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Case Report

Prenatal diagnosis of an SRY-negative XXY female fetus with karyotype of 47,XX,idic(Y)(p11.2)

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Abstract: The *SRY* gene located on the short arm of the Y chromosome at band Yp11.3 is crucial for the sex determination. In this case, we report an SRY-negative XXY female fetus with karyotype 47,XX,idic(Y)(p11.2) which has not been reported before to our knowledge. A pregnant woman presented to our department for amniocentesis because the noninvasive prenatal testing pointed out abnormal screening results with sex chromosomes. The karyotyping result showed 47,XX,idic(Y)(p11.2). The FISH, PCR and array-CGH analysis showed that there were duplications of Y chromosome from Yp11.2-q21 and deletions of the *SRY* gene and pseudoautosomal regions (PAR1). The parents decided to undergo termination of pregnancy. The external genitalia of the aborted fetus were similar to female with clitorimegaly.

Keyword: SRY idic(Y)

Introduction

Klinefelter Syndrome (KS), first described in 1942 as an endocrine disorder, is the most common disorder of sex chromosomes in humans with a prevalence of 1/500-600 in male infants [1]. A majority of KS patients (~80%) show a karyotype of 47,XXY. Apart from that, other karyotypes have been reported, including 46,XX in males, 47,XXY in females, 47,XX,der(Y), 47,X,der(X), Y, 48,XXXY, 48,XXYY, and 49,XXXXY [2]. Most of the patients with KS are males, however, rare female cases are available as the *SRY* gene located on the short arm of Y chromosome at band Yp11.3 is deleted. To date, only fourteen 47,XXY female have been reported in PubMed database [3-5]. In this case, we present a case of SRY-negative XXY female fetus with de novo isodicentric Yp11.2 and deletions of the *SRY* gene and pseudoautosomal regions by conventional cytogenetics, molecular testing and anatomy and pathology detection.

Case report

Patient

The patient was a 41-year-old G_2P_1 woman, with a high risk in the second-trimester Down syndrome serum screening. Despite no abnormalities in the fetal ultrasound scan, the patient was advised to receive further prenatal diagnosis. Finally, noninvasive prenatal testing (NIPT) was chosen, which revealed abnormality of sex chromosomes. Amniocentesis was performed at the 21 week of pregnancy.

Cytogenetic analysis

The chromosome preparation from amniotic fluid cells was performed according to the previous description [Preparation, culture, and analysis of amniotic fluid samples]. The chromosomes were analyzed with GTG-banding. The karyotypes were described based on the International System for Human Cytogenetic Nomenclature (ISCN2009) [6]. Additionally, the karyotypes of the parents were investigated.

Molecular analysis

Fluorescence in situ hybridization (FISH) analysis was performed on amniotic fluid cells to confirm the G-banding chromosome analysis. FISH was performed using centromeric probes for chromosomes 18/X/Y purchased from GP Medical Technologies (Beijing, China), accord-

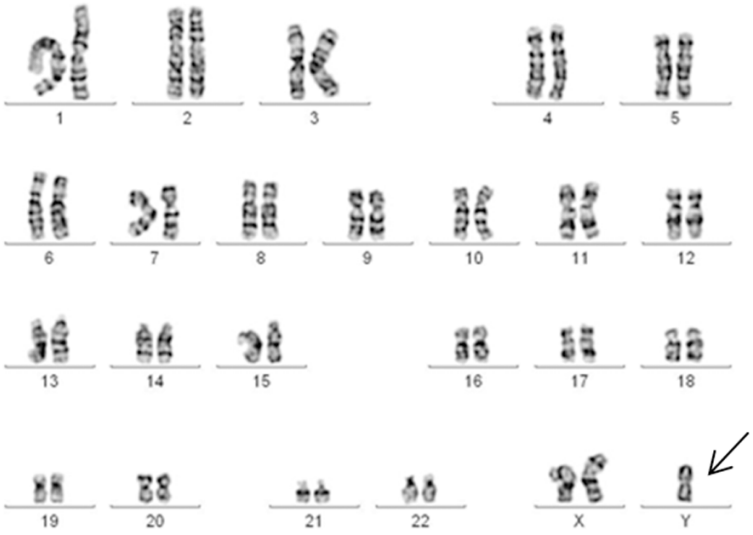

Figure 1. The karyotype 47,XX,idic(Y)(p11.2) of amniotic fluid cells, the arrow points to the idic(Y).

