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DIAGNÓSTICO GENÉTICO PREIMPLANTATORIO (DGP) EN PORTADORES DE REARREGLOS ESTRUCTURALES BALANCEADOS POR aCGH (ARRAY COMPARATIVE GENOMIC HYBRIDIZATION)

PREIMPLANTATIONAL GENETIC DIAGNOSIS (PGDs) IN CARRIERS OF BALANCED STRUCTURAL REARRANGEMENTS BY ARRAY COMPARATIVE GENOMIC HYBRIDIZATION (aCGH)

Ducatelli M.E.[1], A. Grazziotin Mondadori[1], F. Coco[1], N. Neuspiller[1], F.L. Gismondi[1], R. Coco[1]*

[1] Fecunditas, Instituto de Medicina Reproductiva, Afiliada a la Universidad de Medicina, UBA.

*Corresponding author: robertococo@fecunditas.com.ar

ABSTRACT

Carriers of chromosomal rearrangements have an increased risk of producing aneuploid gametes, which originate abnormal embryos, most of them lethal. Only a minority of them complete the pregnancy, delivering malformed children with unbalanced chromosomes. The majority of Preimplantational Genetic Diagnosis (PGDs) reported in these carriers were detected in blastomere using FISH. The objective of this work was to determine the risk of chromosome misbalance in blastocysts. Herein, we report 26 couples with chromosome rearrangements: five with pericentric inversions, four with Robertsonian translocations and 17 with reciprocal translocations. Trophectoderm biopsies were performed on days five or six post Intracytoplasmic Sperm Injection (ICSI). The array Comparative Genomic Hybridization (aCGH) was done with 24 Sure Plus of BlueGnome-Illumina® and the blastocyst transfer in a deferred cycle. Average of aspired oocytes was 16.2 (r= 5-46), of normal fertilized oocytes was 11.1 (r= 2-26) and of obtained blastocysts was 4.8 (r= 1-11). In the reciprocal translocation group, 63 blastocysts were biopsied, 44.4 % of them was normal and 55.6 % abnormal. In the Robertsonian translocations group, 23 blastocysts were studied, 56.5 % of them was normal and 43.5 % abnormal. In the chromosomal inversions group, 25 blastocysts were analyzed, 52 % was normal and 44 % abnormal. We could not obtain any information for one blastocyst due to a failure in DNA amplification. The results allow us to conclude that the theoretical expected risk for structural chromosome rearrangements in trophectoderm biopsy is lower than in blastomeres and/or gametes. Since not all fertilized oocytes reach the blastocyst stage, the quantity of biopsied blastocysts is much lower than in blastomere biopsy, reducing thus the costs of the PGDs.

Key words: Diagnóstico preimplantatorio, cariotipo molecular, biopsia de trofoblasto.

RESUMEN

Los portadores de rearreglos cromosómicos tienen mayor riesgo para generar gametas aneuploides, que originan embriones anormales. Sólo una minoría llega a término y origina niños malformados con cromosomas desbalanceados. La mayoría de los reportes de DGPs (Diagnóstico Genético Preimplantatorio) en portadores de rearreglos corresponden al estudio en blastómeras por FISH. El objetivo de nuestro trabajo fue determinar el riesgo de desbalance cromosómico en el estadio de blastocisto. La serie estuvo compuesta por 26 pacientes portadores de rearreglos: cinco inversiones pericéntricas, cuatro fusiones céntricas y 17 translocaciones recíprocas. La biopsia de trofectodermo fue realizada en el día cinco o seis post Inyección Intracitoplasmática del Espermatozoide (ICSI). El aCGH (array Comparative Genomic Hybridization) fue realizado con el kit 24 Sure Plus BlueGnome-Illumina® y la transferencia del blastocisto en un ciclo diferido. El promedio de ovocitos aspirados fue 16,2 (r= 5-46), el de ovocitos fecundados normales fue 11,1 (r= 2-26) y el promedio de blastocistos obtenidos fue 4,8 (r= 1-11). De los 63 blastocistos biopsiados en el grupo de translocaciones recíprocas, 44,4 % resultaron normal y 55,6 % anormal. De los 23 blastocistos de las fusiones céntricas, 56,5 % resultó normal y 43,5 % anormal. De los 25 blastocistos provenientes de las inversiones pericéntricas, 52 % resultó normal y 44% anormal. Los resultados hallados evidencian que la llegada a blastocisto disminuye sustancialmente el riesgo teórico esperado en la fecundación. Por otro lado, como no todos los ovocitos fecundados llegan a blastocisto, se reduce el costo del DGP.

