RESEARCH NOTE

De novo Xp terminal deletion in a triple X female with recurrent spontaneous abortions: a case report

TAHIR M. MALLA[1], ARSHAD A. PANDITH[1], FAYAZ A. DAR[1], MAHRUKH H. ZARGAR[1] and ZAFAR A. SHAH[2]*

[1]*Advanced Centre for Human Genetics, and* [2]*Department of Immunology and Molecular Medicine, Sher-i-Kashmir Institute of Medical Sciences, Srinagar 190 011, India*

[Malla T. M., Pandith A. A., Dar F. A., Zargar M. H. and Shah Z. A. 2014 *De novo* Xp terminal deletion in a triple X female with recurrent spontaneous abortions: a case report. *J. Genet.* **93**, 819–822]

Introduction

Trisomy X (47,XXX) is a human sex chromosome aneuploidy in which females have an extra X chromosome, compared to the 46,XX karyotype in typical females. Females with 47,XXX karyotypes have been reported to have varied phenotypic changes that include features like tall stature, epicanthal folds, hypotonia and clinodactyly. Seizures, renal and genitourinary abnormalities, and premature ovarian failure are also few associated findings. However, puberty, sexual development and fertility are usually normal in trisomy X females (Tartaglia *et al.* 2010). The case presented in this study had two consecutive spontaneous pregnancy losses though she was devoid of any intrauterine malformation as revealed by her ultrasonography report. Her immunological profile was absolutely normal. Apparently, the female was not having any kind of facial deformity or syndromic features. Cytogenetic evaluation of the female revealed 47(XXX) karyotype with del(Xp21→Xpter) which was possibly the only discernible cause of the recurrent spontaneous abortions. Besides, karyotype analysis of the other family members revealed no structural or numerical abnormality of chromosomes. Cytogenetic evaluation of her husband revealed 46,XY karyotype. To our knowledge, till date a triple X female having recurrent spontaneous abortions with del(Xp21→Xpter) has not been reported.

Trisomy X is a condition caused by the presence of an extra X chromosome in females. It is the most common chromosomal abnormality in females, occurring with an incidence of approximately 1 in 1000 female births. As some individuals are only mildly affected or asymptomatic, it is estimated that only 10% of individuals with trisomy X are actually diagnosed (Tartaglia *et al.* 2010). Marked facial dysmorphism or significant physical features are not commonly associated with 47,XXX, however, minor physical findings can be present in some individuals including epicanthal folds, hypertelorism, upslanting palpebral fissures, clinodactyly, overlapping digits, pes planus and pectus excavatum (Ratcliffe 1985). Although, there have been no direct studies of fertility in trisomy X, many reports of successful pregnancies have been described, and fertility is likely normal in most cases unless complicated by a genitourinary malformation or premature ovarian failure (Linden *et al.* 1988).

Case report

The case presented in this study is a 30-year-old female with a history of recurrent spontaneous abortions. She was married since one year at the time of cytogenetic analysis. On physical examination of the patient, no facial deformity or any other physical anomaly was observed. The ultrasonography of her pelvic organs had normal impression and did not reveal any abnormality in the echotexture of any reproductive parts. Her body weight was 58 kg and her height was 162 cms. The patient had no family history of any genetic disorder and spontaneous abortions. From the menarche age of 13, she had menstruated regularly with a normal flow. The patient conceived normally after two months of marriage and had first spontaneous abortion after two and a half months of conception. During first pregnancy the ultrasonography revealed missed abortion. Immunological profile including anti-phospholipid antibody and anti-cardiolipin antibody status was reported to be normal. Her hormonal profile was within normal limits (LH=5.5 mIU/mL, PRL=17.37 ng/mL, T3=1.34 ng/mL, T4=12.02 g/dL, TSH=3.76 μIU/mL, FSH=11.5 IU/mL, progesterone=0.8 ng/mL). After six months of the first spontaneous abortion, she conceived and had another spontaneous abortion in the first trimester.

*For correspondence. E-mail: tahirachg@gmail.com.

Keywords. trisomy X; Xp deletion; spontaneous abortions.

The ultrasonography revealed no embryonic cardiac pulsation and embryonic motions. Her hormonal profile was again within normal limits. Besides, the immunological profile was normal. Infection as a cause for spontaneous abortion was ruled out owing to the negative test results like TORCH and CBC.