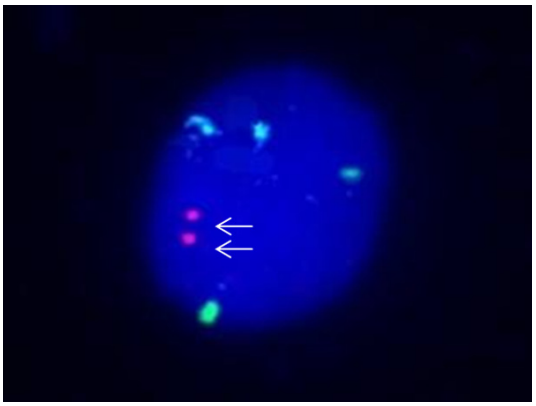

Figure 2. FISH analysis using CEP18/X/Y probes confirms the two normal X (green) and two close but clear distinct Y (red) chromosomes in interphase cells, the arrow points to the idic(Y).

ing to the manufacturer's protocols (CSP X, a green probe specific for Xp11.1-q11.1, and CSP Y, a red probe specific for Yp11.1-q11.1). After hybridization, the chromosomes were counterstained with DAPI. Images were observed using a Zeiss Axiophot fluorescent microscope under a magnification of 100×. Digital images were captured and analyzed using FISH 2.0 software (Video Test).

For PCR and array-CGH investigations, genomic DNA was extracted from the umbilical cord blood samples using H.Q.&.Q blood DNA kit (U-Gene Biotechnology CO., LTD, Anhui, China) according to the manufacturer's instructions. Multiplex PCR amplification was performed to identify Y chromosome microdeletions in the AZF regions with Y chromosome specific STS markers (SRY; AZFa: sY86, sY84; AZFb: sY127, sY134; AZFc: sY254, sY255) using Microdeletion Y Kit (Tellgen Life Science Co., Ltd, Shanghai, China) according to the manufacturer's instructions. The amplification was performed in aGeneAmp PCR System 9600 (Perkin Elmer, USA). The protocols for the PCR were as follows: initial denaturation at 95°C for 3 min and 35 cycles consisting of denaturation 95°C for 30 sec, annealing at 59°C for 30 sec and extension at 72°C for 30 sec. The final extension step was 72°C for 10 min. The *SRY* gene from the normal male and female was used as positive and negative controls. Array-CGH with a 30 kb resolution (SureScan Agilent USA) was done to the DNA sample of umbilical cord blood cells and was performed according to the manufacture's recommendations.

Results

Cytogenetic analysis

Chromosome analysis showed an abnormal karyotype of 47,XX,der(Y;Y) with an additional X chromosome and a structurally abnormal Y chromosome in all metaphase (**Figure 1**). The karyotypes of the patient and her husband were 46, XX and 46, XY.

Molecular analysis

FISH analysis showed the centromeres of two normal X chromosomes and two distinct Y chromosomes in cells (**Figure 2**), confirming the results of the chromosome analysis.

PCR analysis for the Y chromosome indicated that the *SRY* region was lacked while the AZF regions were available (**Figure 3**). Array-CGH results showed that a 3.31Mb copy number gain of chromosome Yp11.2 at 6,628,

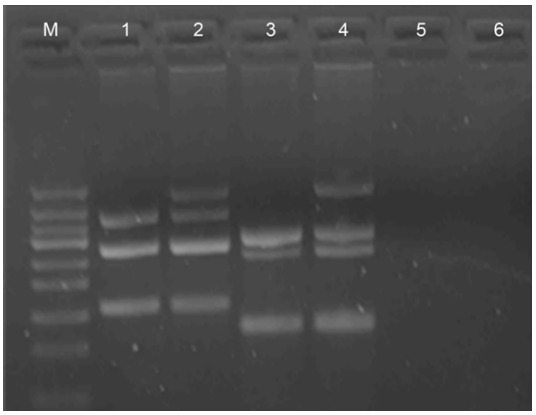

Figure 3. PCR analysis. Lane 1 and 3: the fetus lack of SRY gene but the AZF regions exist; Lane 2 and 4: normal male control; Line 5: negative control; Lane 6: normal female control.