Palabras clave: molecular karyotype, preimplantation diagnosis, throphectoderm biopsy.

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INTRODUCTION

It is well recognized the increased frequency of balanced structural chromosome rearrangements in human infertility. One in every 500 live newborns has a structural balanced rearrangement, such as a reciprocal translocation, a Robertsonian translocation or a pericentric or paracentric inversion (Nielsen, 1991). The carriers of these rearrangements do not express abnormal phenotypes, but the rearrangements may determine sterility and/or infertility in adulthood, due to meiotic arrest or to abnormal chromosome segregation in Anaphase I from the meiotic multivalents or loops.

In meiosis, there are five chromosome segregation alternatives from quadrivalents resulting from reciprocal translocations: alternate, adjacent 1, adjacent 2, 3:1 segregation and 4:0. Only the alternate segregation originates normal gametes, with either the two normal or the two derivative chromosomes. The other segregation types originate abnormal gametes with various chromosomal unbalances. Therefore, in carriers of reciprocal translocations, the theoretical risk of producing abnormal gametes is 80 %. The empiric risk observed in such carriers is almost equal or even higher than the theoretical 80% risk. In trivalents resulting from Robertsonian translocations, there are four segregation alternatives: alternate, adjacent 1, adjacent 2 and 3:0. Only the alternate segregation generates normal gametes, with either the two normal or the two derivative chromosomes. The other segregation types generate abnormal gametes with various chromosomal unbalances. Therefore, in carriers of Robertsonian translocations, the theoretical risk of the producing abnormal gametes is 75%. In contrast to reciprocal translocations, the empiric risk observed in such carriers is lower than the theoretical one.

On the other hand, there are three chromosome segregation alternatives from the meiotic loop of heterozygous for pericentric or paracentric inversions with symmetrical and asymmetrical crossing over: normal, non-disjunction and recombined chromosomes. Therefore, in heterozygous carriers for chromosomal inversions, the theoretical risk of the producing abnormal gametes is 66 %. The observed empiric risk is almost lower than the theoretical one (Durban, 2001; Coco, 2005).

It is important to remark the differences in the percentage of empirical chromosomal risks during gamete production, preimplantational development at cleavage or at the blastocyst, embryonic, fetal stage or newborn. The risks of abnormal gamete formation and preimplantational embryos stage are higher than the observed during pregnancy or in the newborn because most of chromosomal abnormalities are lethal. The lethality of a complete chromosome embryo aneuploidy is 99 %, most embryo aborting during preimplantation or in the postimplantation stage during the first pregnancy trimester. Instead, partial aneuploidies are less lethal than the complete ones and can result in malformed newborns. Thus, it is important that carriers of the balanced chromosomal rearrangements receive advice for the preimplantational diagnoses, even more if they need the *In Vitro* Fertilization/Intracytoplasmic Sperm Injection (IVF/ICSI) procedure to conceive.

There are three types of biopsies: (a) in polar body; (b) in cleaved embryos on day three; and (c) in trophectoderm on day five. According to the last data of the PGD ESHRE Consortium, 16.3 % of the PGDs had been performed in polar bodies, 79.8 % in D3 blastomeres and 2.3 % in D5 blastocysts. According to the last register of the PGD Consortium, from 1,213 PGDs performed in carriers of Robertsonian translocations, 72% of the cycles achieved an euploid embryo transfer with a pregnancy rate per aspiration of 21% and 30% per transfer. From 2,413 PGDs performed in reciprocal translocations carriers, 58% of the cycles achieved an euploid embryo transfer, with 14% of pregnancy rate per aspiration and 24% per transfer (Goossens, 2012).

The blastocyst is the maximum stage reached of *in vitro* preimplantational embryo development. As the cells of the trophectoderm and the mayority of the inner cell mass conform the placenta, the villus and the extraembryonary mesoderm and ectoderm, the trophectoderm biopsy is considered equivalent to the chorionic villus puncture. Therefore, both are indirect procedures whose results are not necessarily in agreement with the constitution of the future embryo-fetal-newborn, being 98 % the estimated diagnostic efficiency.

