

## Identification of a *de novo* inv dup(X) (pter→q22) by multicolor banding in a girl with Turner syndrome

P. Burégio-Frota<sup>1</sup>, L. Valença<sup>1</sup>, G.F. Leal<sup>2</sup>, A.R. Duarte<sup>2</sup>, A.V.S. Bispo-Brito<sup>1</sup>,  
E.M. Soares-Ventura<sup>3</sup>, T.J. Marques-Salles<sup>3</sup>, M.T.M.C. Nogueira<sup>3,4</sup>,  
M.T.C. Muniz<sup>3,5</sup>, M.L.M. Silva<sup>6</sup>, F. Hunstig<sup>7</sup>, T. Liehr<sup>7</sup> and N. Santos<sup>1</sup>

<sup>1</sup>Departamento de Genética, Centro de Ciências Biológicas,  
Universidade Federal de Pernambuco, Recife, PE, Brasil

<sup>2</sup>Instituto Materno Infantil Prof. Fernando Figueira, Recife, PE, Brasil

<sup>3</sup>Centro de Oncohematologia Pediátrica de Pernambuco,  
Universidade de Pernambuco, Recife, PE, Brasil

<sup>4</sup>Departamento de Biologia, Instituto de Ciências Biológicas,  
Universidade de Pernambuco, Recife, PE, Brasil

<sup>5</sup>Departamento de Fisiologia, Instituto de Ciências Biológicas,  
Universidade de Pernambuco, Recife, PE, Brasil

<sup>6</sup>Instituto Nacional do Câncer, Rio de Janeiro, RJ, Brasil

<sup>7</sup>Institut für Humangenetik und Anthropologie,  
Friedrich-Schiller-University, Jena, Germany

Corresponding author: N. Santos  
E-mail: santos\_neide@yahoo.com.br

Genet. Mol. Res. 9 (2): 780-784 (2010)

Received January 12, 2010

Accepted February 12, 2010

Published April 27, 2010

DOI 10.4238/vol9-2gmr777

**ABSTRACT.** We report on a 23-year-old girl with short stature, short and wide neck, low posterior hairline, hypogonadism, underdeveloped breasts, infantile uterus, ovaries not visualized, and primary amenorrhea. Cytogenetic G-banding analysis revealed a mosaic karyotype of 46,X,dup(X)(q22)[35]/45,X[15], confirming the clinical suspicion of Turner syndrome. Molecular cytogenetics using a multicolor banding probe set for the X-chromosome characterized an inverted dup(X). The

karyotype of the patient was therefore interpreted as 46,X,inv dup(X) (pter→q22::q22→pter). This patient had a mosaic Turner syndrome with a cell line comprising partial trisomy Xpter to Xq22 and partial monosomy Xq22 to Xqter.

**Key words:** Inverted duplication; Partial monosomy; Partial trisomy; Multicolor banding; Mosaicism

## INTRODUCTION

Turner syndrome (TS) is one of the most common chromosomal disorders, with an incidence of about 1/2500 live born girls. Minimal diagnostic criterion for this condition is an abnormal karyotype in which all or part of one of the X chromosomes is absent in all or some cells (Hanson et al., 2001). Approximately 30-40% of cases present mosaic karyotypes of which the most common are the following: 45,X/46,XX; 45,X/47,XXX; 45,X/46,XX/47,XXX; 45,X/46,XY, and 45,X/any karyotype with a structurally abnormal X or Y (Gorlin et al., 2001). On the other hand, X-duplications in patients with TS are a rare chromosome abnormality.

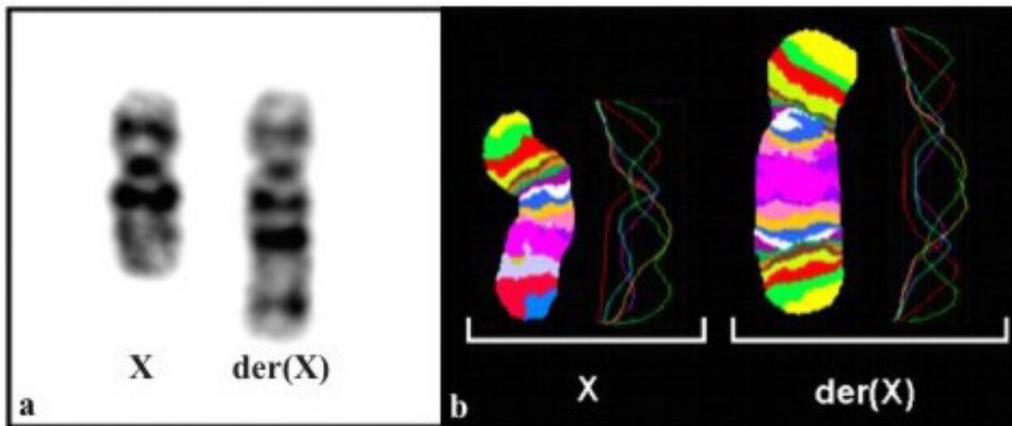
Partial X-chromosome duplications are relatively infrequent and have been identified in males and females. The dup(X) can be inherited or *de novo*, where the former has been detected more frequently, transmitted by female carriers who are usually phenotypically normal, with only short stature. *De novo* duplications are rare and have been associated with abnormalities such as TS, gonadal dysgenesis, or more often congenital anomalies and mental retardation (Van Dyke et al., 1983; Armstrong et al., 2003; Petkovic et al., 2003; Cheng et al., 2005). The aberrant phenotype associated with these duplications is generally attributed to excess gene dosage due to functional disomy for the duplicated region.