Cytogenetic analysis

Lymphocyte culture was carried out according to Moorhead *et al*. (1960). Peripheral venous blood was collected in the heparinized vacutainers and aseptically transferred into a sterile culture bottle with 5–8 mL of RPMI-1640 medium (Sigma, St Louis, USA), supplemented with L-glutamine, 10% feotal bovine serum (Himedia Labs, Mumbai, India), penicillin–streptomycin solution (Invitrogen, California, USA), and phytohaemagglutinin (Himedia Labs, India). The cultures were incubated in a CO_2 incubator for 72 h. Colchicine 50 μL was added at the completion of 70 h to arrest the cells at metaphase. After 72 h of incubation, the cell suspensions were centrifuged for 10 min at 1000 rpm. The supernatant was discarded and the pellet was treated with hypotonic solution (0.075 M KCl) by gentle flushing and cyclomixed. The centrifuge tubes were incubated again at 37°C for 45 min. The tubes were again centrifuged carefully at 1000 rpm for 15 min. The supernatant was removed and 5–8 mL of freshly prepared pre-chilled Cornoys fixative was added to the pellet while mixing in cyclomixer. The tubes were allowed to stand overnight and washed with freshly prepared pre-chilled Cornoy's fixative repeatedly for 3–4 times. GTG banding was performed and the slides were stained with 1% giemsa stain (Seabright 1971). Karyotyping was performed with the help of Cytovision software ver. 3.9 on wellspread G-banded metaphase plates (Applied Imaging, Michigan, USA). At least 30 metaphases were examined to confirm the deletion and to rule out any mosaicism.

Karyotype of the patient was observed to be 47,XXX, del(Xp21→pter) (figures 1 and 2). To rule out the inheritance of the Xp terminal deletion, the other members of the family, including her parents and siblings were also screened. Her elder brother had primary infertility but his karyotype was found to be normal (46,XY). The other siblings and both the parents had normal karyotypes. Besides, the cytogenetic profile of the husband was found to be normal.

Discussion

Recurrent spontaneous abortion is the miscarriage of two or more consecutive pregnancies in the first or early second trimester of gestation (Carp *et al.* 2004). Among all causative factors the few undisputed causes of recurrent pregnancy loss are genetic, anatomic or immunologic factors. However, even after a comprehensive evaluation, recurrent pregnancy loss remains unexplained in more than half of affected couples (Branch *et al.* 2010). As no anatomic or immunologic disorder was found in the present case, the cytogenetic analysis of the affected female along with her family for the detection of any chromosomal anomaly was carried out.

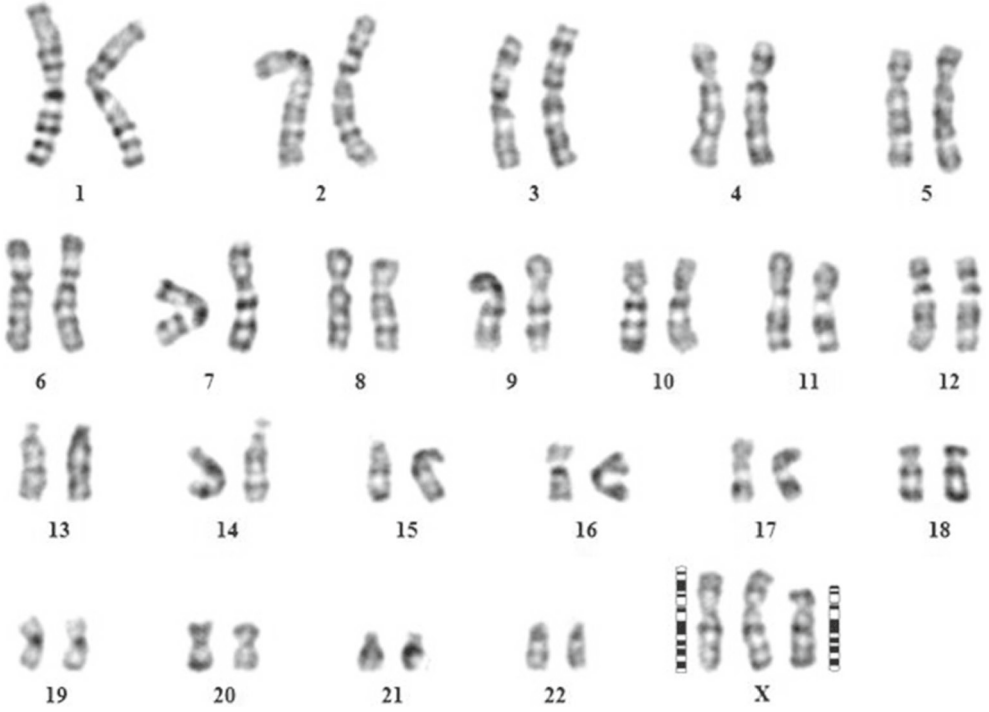

Figure 1. Karyotype of the triple X female showing 47,XXX del(Xp21-pter). Ideograms of the normal X chromosome and the X chromosome with terminal deletion are also shown.