When the choice is the trophectoderm biopsy, it is preferable to perform the blastocyst transfer in a deferred cycle, due to the fact that not all the blastocysts are obtained on day five, the genetic laboratory has more time for performing the studies and, principally, because the pregnancy rate is equal or better than with the fresh embryo transfer. Nowadays, with better laboratory conditions and vitrification success, the blastocyst biopsy is the PGD best option, due to the robust results, the lower cost and the fact

that the genetic study can be programmed for working days.

Herein, we report our experience with blastocyst biopsy and deferred transfer in 26 PGD cycles in carriers of 17 reciprocal translocation, four Robertsonian translocations and five pericentric inversions, all of them evaluated by aCGH with the 24 Sure Plus BlueGnome-Illumina® platform.

PATIENTS AND METHODS

The couples accessed to PGD because of their recurrent abortion experiences and failures in IVF/ICSI procedures, the malformed stillbirths or the birth of a malformed child. The balanced rearrangements observed were:

(a) reciprocal translocations: 46,XX,t(7;11)(q11.2;q12); 45,XX,der(14)t(14;22)(q32;q11.1),-22; 46,XY,t(13;17)(q14;q23); 46,XX,t(6;10)(q13;q24); 46,XY,t(3;6)(q26;q24); 46,XX,t(9;13)(q34.3;q14.3); 46,XX,t(1;8)(q41-42;q12); 46,XX,t(6;7)(q23;q34); 46,XY,t(3;8)(p21;p11.2); 46,XX,t(9;13)(q21;q21.2); 46,XX,t(2;22)(q35;q13); 46,XY,t(6;14)(q13;q31); 46,XY,t(6;7)(q25;p15); 46,XY,t(9;11)(q32;q21); 46,XY,t(2;9)(q37;p12); 46,XX,t(4;9)(q21;p22); 46,XY,t(10;19)(q10;q10).

(b) Robertsonian translocations or centric fusions: 45,XY,rob(13;21)(q10;q10); 45,XY,rob(13;14)(q10;q10),9ph;45,XY, rob(14;21)(q10;q10); 45,XX, rob(13;14)(q10;q10).

(c) Pericentric inversions: 46,XY,inv(5)(p12q22); 46,XX,inv(5)(p14q21); 46,XY,inv(9)(p21q22); 46,XY,inv(8)(p23q11.2); 46,XY,inv(10)(p15q25.2).

The women´s average age was 36.1 years old (range: 25-46). Previous to the PGD procedure, all women were subjected to the following hormonal studies: FSH, LH, E2 y AMH, antral follicles count and the hysterosalpingography. All men underwent a complete semen analysis with bacteriological sperm culture.

The ovarian stimulation was done with recombinant gonadotrophines and GnRH antagonist or agonist. Thirty six hours post-HCG application, oocytes were retrieved by trans-vaginal ultrasonography. ICSI procedure was performed in all cases with ejaculated semen. The normal fertilized oocytes were cultured until reaching the blastocyst stage on day five or six. On day four post -ICSI the *zona pellucida* was perforated with laser shots to allow the aspiration of the hatched trophectoderm cells. After biopsy, the blastocysts were vitrified and the removed and washed trophectoderm cells were placed in Eppendorf microtubes for the molecular karyotype study. This study was performed with the BlueGnome-Illumina® 24 Sure Plus platform. Only the normal blastocysts were devitrified and transferred in a deferred cycle. The endometrium was prepared with estrogens and progesterone.

RESULTS

The average of aspired oocytes was 16.2 (5-46), the average of normal fertilized oocytes was 11.1 (2-26) and the average of reached blastocysts was 4.8 (1-11). Every couple achieved at least one blastocyst for biopsy.

In the reciprocal translocations group, 63 blastocysts were biopsied. Twenty eight (44.4 %) had normal karyotypes and 35 (55.6 %) had abnormal chromosome complements, being 15 of them (42.9%) product of a homogeneous missegregation of the meiotic quadrivalent, nine of them (25.7%) associated with aneuploidies, and 11 (31.4 %) of them with *de novo* aneuploidies unrelated to reciprocal translocations.

In the Robertsonian translocations group, 23 blastocysts were biopsied. Thirteen of them (56.5%) had normal karyotypes, and 10 (43.5 %) had abnormal karyotypes, with one of the abnormal karyotype (10%) being product of missegregation of the meiotic trivalent, one (10 %) associated with aneuploidy and eighty (80%) with *de novo* aneuploidies unrelated to the Robertsonian translocations.

