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CASE REPORT

Cytogenetic Analysis of a Patient with Turner Syndrome (45,X/47,XXX/48,XXXX/46,XX)

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SUMMARY

Background: **Turner syndrome (TS) is a female genetic disorder. Most patients with TS have a 45,X haplotype, but a small proportion have low nonholic chimerism. We here report a rare case of chimeric Turner syndrome in an individual with no phenotype aside from difficulties in conception, which may have been due to TS-associated decreased ovarian function.**
Methods: **A 41-year-old female presented with no family history of TS, normal facial build, normal intelligence, and no other common clinical features of TS. The patient experienced spontaneous puberty, regular menstruation of a normal volume, bilateral fallopian tube blockage, and multiple cervical cysts.**
Results: **Karyotype analysis showed 45,X/47,XXX/46,XX cells, whereas fluorescence in situ hybridization also revealed the presence of 48,XXX cells.**
Conclusions: **There is growing evidence that ovarian function declines with age among those with chimeric TS, reducing their chances of conception. Fluorescence in situ hybridization should be recommended among those with difficulties conceiving to detect those with atypical chimeric TS, who may experience ovarian failure at an early age, to enable timely fertility interventions.**
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KEYWORDS

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INTRODUCTION

Turner syndrome (TS) is a common female chromosomal disorder that occurs at a rate of approximately one in 2,500 live female births. TS patients are typically characterized by short stature, gonadal hypoplasia, neck webbing, and elbow eversion. In addition to kidney and heart disorders [1], the incidence of ovarian insufficiency and primary amenorrhea is also higher among TS patients than in the general population [2,3], making spontaneous pregnancy unlikely. When spontaneous pregnancy does occur, there is a high likelihood of pregnancy loss or fetal abnormalities [4-5].
The characteristic morphological TS phenotypes primarily occur in those with the 45,X haplotype [4], which is found in 45 - 50% of TS patients [4]. Other TS

patients are chimeric, with haplotypes including 46, X,I(Xq) and 46,XX/47,XXX. Triple X syndrome, which occurs in individuals with exclusively or primarily a 47,XXX haplotype, is a related disorder that is generally associated with normal gonadal development and reproductive function during puberty [6]. Thus, the ~4% of TS patients with 47,XXX chimerism (including 45,X/47,XXX and 45,X/46,XX/47,XXX cell lines) generally have milder phenotypes than those with the typical 45,X haplotype. Indeed, the relatively rare 45,X/47XXX patients do not require hormone therapy [6] and most spontaneously menstruate without estrogen.

Although chimeric TS patients are known to generally have no cardiac or renal problems, ovarian function has not been well characterized among these patients. In the present study, we describe the phenotypes and genotype of a 41-year-old female with chimeric TS. This case study was designed to increase understanding in the field of reproductive biology regarding possible phenotypes and recommended genotyping methods and support systems for those with chimeric TS [5].

CASE DESCRIPTION

This study was approved by the Medical Ethics Committee of Ganzhou People's Hospital and the patient provided written informed consent. In 2015, the patient delivered a full-term female infant by Cesarean section. From year to year, the patient received medical treatment in the reproductive department of Ganzhou People's Hospital after a lesion removal for a right fallopian tube ectopic pregnancy. Since that time, the patient had used no contraception but had not experienced pregnancy. The patient now presented with bilateral polycystic changes and infertility.

The patient had no medical history of diseases of the kidney, urinary system, heart, or other organs. A physical examination showed no renal or cardiac abnormalities, a normal height and weight (156 cm and 51 kg, respectively), and no multiple nevi. She had no facial abnormalities or other characteristic TS symptoms. In a laboratory examination of liver and kidney function, blood glucose and lipid levels were normal, as was the metabolic composite index. Hormone levels were as follows: follicle-stimulating hormone (FSH), 6.36 mIU/mL (3.5 - 12.5 mIU/mL); luteinizing hormone (LH), 7.89 mIU/mL (2.4 - 12.6 mIU/mL); prolactin (PRL), 24.0 ng/mL (4.79 - 23.3 ng/mL); progesterone (Pro iii), < 0.05 ng/mL (0.057 - 0.893 ng/mL); testosterone (Testo); estradiol (e2Iii), 129.0 pmol/L (45.5 - 854 pmol/L); and anti-Mullerian hormone (AMH), 9.85 ng/mL (0.147 - 7.49 ng/mL). Pelvic ultrasound examination showed normal uterine size and shape, mature follicles on the right side, no follicles on the left side, multiple cervical cysts, and a small amount of fluid in the cervical canal.