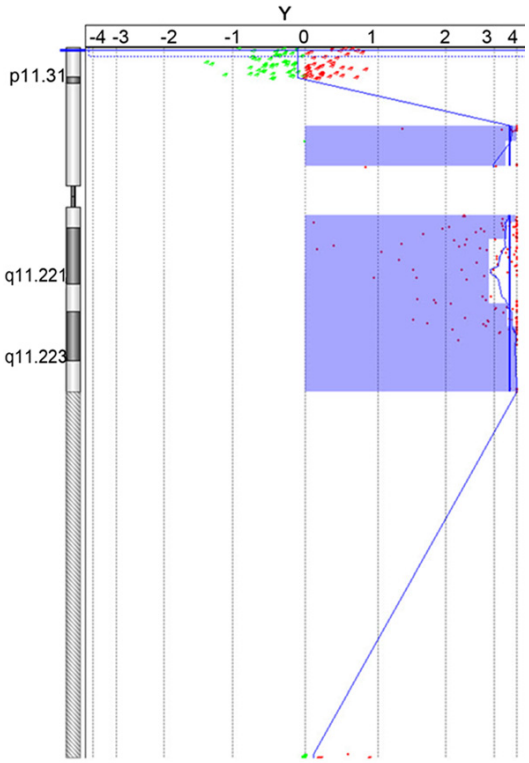

Figure 4. Array-CGH of the idic(Y) showing duplication of Y chromosome from Yp11.2-q21.

691-9,940,478 nt, a 0.55Mb copy number gain of chromosome Yq11.21 at 14,061,053-14,611,143 nt, a 14.16Mb copy number gain of chromosome Yq11.21-11.23 at 14,611,203-28,767,604 nt, a 5.42Mb copy number gain of chromosome Yq11.221-11.222 at 16,017,052-21,441,503 nt, a 5.41Mb copy number gain of chromosome Yq11.223-11.23 at 23,354,523-28,767,604 nt, as well as a 0.26Mb copy number gain of chromosome Yq12 at 59,031,421-59,293,089 nt (**Figure 4**). The findings were consistent with the results obtained from chromosome analysis, showing that this cytogenetic aberration results in two copies of Yp11.2-q12.

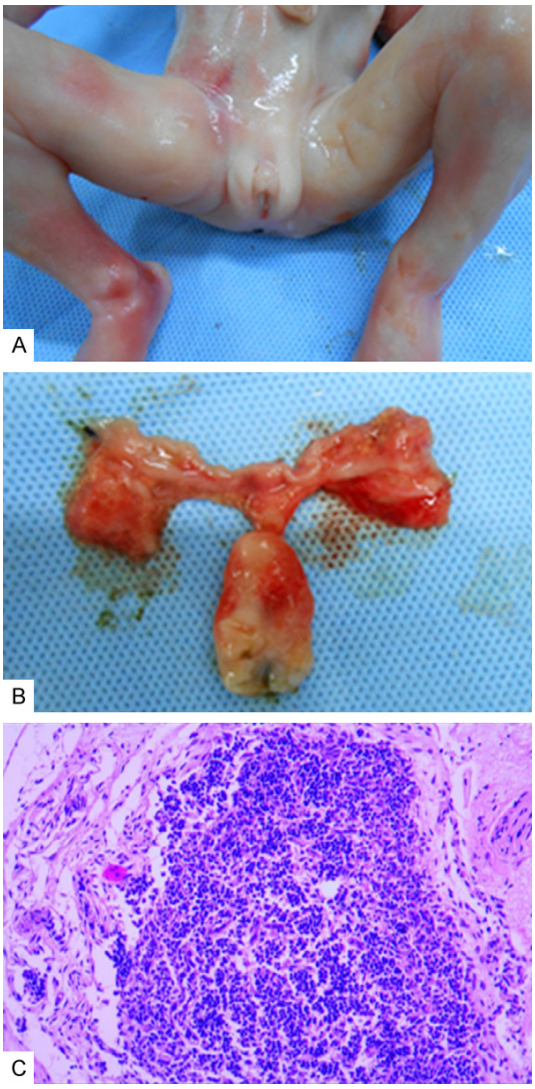

Figure 5. A: The external genitalia of the aborted fetus were female. B and C: The aborted fetus had normal internal genitalia with a uterus and two gonads which were ovaries showing abundant oocytes.

Based on the results of cytogenetic analysis, FISH, PCR and array-CGH, the karyotype of the fetus was 47,XX,idic(Y)(p11.2).

After genetic counseling, the couple decided to terminate the pregnancy. Pathologic exami-

nation of the induced fetus showed a female fetus with clitorimegaly. The fetus (a gestation age of approximately 24 weeks) weighted 498 g with a height of 25 cm. Vigina, uterus, two fallopian tubes and two gonads were indentified (**Figure 5**).