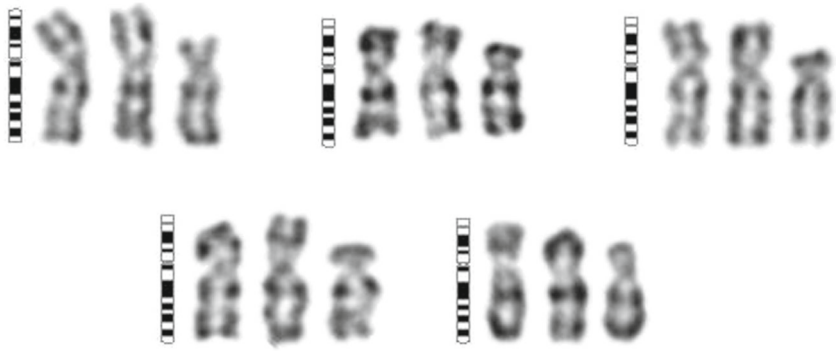

Figure 2. Partial karyotypes of the female showing del(Xp21-pter).

Chromosomal anomalies are known to be the single most common cause of spontaneous abortion. Previous studies indicate that 50% of spontaneously expelled abortuses have been thought to be chromosomally abnormal (Gardner and Sutherland 1996). Among couples with recurrent miscarriages, about 3–5% have one partner with a cytogenetic abnormality (Tharapel *et al.* 1985, Portnoi *et al.* 1988; Fryns and Van Buggenhout 1998). Even higher frequencies of karyotypic abnormalities are found among couples with recurrent miscarriages if other causes, such as uterine malformations, are excluded first (Carp *et al*. 2004).

The risk of spontaneous miscarriage is reportedly high in women with Turner syndrome (TS) who are mosaic for a 46,XX cell line, a 47,XXX cell lineage, or have very distal Xp deletions (Sybert and McCauley 2004). Sybert (2005) summarized the work regarding pregnancy outcomes in women with TS, and reported that up to 39% ($N = 93/233$) of the pregnancies result in spontaneous abortion. Moreover, it has been reported that in females with 46,XX karyotype, partial deletion of Xp may result in TS (Zinn *et al.* 1993). Microphthalmia with linear skin defects (MLS), a rare congenital, X-linked dominant syndrome is also most commonly caused by terminal deletions of Xp (Vergult *et al.* 2013). In contrast with these reports, the patient in discussion was a true 47,XXX female without any mosaic cell line, but had an Xp terminal deletion in the extra copy of the X chromosome that had caused the spontaneous abortions. Females with Xp deletions are phenotypically normal except for short stature, as they need only one active copy of this region to be normal. In the present case, as the female had an extra copy of X chromosome, she did not show any phenotypic changes like short stature.

The Xp21-Xpter region of X chromosome contains various genes like *SS* (MIM312865), *CDPX* (MIM3O 295O), *MRX* (MIM309530), *XLI* (MIM3O81OO), *KAL* (MIM30 870O), *AIC* (MIM3O4O5O), and *FDH* (MIM305600) whose deletion causes multiple phenotypes (Ballabio and Andria 1992). It has been reported that inactivation of the holocytochrome c-type synthase gene (*HCCS*), present on Xp22.2 is lethal in 46,XY condition (Qidwai *et al.* 2010). Therefore, we presume that the case under discussion has been harboring 46,XY feotuses in which, the possible nullisomy for the genes has not been compatible with life and may have therefore resulted in spontaneous abortions.

The present study indicates that the patient had spontaneous abortions due to the *de novo* Xp terminal deletion, the sole discernible abnormality keeping in view of her normal anatomic, immunological, and other profiles. Although hidden mosaicism for a 47,XXX devoid of Xp terminal deletion cell line might be possible, but none was detected pointing towards the likeliness of spontaneous abortions of the patient by the X chromosome terminal deletion. The role of Xp terminal deletion as a cause of abortion in this case was also validated by the normal chromosomal constitution of the husband.

It has been reported by the researchers that a partial deletion of Xp may arise *de novo* or as the result of parental balanced translocations involving a segment of the short arm of chromosome X (Prieto *et al.* 1978; Lai *et al.* 1992; Zinn *et al.* 1993; Hon *et al.* 1995). In the present case, karyotypes of both the parents were devoid of del(Xp21→pter), which confirmed that it was a *de novo* deletion of Xp. To the best of our knowledge, *de novo* terminal deletion of Xp [del(Xp21-pter)] in a triple X female with a history of recurrent spontaneous abortions has not been reported anywhere in the previous works making this case as rare one.

