortion and delivery of unbalanced karyotype with full aneuploidy for chromosome 13 or 21; however, these couples show better reproductive outcome. In the present study, six Robertsonian translocation couples were included among the 52 carrier couples and there was no Robertsonian translocation carrier couple in the young male carrier couple group (Table 3). The cumulative live birth rate in the Robertsonian carrier couple group was 66.7% (4/6), which was considered to be the same level as compared with autosomal reciprocal translocation group.

Table 2 Reproductive outcome I (segregated by carrier gender)

Factor	Total	Female carriers	Male carriers
Number of couples	52	33	19
Maternal average age	36.5 ± 3.8	36.2 ± 3.9	37.2 ± 3.6
OR cycles	239	174	65
Cycles canceled before IVF (%)	19 (7.9)	17 (9.8)	2 (3.1)
OR cycles per patient	4.6	5.3	3.4
COC	599	409	190
Median no. of COC/OR $\pm$ s.d. (range)	2.51 ± 2.1 (1–16)	2.35 ± 2.1 (1–16)	2.92 ± 2.0 (1–10)
No. of 2PNs	437	296	141
2PN formation rate (per COC; %)	73.0	72.4	74.2
Successfully biopsied	414	278	136
FISH applied	400	264	136
Diagnosed	400	264	136
Transferable (% per diagnosed)	105 (26.3)	70 (26.5)	35 (25.7)
Frozen after diagnosed (morphologically normal BLs)	82	52	30
Transferred with single blastocyst transfer	71	45	26
hCG-positive	51	32	19
Gestational sac	47 ^a	29 ^a	18
Positive FHB	44 ^a	28 ^a	16
Cases of clinical pregnancy (cases of FHB+)	43	27	16
Clinical pregnancy rate (FHB+), % per OR/% per ET	18.0/60.6	15.5/60.0	24.6/61.5
No. of deliveries	40	25	15
Live births	41 ^a	26 ^a	15
Live birth rate, % per OR/% per ET/per couple	16.7/56.3/76.9	14.4/55.6/75.8	23.1/57.7/78.9
No. of miscarriages after FHB+	3	2	1
Miscarriage rate, % per clinical pregnancy	7.0	7.4	6.3

Abbreviations: BL, blastocyst; COC, cumulus oocyte complex; FHB, fetal heartbeat; FISH, fluorescence *in situ* hybridization; hCG, human chorionic gonadotropin; IVF, *in vitro* fertilization; OR, oocyte retrieval; PN, pronucleus; SET, single embryo transfer.

^aIncluding one case of identical twins.

Table 3 Reproductive outcome II (segregated by carrier gender and maternal age for primiparity)

Carrier gender	Age (years)	Total no. of carrier	No. of ROB carrier	Total no. of successful couples	No. of ROB successful cases	Final successful rate	No. of applied PGD cycles/ couple to obtain a live birth of primiparity			Ave. PGD cycles for successful cases (ranges)	No. of unsuccessful couples	Ave. PGD cycles for unsuccessful cases (ranges)	Ave. no. of PGD cycles/total couples
							1st	2nd	3rd≤				
Female carriers													
^a Young female	32.4	18	(1)	14	(1)	77.8%	3	4	7	3.8	4	4.8	4.1
Male	34.5	—	—				—	2	—	8	(1–14) 7.4	5	
^a Advanced age female	38.9	15	(4)	10	(2)	66.7%	(1–18)						(2–17)
Male	40.9	—	—					—	—	—	—	—	
Male carriers													
^a Young female	35.0	—	—	8	—	72.7%	4	3	1	1.6	3	3.3	2.1
Male	36.6	11	(0)				(0)	—	—	—	—	—	
^a Advanced age female	39.5	—	—	4	—	50.0%	1	—	3	3.8	4	5.3	4.5
Male	42.1	8	(1)				(1)	—	—	—	—	—	

Abbreviations: FHB, fetal heartbeat; PGD, prenatal genetic diagnosis; ROB, Robertsonian translocation.
^aReproductive outcome was segregated by maternal age (<38 and $\geq$ 38) in the carrier gender group.

