Clin Pediatr Endocrinol 2015; 24(1), 11–14

Original Article

A 45,X/46,XY DSD (Disorder of Sexual Development) case with an extremely uneven distribution of 46,XY cells between lymphocytes and gonads

Risa Nomura[1], Kentaro Miyai[1], Michiyo Okada[2], Michiko Kajiwara[1], Makoto Ono[1], Tsutomu Ogata[2, 3], Iichiro Onishi[4], Mana Sato[4], Masaki Sekine[4], Takumi Akashi[4], Shuki Mizutani[1], and Kenichi Kashimada[1]

[1]*Department of Pediatrics and Developmental Biology, Graduate School, Tokyo Medical and Dental University, Tokyo, Japan*

[2]*Department of Endocrinology and Metabolism, National Research Institute for Child Health and Development, Tokyo, Japan*

[3]*Department of Pediatrics, Hamamatsu University School of Medicine, Hamamatsu, Japan*

[4]*Department of Pathology, Graduate School, Tokyo Medical and Dental University, Tokyo Japan*

Abstract. In 45,X/46,XY DSDs, the proportion of the two cell lineages is uneven in different organs and tissues, and 45,X and 46,XY cells can be found throughout the body. The gonadal development of 45,X/46,XY patients depends on the population of 46,XY cells in the gonads and the clinical features are variable. We had a 45,X/46,XY DSD patient whose 46,XY population in peripheral blood was extremely low, less than 0.2%, and was not detected by FISH analysis. However, the patient showed bilateral testicular development and more than 50% of the cells in the gonads had the 46,XY karyotype. This case suggests that a drastically imbalanced distribution could occur in 45,X/46,XY DSD cases.

Key words: 45,X/46,XY DSD (disorder of sexual development), mosaicism, chimerism, SRY, FISH (fluorescence *in situ* hybridization)

Introduction

The clinical features of 45,X/46,XY patients are variable, and the typically include, Turner-like syndrome with bilateral streak gonads with Müllerian structures and Turner Syndrome stigmata, mixed gonadal dysgenesis (MGD) with unilateral testis and a streak gonad, male gender with bilateral testes (any combination of dysgenetic or normal testes) or, as is usually the case, male gender with bilateral testes and normal male genitalia (1–3). Generally, the proportion of the two cell lineages is uneven in different organs and tissues (3), and 45,X and 46,XY cells can be found throughout the body.

We had a 45,X/46,XY DSD (disorder of sexual development) patient whose 46,XY population was extremely low in peripheral blood. This case suggests that a drastically imbalanced distribution could occur in 45,X/46,XY cases, and

Received: April 21, 2014
Accepted: August 28, 2014
Corresponding Author: Dr. Kenichi Kashimada, Department of Pediatrics and Developmental Biology, Graduate School, Tokyo Medical and Dental University, 1-5-45 Yushima, Bunkyo-ku, Tokyo 113-8510, Japan
E-mail: kkashimada.ped@tmd.ac.jp

the lessons from the case may provide valuable information for the clinical management of 45,X/46,XY DSD cases.

Case Presentation

Written informed consent was obtained from the parents of the patient, in accordance with the Institutional Review Board of Tokyo Medical and Dental University. The case was conceived through IVF using a sperm donor due to azoospermia of the patient's father and was born at 37 wk of gestational age with a birth wt of 2248 g. Twin pregnancy was not documented during prenatal examinations. The external genitalia of the patient were atypical with clitoromegaly and partial fusion of the labia. The vagina and urethra opened separately (Fig. 1). The right and left gonads were palpable in the scrotum and in the inguinal canal, respectively. No organs derived from Müllerian ducts were identified by abdominal MRI scan, and the peak level of testosterone in the human chorionic gonadotropin (hCG) test was within the normal range for male infants (peak level of testosterone: 8.5 ng/ml), suggesting the gonads had testicular function. G banding chromosomal analysis using peripheral lymphocytes suggested a 45,X karyotype without the 46,XY lineage, and this finding was supported by fluorescence *in situ* hybridization (FISH) analysis using 500 peripheral lymphocytes (Fig. 2A). FISH analysis by using other cells, such as buccal cells or fibroblasts, was not performed. No marker chromosomes were identified. In contrast to the results of G banding and FISH analyses, *SRY* and a long arm region of the Y chromosome, *DYZ1,* were detected by PCR from peripheral lymphocytes (Fig. 2B). Based on these findings, we concluded that the patient had the 46,XY lineage; however, the population of 46,XY cells was extremely low.