As postkaryotyping genetic counselling is very important, genetic counselling was offered to this case as well and the possible pattern of inheritance of the Xp terminal deletion was explained to the couple in detail.

Conclusion

We conclude from this case study that Xp terminal deletion of the extra copy of X chromosome was the only possible cause of recurrent spontaneous abortions. But, as the female had two normal copies of X chromosome, no syndromic facies or any kind of dysmorphism was observed.

References

Ballabio A. and Andria G. 1992 Deletions and translocations involving the distal short arm of the human X chromosome: review and hypotheses. *Hum. Mol. Genet.* **1**, 221–227.

Branch D. W., Gibson M. and Silver R. M. 2010 Clinical practise. Recurrent miscarrige. *N. Engl. J. Med.* **363**, 1740–1747.

Carp H., Feldman B., Oelsner G. and Schiff E. 2004 Parental karyotype and subsequent live births in recurrent miscarriage. *Fertil. Steril.* **81**, 1296–1301.

Fryns J. P. and Buggenhout V. G. 1998 Structural chromosome rearrangements in couples with recurrent fetal wastage. *Eur. J. Obstet. Gynecol. Reprod. Biol.* **81**, 171–176.

Gardner R. J. M. and Sutherland G. R. 1996 Chromosome abnormalities and genetic coumseling, 2nd edition, pp. 313–317. Oxford University Press, New York, USA.

Hon E., Chapman C. and Gunn T. R. 1995 Family with partial monosomy 10p and trisomy 10p. *Am. J. Med. Genet.* **56**, 136–140.

Lai M. M. R., Sriven P. N., Ball C. and Berry A. C. 1992 Simultaneous partial monosomy 10p and trisomy 5q in a case of hypoparathyroidism. *J. Med. Genet.* **29**, 586–588.

Linden M. G., Bender B. G., Harmon R. J., Mrazek D. A. and Robinson A. 1988 47,XXX: what is the prognosis? *Pediatrics* **82**, 619–630.

Moorhead P. S., Nowell P. C., Mellman W. J., Battips D. M. and Hungerford D. A. 1960 Chromosome preparation of leukocytes cultured from human peripheral blood. *Exp. Cell. Res.* **20**, 613–616.

Otter M., Stumpel S. C. and Curfs L. M. G. 2010 Triple X syndrome: a review of the literature. *Eur. J. Hum. Genet.* **18**, 265–271.

Portnoi M. F., Joye N., Akker V. D. J., Morlier G. and Taillemite J. L. 1988 Karyotypes of 1142 couples with recurrent abortion. *Obstet. Gynecol.* **72**, 31–34.

Prieto F., Badia L., Moreno J. A., Barbere P. and Asensi F. 1978 10p syndrome associated with multiple chromosomal abnormalities. *Hum. Genet.* **45**, 229–235.

Qidwai K., Pearson D. M., Patel G. S., Pober B. R., Immken L. L., Cheung S. W. and Scott D. A. 2010 Deletions of Xp provide evidence for the role of holocytochrome c-type synthase (hccs) in congenital diaphragmatic hernia. *Am. J. Med. Genet. A* **152A**, 1588–1590.

Ratcliffe S. G. 1985 Longitudinal growth studies on children with sex chromosome abnormalities. *Prog. Clin. Biol. Res.* **200**, 301–309.

Seabright M. 1971 A rapid banding technique for human chromosomes. *Lancet* **2**, 971–972.

Sybert V. P. 2005 Turner syndrome. In *Management of genetic syndrome* (ed. S. B. Cassidy and J. E. Allanson), pp. 459-484. Wiley, New York, USA.

Sybert V. P. and McCauley E. 2004 Turner's syndrome. *N. Engl. J. Med.* **351**, 1227–1238.

Tartaglia N. R., Howell S., Sutherland A., Rebecca W. and Lennie W. 2010 A review of trisomy X (47,XXX). *Orphanet. J. Rare Dis.* **5**, 8.

Tharapel A. T., Tharapel S. A. and Bannerman R. M. 1985 Recurrent pregnancy losses and parental chromosome abnormalities: a review. *Br. J. Obstet. Gynaecol.* **92**, 899–914.

Vergult S., Leroy B., Claerhout I. and Menten B. 2013 Familial cares of a submiscroscopic Xp 22.2 deletion: genotype-phenotype correlation in microphthalmia with linear skin defects syndrom. *Mol. Vis.* **19**, 311–318.

Zinn A. R., Page D. C. and Fisher E. M. C. 1993 Turner syndrome: the case of the missing sex chromosome. *Trends. Genet.* **9**, 90–93.

Received 17 March 2014, in final revised form 10 May 2014; accepted 14 May 2014
Unedited version published online: 24 June 2014
Final version published online: 8 October 2014