Discussion

Rare cases of KS are reported in female. In our study, we present a female fetus with a karyotype of 47,XX,idic(Y)(p11.2) having a derivative Y chromosome consisted of two Y chromosomes. FISH analysis testified two Y chromosome centromeric signals in the derivative Y chromosome. PCR amplification showed no *SRY* gene in the fetus, but the AZFs were available. Array-CGH showed that the duplicated parts of Y chromosome were derived from Yp11.2-q12 exclusive of the region of *SRY* gene. However, the autopsy and the pathology examination displayed that it was a female fetus with female external and internal genitalia. Taken together, the karyotype 47,XX,idic(Y)(p11.2) identified in this study is a rare kind of KS with structure abnormality of Y chromosome which has never been reported before.

To date, structural rearrangements involving the Y chromosome have been frequently reported, among which dicentric Y chromosome is the most common type [7-11]. As idic(Y) chromosome is unstable, the fate of such chromosome is to break down. Heinrita et al [7] reported a patient with karyotype 47,XX,idic(Y)(q12) which the breakpoints in the idic(Y) chromosome occur in the long arms of the Y chromosome and the *SRY* gene was present. If the breakpoints are in the short arms of the Y chromosome and the *SRY* gene is deleted, a female phenotype can be expected. In this case, the derivative Y chromosome was derived from two broken Y chromosomes jointed together at the breakpoint of Yp11.2 with the loss of *SRY* gene. As is known to all, *SRY* gene specifically controls male sexual development by encoding for the gene product the testis determining factor, which promotes the undifferentiated gonadal tissue of the embryo to form the male testes.

In conclusion, a rare case of KS with non-mosaic isodicentric Y chromosomes together with loss of *SRY* gene is present in this study. Our study contributes to the clinical genetic counselling and prenatal diagnosis for such disease.

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Disclosure of conflict of interest

None.

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References

[1] Song SH, Won HJ, Yoon TK, Cha DH, Shim JY and Shim SH. A case of the rare variant of Klinefelter syndrome 47, XY, i (X)(q10). Clin Exp Reprod Med 2013; 40: 174-176.

[2] Lanfranco F, Kamischke A, Zitzmann M and Nieschlag E. Klinefelter's syndrome. Lancet 2004; 364: 273-283.

[3] Lin B, Tan F, Xu H, Wang P, Tang Q, Zhu Y, Kong X and Hu L. De novo Structure Variations of the Y Chromosome in a 47, XXY Female with Ovarian Failure: A Case Report. Cytogenet Genome Res 2014; 143: 221-224.

[4] Khandelwal A, Agarwal A and Jiloha R. A 47, XXY female with gender identity disorder. Arch Sex Behav 2010; 39: 1021-1023.

[5] Frühmesser A and Kotzot D. Chromosomal variants in Klinefelter syndrome. Sex Dev 2011; 5: 109-123.

[6] Shaffer LG, Slovak ML and Campbell LJ. An International System for Human Cytogenetic Nomenclature. Karger, Basel: International Standing Committee on Human Cytogenetic Nomenclature; 2009.

[7] Heinritz W, Kotzot D, Heinze S, Kujat A, Kleemann WJ and Froster UG. Molecular and cytogenetic characterization of a non-mosaic isodicentric Y chromosome in a patient with Klinefelter syndrome. Am J Med Genet A 2005; 132: 198-201.

[8] DesGroseilliers M, Beaulieu Bergeron M, Brochu P, Lemyre E and Lemieux N. Phenotypic variability in isodicentric Y patients: study of nine cases. Clin Genet 2006; 70: 145-150.

[9] Iourov I, Vorsanova S, Liehr T, Monakhov V, Soloviev I and Yurov Y. Dynamic mosaicism manifesting as loss, gain and rearrangement

of an isodicentric Y chromosome in a male child with growth retardation and abnormal external genitalia. Cytogenet Genome Res 2008; 121: 302-306.

[10] Lange J, Skaletsky H, van Daalen SK, Embry SL, Korver CM, Brown LG, Oates RD, Silber S, Repping S and Page DC. Isodicentric Y chromosomes and sex disorders as byproducts of homologous recombination that maintains palindromes. Cell 2009; 138: 855-869.

[11] Pasantes J, Wimmer R, Knebel S, Münch C, Kelbova C, Junge A, Kieback P, Küpferling P and Schempp W. 47, X, idic (Y), inv dup (Y): a non-mosaic case of a phenotypically normal boy with two different Y isochromosomes and neocentromere formation. Cytogenet Genome Res 2012; 136: 157-162.