After careful discussions with urologists and the parents, we determined the rearing sex of the patient to be female. Genitoplasty and left gonadectomy were performed at the age of one yr, and right gonadectomy was performed at the age of ten yr. The diameters of the left and right testes were 5 mm and 28 mm respectively. In spite of absence of germ cells, the epididymis, vas deferens and seminal vesicle were observed in both testes. Sertoli cells (SOX9 positive cells) and immature Leydig cells were observed (Fig. 3A). Ovarian tissue or cells (FOXL2 positive cells) were not found (Fig. 3B). During the routine examination for surgical treatment, her ABO blood type was revealed to be atypical ABO blood type, A1B3.

Histological FISH analyses of the right gonad revealed 73% of cells in the gonad had the Y chromosome (Fig. 3C), suggesting that the karyotype of the patient was 45,X/46,XY with extreme discordance of the 46,XY population between the lymphocytes and gonads.

She showed intellectual disability and Turner stigmata, such as a webbed neck, broad chest and short stature (less than –2SD). She received GH therapy from ten yr of age and estrogen therapy from fifteen yr of age. Her adult height was 147 cm (–2.09 SD).

Discussion

From the viewpoint of the uneven distribution of the 46,XY lineage, our case is remarkable. In 45,X/46,XY testicular DSD cases (classically called male "pseudohermaphroditism: MPH"), it has been reported that there is no correlation between the proportion of the 45,X/46,XY cell lineages in the blood and in the gonads; however, based on previous reports that have described testicular DSDs with the 45,X/46XY karyotype, the 46,XY lineage was detected in peripheral blood by cytological analyses, e.g., G banding or FISH (2, 4, 5). To date, there has been a case report of a 45,X patient whose gonads developed as testes, whereas, in this case, PCR analyses detected only SRY but no other genes of the Y chromosome, suggesting that the case had autosomal translocation of SRY instead of the 45,X/46,XY karyotype (6).

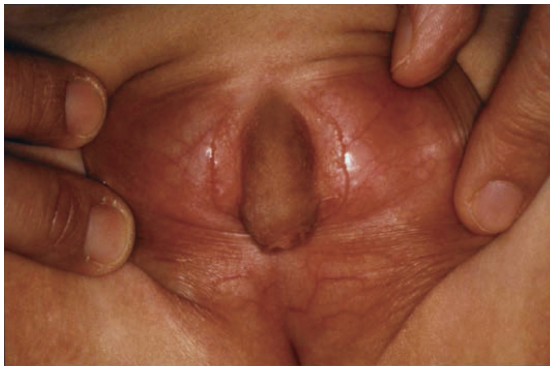

Fig. 1. External genitalia of the patient.

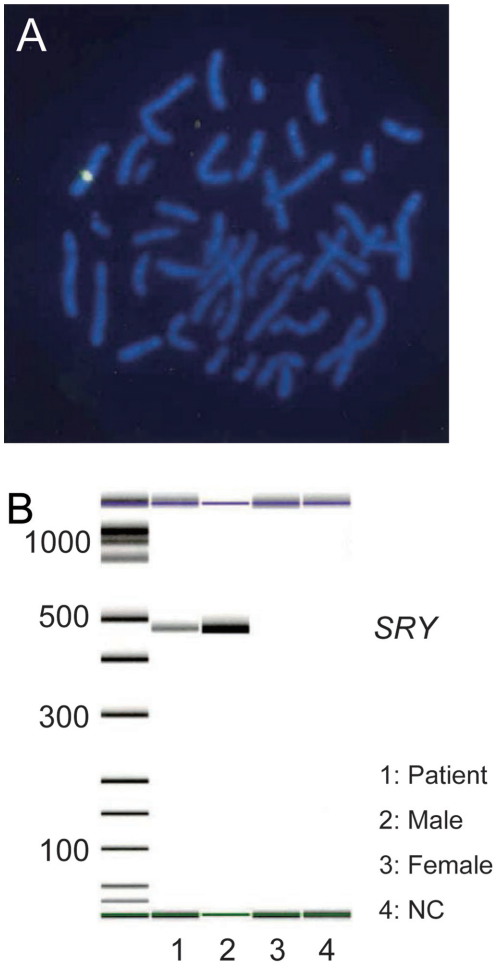

Fig. 2. A: FISH analysis of the peripheral lymphocytes. The X and Y chromosomes were labeled with green and red, respectively. The patient had a single green-labeled chromosome, indicating the 45, X karyotype. B: PCR analysis of *SRY* in peripheral lymphocytes.