In the pericentric inversion group, 25 blastocysts were biopsied. Thirteen (52%) had normal karyotypes, 11 (44%) had abnormal karyotypes and one (4%) was an amplification failure. Two of abnormal karyotypes (18.2 %) were products of a recombinant aneusomy of the meiotic loop, seven (63.6 %) with de novo aneuploidies unrelated with the rearrangement and two (18.2 %) were unbalanced segregation of a balanced complex rearrangement between a reciprocal translocation and a pericentric inversion. This case was a finding because the couple consulted for a chromosome 10 inversion but they really had a reciprocal translocation between chromosome 8 and an inverted chromosome 10. The abnormal karyotypes obtained in each group of balanced structural chromosome rearrangements (BSR) are presented in Table 1.

Twenty-six PGD cycles were performed; 20 of them were transferred with a normal devitrified blastocyst. Eight couples achieved an ongoing pregnancy, but one of them ended in a spontaneous abortion at eight weeks of gestation. In six couples, their blastocyts have not transferred yet.

Table 1. Blastocyst chromosome abnormalities of balanced structural rearrangement carriers.

Balanced Structural rearrangements (BSR)	Abnormal blastocyst karyotypes	Types of abnormal segregations	Chromosome anomalies unrelated to the BSR
	Reciprocal Translocations		
46,XY,t(3;8)(p21;p11.2)	46,XY,der(3)t(3;8)(p21;p11.2)	Adjacent 1	
	46,XY,der(3) t(3;8)(p21;p11.2)	Adjacent 1	
	46;XX,+3,-22	Seg 3:1	Monosomy 22
46,XX,t(6;10)(q13;q24)	45,X		Monosomy X
	46,XX,der(6)t(6;10)(q13;q24)	Adjacent 2	
46,XX,t(6;7)(q23;q34)	45,XY,-22		Monosomy 22
	46,XX,-7,+13	Seg 3:1	Trisomy 13
	46,XY, der(6)t(6;7)(q23;q34)	Adjacent 2	
	46,XX,-7,+22	Seg 3:1	Trisomy 22
46,XX,t(1;8)(q41-42; q12)	46,XY,der(1)t(1;8)(q41-42;q12)	Adjacent 1	
	45,XY,-8	Seg 3:1	
	46, XY, der(1)t(1;8)(q41-42;q12)	Adjacent 1	
	45,X,der(1)t(1;8)(q41;q12),del(3q10)	Adjacent 1	Partial Monosomy 3
	46,XX,der(8)t(1;8)(q41;q12),del(6)	Adjacent 1	
	45,XY,-15		Partial Monosomy 6
	46,XX,-3,+21		Monosomy 15
			Monosomy 3 Trisomy 21
46,XY,t(3;6)(q26;q24)	45,XY,-15		Monosomy 15
46,XX,t(9;13)(q21;q21.2)	46,XX,+9,-21	Seg 3:1	Monosomy 21
	47,XX,+2,-9,+13,+17,-18,+20,-21		Caotic
	46,XY,+14,-15		Trisomy 14 Monosomy15
	48,XXY,+19		Disomy X Trisomy 19

Balanced Structural rearrangements (BSR)	Abnormal blastocyst karyotypes	Types of abnormal segregations	Chromosome anomalies unrelated to the BSR
46,XY,t(6;14)(q13;q31)	46,XX,der(6) t(6;14)(q13;q31)	Adjacent 1	
	46,XX,der (6)t(6;14)(q13;q31)	Adjacent 1	
	46,XX,+19		Trisomy 19
46,XY,t(6;7)(q25;p15)	45,XX,der(7)t(6;7)(q25;p15),- 22	Adjacent 1	Monosomy 22
	46,XX,der(6)t(6;7)(q25;p15)	Adjacent 1	
	46,XY,+14,-15		Trisomy 14 Monosomy 15
	46,XX,-7,+22	Seg 3:1	Trisomy 22
46,XY, t(9;11)(q32;q21)	46,XY, der(11)t(9;11)(q32;q21)	Adjacent 1	
46,XY, t(2;9)(q37;p12)	46, XX, der(2)t(2;9)(q37;p12)	Adjacent 1	
46,XX,t(4;9)(q21;p22)	45,XX,-9	Seg 3:1	
	46,XX,+4,-22	Seg 3:1	Monosomy 22
	47,XX,+19		Trisomy 19
46,XX, t(7;11)(q11.2;q12)	46,XX,der(11) t(7;11)(q11.2;q12)	Adjacent 1	
46,XY,t(10;19)(q10;q10)	46,XX,der(10)t(10;19)(q11.21;q12)	Adjacent 1	
	Robertsonian Translocations		
45,XY,rob(13;21)(q10;q10)	45,XY,-13	Adjacent 2	
	47,XY,+3		Trisomy 3
	45,XX,-16		Monosomy 16
	47,XXY		Disomy X
45,XY,rob(13;14)(q10;q10),9ph	45,XX,-21		Monosomy 21
45,XY,rob(14;21)(q10;q10)	48,XX,+2,+9		Trisomy 2 y 9
	44,XY,-13,-22		Monosomy 13 y 22
	47,XX,+16		Trisomy 16
	46,X,+21	Adjacent 2	Monosomy X
	47,XX,-7,+15,+16		Monosomy 7
			Trisomies15, 16