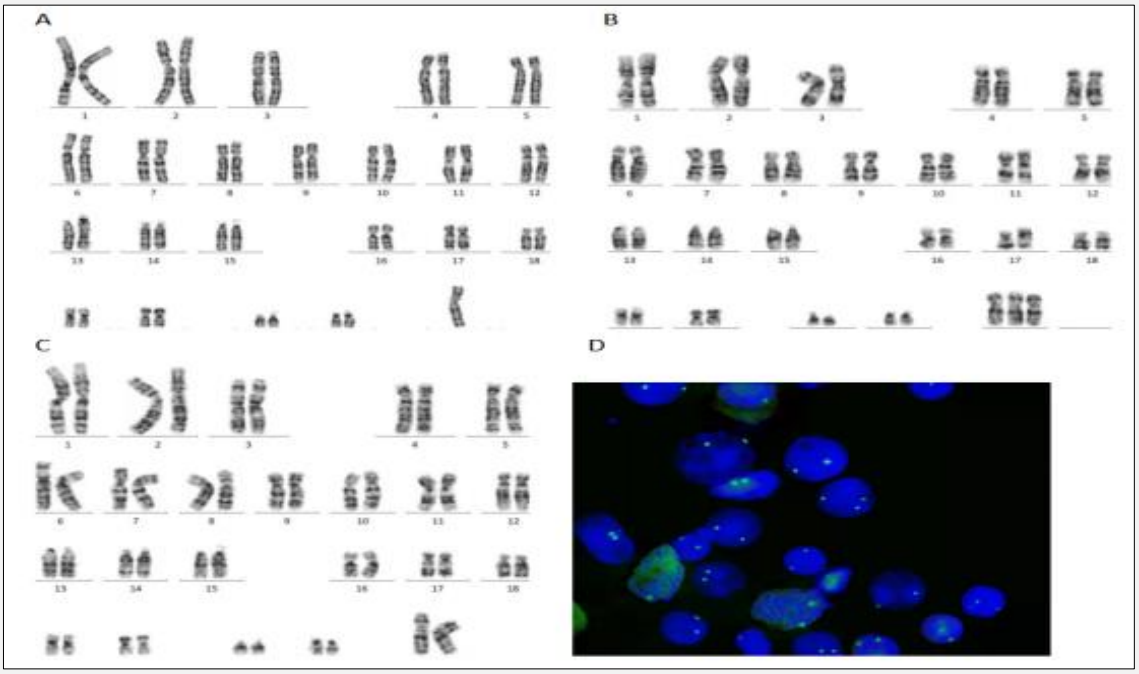
Due to the normal values of the above indicators but the apparent infertility, we conducted chromosome karyotype analysis and fluorescence in situ hybridization (FISH) to identify potential chromosomal abnormalities. Traditional karyotype analysis was performed using g-band lymphocyte medium culture. Of 150 examined cells, 13 were 45,X; six were 47,XXX; and 132 were 46,XX (Figure 1). FISH was used to confirm the chimerism suggested by karyotyping; this revealed 48,XXXX cells in addition to 45,X, 47,XXX, and 46,XX cells. The patient's final haplotype was thus 45,X/47,XXX/46,XX/48,XXXX (Figure 1).

DISCUSSION

Clinical features typical of TS are short stature, multiple moles, heart and kidney disease, and growth delays. TS is typically diagnosed when patients are very young due to these characteristic abnormal morphological phenotypes. However, phenotypes are known to differ between haplotypes, and those with chimeric types may not be diagnosed until much later (e.g., after years of infertility), as occurred in this case. For example, phenotypes are milder among those with the 45,X/47,XXX/46,XX/48,XXXX haplotype than among those with 45,X only cell lines [7]. Consistent with those prior findings, the patient in this case had a relatively high proportion of 46,XX cells and no characteristics typical of TS patients aside from a reduced pregnancy rate, which may have been due to the impacts of the 45,X, 47,XXX, and 48,XXXX cells on ovarian function [8]. However, the specific mechanism associated with these phenomena remains to be studied.

The haplotype identified here is extremely rare; a population-scale frequency has not been reported in the literature due to low sample sizes. Furthermore, 48,XXXX cells were not detected here with the conventional karyotyping approach because there were so few of these cells. This suggests that instances of 45,X/47,XXX/46,XX/48,XXXX chimerism may have been overlooked in the past when only conventional karyotyping was used, resulting in missed diagnoses. The use of FISH with a larger number of cells can therefore complement karyotype analysis in such cases. Importantly, FISH can also be used to detect Y-chromosomal fragments (i.e., mosaic TS) [8]. Due to the rarity of patients with haplotypes such as 45,X/47,XXX/46,XX/48,XXXX, a comprehensive TS karyotype database should be constructed in the future to enable identification of correlations between TS genotypes and phenotypes.

Ovarian function is a considerable concern for TS patients. Chimeric patients have normal initial ovarian function, which declines with age. However, the timings of functional decline and disease onset remain unclear, complicating genetic counseling efforts for those with TS. The development of appropriate indicators to predict ovarian function and patient age at menopausal onset will be critical in such efforts. Prior studies have indicated that serum AMH and AFC levels may be useful markers of ovarian and follicular reserves and accurate



Figures 1. A: 45,X, B: 47,XXX, C: 46,XX, D: FISH:48,XXXX.

predictors of menopausal age [9]. Furthermore, AMH levels are closely related to TS haplotypes [10] and could be used for highly sensitive and specific prediction of impending premature ovarian failure in TS patients. Young TS patients should therefore undergo regular AMH and AFC monitoring for fertility preservation.

CONCLUSION

Our findings highlight the issues faced by individuals with TS who present no or few abnormal phenotypes. To prevent easily missed diagnoses of TS, clinicians diagnosing common genetic disorders should routinely employ two or more methodologies (such as conventional karyotyping combined with FISH) to ensure accurate results. Clinicians are advised to recommend such karyotype analyses for women with any of the following conditions: short stature or growth delay; primary amenorrhea; or ovarian function decline. Patients determined to have TS and chimeric haplotypes (such as 45,X/47,XXX/46,XX/48,XXXX) should be examined and counseled throughout their growth and development to assess ovarian function. If necessary, appropriate technologies can then be applied in a timely fashion to preserve fertility. Overall, our findings expand current knowledge of the clinical symptoms presented by TS patients and promote the application of methods for accurately identifying the condition early, allowing adequate time for appropriate interventions.

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Declaration of Interest:
The authors declare that they have no known competing financial interests. The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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