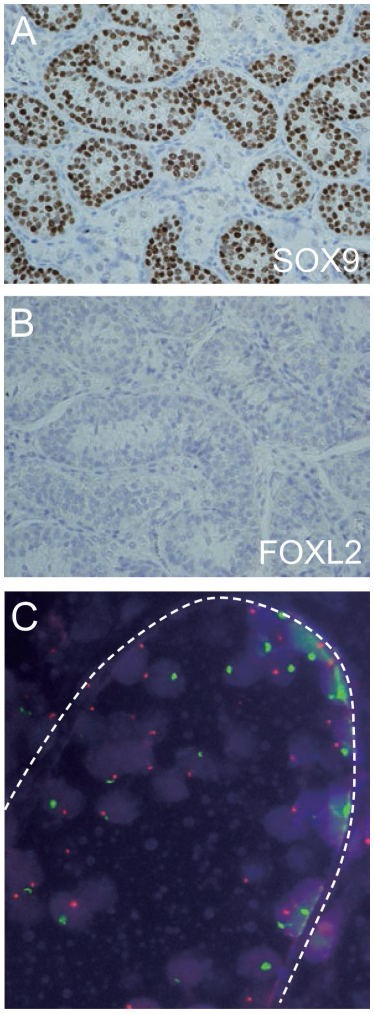

Fig. 3. A and B: Immunohistochemical analysis of the right gonad. Sertoli cells, SOX9-positive cells, were observed in the seminiferous tubules (A), and there were no ovarian cells stained by a granulosa cells marker, FOXL2 (B). C: FISH using probes for the X (red) and Y (green) chromosomes showed that most of the Sertoli cells had the 46,XY karyotype. The number of Y-positive cells was counted and averaged in XX fields under a light microscope at ×40 magnification, and representative fields were photographed. A white dotted line indicates a seminiferous tubule.

Generally, 45,X/46,XY DSDs are considered to be mosaicisms due to the loss by nondisjunction of the Y chromosome after normal disomic fertilization. Because this case was conceived

through IVF and had an atypical ABO blood type, A1B3, we considered the possibility of chimerism, instead of mosaicism. IVF has been suggested to increase the risk of rare twin-associated anomalies and chimerism (7, 8). Further, one of the major explanations for a rare atypical blood type, such as A1B3, is chimerism (9, 10). In the present case, however, the lack of DNA samples for the biological father prevented efficient genetic analysis to prove the existence of chimerism.

Although we were not able to identify the precise mechanisms of the uneven distribution of the two cell lineages, our case might provide valuable insight for the approach to 45,X/46,XY DSD cases. In particular, the possibility of a drastically imbalanced distribution of 45, X/46, XY cells should be considered in DSD cases, and careful analyses and evaluation of the karyotype are important.

References

1. Knudtzon J, Aarskog D. 45,X/46,XY mosaicism. A clinical review and report of ten cases. Eur J Pediatr 1987;146: 266–71. [Medline] [CrossRef]
2. Chang HJ, Clark RD, Bachman H. The phenotype of 45,X/46,XY mosaicism: an analysis of 92 prenatally diagnosed cases. Am J Hum Genet 1990;46: 156–67. [Medline]
3. Telvi L, Lebbar A, Del Pino O, Barbet JP, Chaussain JL. 45,X/46,XY mosaicism: report of 27 cases. Pediatrics 1999;104: 304–8. [Medline] [CrossRef]
4. Tosson H, Rose SR, Gartner LA. Description of children with 45,X/46,XY karyotype. Eur J Pediatr 2012;171: 521–9. [Medline] [CrossRef]
5. Hersmus R, Stoop H, Turbitt E, Oosterhuis JW, Drop SLS, Sinclair AH, *et al*. *SRY* mutation analysis by next generation (deep) sequencing in a cohort of chromosomal Disorders of Sex Development (DSD) patients with a mosaic karyotype. BMC Med Genet 2012;13: 108. [Medline] [CrossRef]
6. Copelli S, Castiñeyra G, Levalle O, Aszpis S, Mormandi E, Targovnik H. PCR analysis of Y-chromosome sequences in a 45,X male patient and a review of the literature. Arch Androl 2000;44: 137–45. [Medline] [CrossRef]
7. Strain L, Dean JC, Hamilton MP, Bonthron DT. A true hermaphrodite chimera resulting from embryo amalgamation after *in vitro* fertilization. N Engl J Med 1998;338: 166–9. [Medline] [CrossRef]
8. Simon-Bouy B, Plachot M, Mokdad A, Lavaud N, Muti C, Bazin A, *et al*. Possible human chimera detected prenatally after *in vitro* fertilization: a case report. Prenat Diagn 2003;23: 935–7. [Medline] [CrossRef]
9. Tippett P. Blood group chimeras. A review. Vox Sang 1983;44: 333–59. [Medline] [CrossRef]
10. Cho D, Lee SK, Yazer MH, Shin MG, Shin JH, Suh SP, *et al*. A dispermic chimera with mixed field blood group B and mosaic 46,XY/47,XYY karyotype. J Korean Med Sci 2007;22: 553–6. [Medline] [CrossRef]