Balanced Structural rearrangements (BSR)	Abnormal blastocyst karyotypes	Types of abnormal segregations	Chromosome anomalies unrelated to the BSR
	Pericentric inversions		
46,XY,inv(5)	45,XX,-16		Monosomy 16
	47,XXY		Disomy X
	46,XY,+6,-10		Monosomy 10 Trisomy 6
46,XY,inv(9)	47,XXY		Disomy X
46,XX,inv(5)	46,XX,rec(5)del5p,dup5q	Recombined	
	46,XX,rec(5)del5q	Recombined	
	48,XX,+17,+20		Trisomies 17, 20
46,XY,inv(8)	47,XX,+15		Trisomy 15
	46,XX,+16,-18		Trisomy 16 Monosomy 18
46,XY,inv(10)	46,XX,der(8)t(8;10)(q24.23;q25.1)[a]	Adjacent 1	
	46,XX,der(8)t(8;10)(q24.23;q25.1)[a]	Adjacent 1	

[a]: These abnormal unbalances allow us to infer that the true karyotype of the patient is a reciprocal translocation between a chromosome #8 and an inverted chromosome #10

DISCUSSION

Before the development of the arrays technology, the PGDs in balanced rearrangement carriers were approached by FISH or by quantitave fluorescent Polymerase Chain Reaction (qfPCR) using probes or polymorphic markers linked to the involved chromosomes.

Actually, we have the possibility of molecular karyotying, by using the qf-PCR, aCGH, simple nucleotide polymorphism arrays (aSNPs) and, ultimately, NGS or new generation sequencing (Bono, 2015), all of them better that the FISH technique because information is obtained for all chromosomes. Because of the high cost of the molecular studies it is preferable to perform it in embryos at the maximum stage of *in vitro* preimplantation development, because they have higher chances of becoming implanted in the endometrium. However, in the last report of the ESRHE PGD Consortium, it can be appreciated that only 2.3 % of the PGDs are performed in the trophectoderm with deferred cycle transfer. We decided to end with the blastomere biopsies for several reasons: the best results published with devitrified blastocyst transfer, the less invasiveness of the trophectoderm biopsy *vs.* cleavage embryos, the largest number of aspirated cells for the genetic studies, the robust results and the possibility to organize the studies in workable days. The genetic study in a single cell is difficult and becomes a real challenge for the patients and the medical team.

In the present series of 26 cycles in carriers of balanced structural rearrangements, 111 blastocysts were biopsied, 54 had normal karyotypes (48.6%), 56 were abnormal (50.4%) and for one (0.09 %) no results were obtained due to a DNA amplification failure.

In Table 1, we report the details of the abnormal karyotypes product of a homogeneous missegregation

of the meiotic multivalent and loops, an unbalanced rearrangement associated with aneuploidies and with *de novo* simple or more complex aneuploidies. The aneuploidies associated or not with the unbalanced karyotypes related to the balanced structural rearrangements could be due to an interchromosomal effect during meiosis in the carrier, or to other causes, such as the employed ICSI procedure, the suboptimal conditions of *in vitro* embryo development, or an inherent aneuploidy risk related to the maternal age; however, with the aCGH performed in this series, we could not know the origin of the aneuploides to infer a likely explanation.

In the present series we found 44.4% of transferrable blastocyst in reciprocal translocations carriers, 56.5% in Robertsonian translocations carriers and 54.2% in pericentric inversion carriers. These results agree with those reported by Xiong *et al.* (2014) and Idowu *et al.* (2015), who performed the PGDs with aSNPs.