Table 4 Confirmation of PGD results

Factor	Present study	Scriven ¹⁶	Keymo-len ¹⁵		Rycke ¹⁹	Hirshfeld ⁸³	Franssen ³⁴
			COH	COH			
Ovarian stimulation method (or infertility treatment)	Mild stimulation/natural	COH	COH	COH	Natural conception with medical management	Natural conception	
No. of couples	52	59	142	445	109	278	
Avg. female age	37	34	33	—	30	32	
No. of OR cycles	239	113	312	946	—	—	
Total no. of COC	599	—	3895	11815	—	—	
Avg. no. of COC/cycle	2.5	13	12.5	12.5	—	—	
No. diagnosed	400	708	1553	5160	—	—	
Alternate (% /Diagnosed)	105 (26.3)	184 (26.0)	323 (20.8)	1181 (22.9)	—	—	
Embryos transferred	71	131	—	808	—	—	
No. of fetal hearts	44	36	47	209	—	—	
Patients FHB+	43	29	40	176	—	—	
Implantation rate %	62.0	27.5	—	25.9	—	—	
No. of deliveries	40	26	40	144	81	205	
No. of births	41	32	47	—	—	—	
Deliveries per OR %	16.7	23.0	12.8	15.2	—	—	
Deliveries per ET %	56.3	19.8	—	17.8	—	—	
Cumulative live birth rate per couple %	76.9	44.1	28.2	32.4	74.3	83.0	
Ave. no. of OR/couple	4.6	1.9	2.2	2.1	—	—	
Miscarriage rate %	7.0	10.3	0	4.6	55.0	48.6	

Abbreviations: COC, cumulus oocyte complex; COH, conventional ovarian stimulation; FHB, fetal heartbeat; OR, oocyte retrieval; PGD, preimplantation genetic diagnosis.

In general, the IVF success rate increases with the numbers of OR cycles until reaching a plateau. Our cumulative success data were the most effective; however, the number of OR cycles was also the highest, which was 4.6 times on average and exceeded the number obtained by another institute by approximately twofold.^{6–20} Young females whose husbands were in the young carrier group required the fewest PGD cycles: 1.6, compared with 7.4 for the advanced age female carrier group.

In addition, we presented PGD outcome measures for patients with reciprocal translocations from both single centers^{15,16} and a multiple center, which were collected by the European Society of Human Reproduction and Embryology (ESHRE) PGD Consortium¹⁹ along with data from the reproductive immunologist group from natural pregnancy^{23,36} (Table 4). Overall, 52 translocation carrier couples with RPL in the present study achieved an average 76.9% (40/52) cumulative live birth rate, which included four couples trying for

their second child with PGD. This rate is higher than that obtained in previous PGD reports.

DISCUSSION

There are two primary choices of RPL treatments for couples in which either partner is a translocation carrier: natural pregnancy with concomitant reproductive immunological management and IVF with PGD; the issue of which method should be utilized preferentially is not clear. To our knowledge, based on results from reports of PGD^{6–20} and the conservative strategy using natural pregnancy,^{23,24,36} we consider that the natural pregnancy strategy seems to have better prognosis in young reciprocal translocation carrier couples with RPL. It is difficult to understand these results; however, the patient background, biopsy invasiveness and diagnostic accuracy might influence the final outcome. Consistent with this, Ikuma *et al.*²⁴ reported a 68% cumulative live birth rate using PGD in a 31-year maternal age RPL couple group in a randomized control study, with the conclusion that PGD might perform similarly with respect to the live birth rate compared with natural pregnancy while preventing further miscarriages in the young RPL couple group. However, there are very few clinical reports that are concerned about the reproductive outcome of advanced maternal age couples wherein either partner is a translocation carrier with RPL. Keymolen *et al.*¹⁵ reported that the cumulative pregnancy rate (number of pregnancies/couple) in an advanced age (>35 years) group was 22% (4/18) and 5% (1/20) in the female and male carrier groups, respectively. In addition, another report described a cumulative pregnancy rate of 8% (1/12) in advanced maternal age (>38 year) couples.¹⁴ In the present study, we achieved 67 and 50% cumulative live births in the advanced maternal age (≥38 year) female and male carrier groups using PGD.

It is known that maternal age strongly influences IVF outcomes, and our results suggested that it also affects reproductive outcome in the chromosomal structural abnormality carrier couples. In the young maternal age group (≤37 years), the average number of PGD cycles as OR cycles to obtain about a 70% live birth rate was 4.1 and 2.1 in female carriers and normal karyotype females whose husbands were in the carrier group, respectively. Furthermore, in the advanced maternal aged group (≥38 years), 6.7 PGD cycles were required in the female carrier group, and 4.5 cycles were needed to obtain 50% successful rate in women with a normal karyotype whose husbands were in the carrier group (Table 3).

However, these results did not allow a conclusion to be drawn regarding the underlying mechanism of how translocation influences the prognosis of reproductive outcome. Some studies indicated some trend for the relationship between the type of chromosomal abnormality or carrier gender and reproductive outcome. It has been established that young carrier Robertsonian translocation couples have good prognosis not only without PGD^{36,37} but also with PGD.¹⁹ It was revealed that young male Robertsonian translocation carriers showed statistically higher¹⁷ or trends toward higher^{12,13,15,20} reproductive outcomes than did young female carriers in the PGD clinical study. However, no young male Robertsonian translocation carriers were enrolled in the present study, and the cumulative live birth rate of all Robertsonian translocation carrier couples was 66.7% (4/6), which was the same result as that of autosomal translocation couples. Furthermore, Keymolen *et al.*¹⁵ suggested that the younger male reciprocal translocation carrier group showed the best prognosis of live birth rate per couple and the worst rate was obtained in the advanced age male carrier couple group. In addition, the younger female group performed better than the advanced age female group, but this difference was small. The same trend was observed in the present study, wherein

the most effective average number of PGD cycles as oocyte retrieval was 2.1 in young normal karyotype females whose husbands were in the young carrier (avg. 36.6 years) group. The number of IVF/PGD cycles available or required represents one of the relevant factors, as Scriven *et al.*¹⁶ described that carrier couples would be afforded a 50% chance of successful live birth following three cycles of PGD with an ovarian hyperstimulation protocol. Comparatively, in the present study, 4.6 PGD cycles with mild ovarian stimulation were sufficient to obtain a 77% chance of successful live birth (Table 4).

The first report of the existence of an interchromosomal effect was written by Lejeune in 1963;³⁸ this has been a controversial topic ever since. Some patients exhibiting a higher than average rate of chromosomal abnormality have been discovered among the translocation carrier population;^{39–44} however, we consider that whether this phenomenon was caused by the concurrent translocation remains unclear. In the present study, FISH was only applied for examination of the two translocated chromosomes during the process of PGD, whereas the transferred embryos subsequently had equivalent aneuploidy risk as compared with the general embryos used in non-PGD IVF cycles. However, we obtained relatively favorable reproductive outcomes in the translocation carrier group with 37 years average maternal age, and the clinical pregnancy rate (fetal heartbeat positive; implantation rate) per single blastocyst transfer and miscarriage rate per clinical pregnancy were 62.0% and 7.0%, respectively. Out of three miscarriage cases, chromosomal analysis of the abortus was performed for two cases, identifying 45,X and trisomy 21. In comparison, in the general sub-fertile couple group at 35–37 years of maternal age treated in our IVF center, the clinical results for these rates were 45.0% and 17.5%,³⁵ respectively. Furthermore, in the case of natural conception with a maternal age of 30–40 years, the rate of early pregnancy loss was 8–15%,^{45–48} and 57–75% of chromosomal abnormality was found in the product of conception.^{49–51} Thus, we considered that interchromosomal effect might not substantively influence meiosis in our translocation carrier patient group.

In the present study, we achieved a 76.9% cumulative live birth rate with 4.6 oocyte retrieval cycles using FISH-based PGD for 52 couples with a history of RPL who had 46 reciprocal and 6 Robertsonian translocations. The average chromosomal abnormality rate of the embryos was 73.7%. The IVF procedure was performed with our original mild ovarian stimulation protocol and single blastocyst transfer policy. Using our IVF protocol for general infertile patients, the expected cumulative live birth rate is 77, 70 and 45% in <35, 36 and 39 year maternal age groups during five OR cycles.²⁸ In the present study, translocation carrier couples showed the same reproductive outcome compared with the general infertility population by excluding the unbalanced rearrangement embryos for transfer using PGD. However, considering that the number of embryos was reduced to only 1/5 to 1/3, because the average transferrable rate of reciprocal translocation carrier couples was 20–30% on a global basis^{6–20} because most embryos were eliminated by PGD as being affected with a chromosomal abnormality related to translocated chromosomes, this RPL couple group might have a better ability for reproduction compared with general infertility patients. In particular, the best group, which showed a 72.7% live birth rate for primiparity with the fewest average number of OR cycles (2.1 cycles), was that of young normal karyotype females whose husbands were in the young carrier group. These young couples might have the chance to obtain live birth with natural pregnancy if they were not adverse to a high abortion risk, which is associated with the reciprocal translocation; conversely, in the case of advanced age couples, the worthwhile way to achieve the goal of a live birth might be to use an

IVF/PGD protocol if the higher cost, time and effort required for IVF and the invasiveness of the biopsy are fully acceptable.

More studies are needed to confirm these results and to interpret the results with respect to underlying differences in the mechanisms of meiosis in spermatocytes and primary oocytes. Overall, we consider that the accumulation of these detailed reproductive outcomes of translocation carriers with or without PGD will likely contribute to an extensive discussion of genetic counseling.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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