We consider that eight pregnancies out of 20 with only one euploid blastocyst transfer is a great stimulus to continue with the trophectoderm biopsy and deferred cycle transfer. In all cases we achieved at least one blastocyst for biopsy and one euploid for transfer.

Another important fact from the economic point of view is that the genetic studies are less expensive because it is performed only in those which reach the blastocyst stage, which are transferrable. Molecular karyotyping performed in few cells is expensive. For that reason, the PGD study in potentially transferrable embryos becomes relevant, especially in countries with few economic resources. The change of studying only one cell in cleavage embryos versus several cell at the blastocyst stage is significant because results are obtained in most of the assays performed. In fact, in our series of 111 blastocyst biopsied with five to 10 removed trophectoderm cells, we had a result in almost all of them, except in one due to an amplification failure. It is an important difference with the biopsy of a single blastomere in day three, in which the assays without results varied between 10 and 15 %.

A PGD program with fresh transfer requires that the molecular geneticist team works all the day, every day, without free days. Instead, a PGD program with deferred transfer is more organized for the genetic laboratory and, if there were doubts about some of the results, the studies can be either completed or repeated. Actually, there are no doubts that the pregnancy rate in cases of deferred cycle transfer is better than in the fresh transfer, not only due to the pregnancy rate but also because of the better obstetric and perinatal results that can be obtained. These better results probably are due to the embryo-endometrium synchronization and the better endometrial receptivity in a non-stimulated cycle. In fact, it is known that the high concentration of estrogens in a stimulated cycle disturbs and damages the endometrial receptivity (Haouzi, 2010).

BIBLIOGRAPHY

Bono S., A. Biricik, L. Spizzichino, A. Nuccitelli, M.G. Minasi, E. Greco, F. Spinella, F. Fiorentino (2015) Validation of a semiconductor next-generation sequencing-based protocol for preimplantation genetic diagnosis of reciprocal translocations. Prenatal Diagnosis 35: 938-944.

Coco R., F. Coco Ludueña, M. Urquiza, J. Mincman, A. Gallo, N. Neuspiller (2005) Riesgo genético reproductivo en portadores de rearreglos cromosómicos. Reproducción 20 (1): 25-36.

Coco R., A. Mondadori, M.E. Ducatelli, J. Mincman, A. Gallo, F. Coco, S. Neuspiller, F.L. Gismondi, N. Neuspiller (2012) Preimplantation diagnosis in blastocyst biopsy and deferred cycle transfer. JBRA Assist Reprod. 16 (5): 268-270.

Durban M., J. Benet, M. Boada, E. Fernandez, J.M. Calafell, J.M. Lailla (2001) PGD in female carriers of balanced Robertsonian and reciprocal translocations by first polar body analysis. Hum. Reprod. Update 7 (6): 591-602.

Goossens V., J. Traeger-Synodinos, E. Coonen, M. De Rycke, C. Moutou, T. Pehlivan, I.A.P. Derks-Smeets, G. Harton (2012) ESHRE PGD Consortium data collection XI: cycles from January to December 2008 with pregnancy follow-up to October 2009. Hum. Reprod. 27 (7): 1887-1911.

Haouzi D., S. Assou, C. Dechanet, T. Anahory, H. Dechaud, J. De Vos, S. Hamamah (2010) Controlled ovarian hyperstimulation for *In Vitro* Fertilization alters endometrial receptivity in Humans: Protocol effects. Biol. Reprod. 82 (4): 679-686.

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Idowu D., K. Merrion, N. Wemmer, J. Gessner Mash, B. Pettersen, D. Kijacic, R. Lathi (2015) Pregnancy outcome followinf 24-Chromosome preimplantation genetic diagnosis in couple with balanced reciprocal or Robertsonian translocation. Fertility and Sterility 103: 1037-42.

Nielsen J., M. Wohlert (1991) Chromosome abnormalities found among 34,910 newborn children: results from a 13-year incidence study in Arhus. Denmark. Hum. Genet. 87 (1): 81-3.

Xiong B., K. Tan, Y.Q. Tan, F. Gong, S.P. Zhang, C.F. Lu, K.L. Luo, G.X. Lu, G. Lin (2014) Using SNP array to identify aneuploidy and segmental imbalance in traslocation carriers. Genomics Data 2: 92-95.