In this paper, we describe an unusual case of *de novo* Xp to Xq22 duplication in a patient with TS. To our knowledge, the present report represents the first case of 46,X,inv dup(X) (pter→q22::q22→pter) characterized by molecular cytogenetic multicolor banding (MCB).

## Case report

The patient was born to a normal 39-year-old woman and her healthy and nonconsanguineous 39-year-old husband. The couple had a son and five other daughters with no clinical problems, but the mother had one first-trimester spontaneous abortion. Pregnancy was unremarkable except for an episode of vaginal bleeding in the sixth month. Delivery was at term by cesarean section. Birth weight was 3.0 kg. Motor development was normal. She was able to read and write at the age of 6 years and finished high school at 18 years. The patient had primary amenorrhea. Clinical examination at the age of 23 years showed the following: height 146 cm (slightly below the 3rd percentile), weight 64.5 kg (10th percentile), occipitofrontal circumference 56 cm (25-50th percentile). The girl presented short and webbed neck, low posterior hairline, posteriorly rotated ears, asymmetry of palpebral fissure length, absence of upper lateral incisors, scarce axillary hairs, and short third and fourth metacarpals. Tanner's stage was M2, P2. Ultrasound studies showed a hypoplastic uterus and normal kidneys. Ovaries were not seen by ultrasound studies. Endocrinologic evaluation revealed secondary hypogonadism (decreased estradiol, FSH and LH levels). G-banding chromosome analysis revealed a karyotype

of 46,X,dup(X)(q22)[35]/45,X[15] (Figure 1a). Karyotype analysis of the parents showed normal results. Multicolor banding for the X chromosome was used to characterize the duplicated region in more detail, and it was done as previously described (Liehr et al., 2002). The chromosomal constitution of the patient was interpreted as 46,X,inv dup(X)(pter→q22::q22→pter) comprising a partial Xpter to Xq22 duplication and partial Xq22 to Xqter deletion (Figure 1b). C-banding confirmed that the derivative X-chromosome had two centromeres.



**Figure 1.** Partial karyotype of the proband. **a.** G-banding showing a normal X-chromosome (left) and derivative chromosome (right). **b.** Multicolor banding of X-chromosome characterized a normal X-chromosome and an inv dup(X)(pter→q22::q22→pter).

## DISCUSSION

Duplications of the X chromosome are rare chromosome rearrangements and have been reported predominantly in males with multiple congenital anomalies and developmental delay (Armstrong et al., 2003; Cheng et al., 2005; Stankiewicz et al., 2005). Approximately 20 cases of Xq duplication have been described in females, and the abnormal phenotype usually included short stature, mental retardation/developmental delay, hypotonia, hypogonadism, microcephaly, and various minor dysmorphic anomalies. As a consequence of skewed X-chromosome inactivation resulting in inactivation of the dup(X) chromosome, and selection against cells with active abnormal X in carrier females, most dup(Xq) females appear phenotypically normal (Armstrong et al., 2003; Stankiewicz et al., 2005). Here, we report on a patient with an inverted duplication in 70% of the cells analyzed and Turner stigmata. As her parents were normal and did not show the duplicated segments, the inv dup(Xq) was believed to be *de novo* in origin. Only a few patients with tandem duplications have been found to be mosaic with a normal cell line (Van Dyke et al., 1983). The majority of the cases have occurred *de novo* and are characterized by multiple congenital anomalies and developmental delay. To date, it has been difficult to determine the exact cytologic nature of chromosomal duplication, which can be direct and inverted. Mismatched pairing of homologs and unequal crossover between sister or non-sister chromatids, as well as three break rearrangements that include a U-type rear-

rangement for inverted duplications, have generally been accepted (Taylor et al., 1977; Van Dyke et al., 1983; Kotzot et al., 2000). Nevertheless, Petkovic et al. (2003) discussed that a dicentric X-chromosome, in a girl with moderate growth retardation and 45,X/46,X,psu dic(X)(q22.3) karyotype, was the result of an isocal break in both chromatids of the paternal X and subsequent rejoining of the broken ends, followed by the inactivation of one centromere.

The stigmata of TS are growth retardation with reduced adult height, failure to undergo puberty, and an accelerated rate of atresia of ovarian follicles, causing gonadal insufficiency and infertility (Bondy 2005). Phenotypic expression in TS patients depends on the karyotype, and the identification of sex chromosome mosaicism plays a key role in clinical management. However, the presence in our patient, as in most other reported cases, of a 45,X cell line makes any genotype-phenotype correlation complicated. Thus, the evaluation of clinical features associated with gain and loss of the particular X chromosomal segment in this case is inconsistent. We believe that the clinical data described here are probably caused by the 45,X cell line, as a consequence of loss of an unstable der(X). If the rule of selective lyonization holds, the dup(X) is preferentially inactivated, and normality should be expected.

Multicolor banding is a recently developed technique that allows the differentiation of chromosome region-specific areas at the band and sub-band level, producing changing fluorescence intensity ratios along the chromosomes. The color bands have a great value in identifying chromosomal abnormalities, particularly complex chromosome rearrangements, and intrachromosome exchanges (i.e., inversions, deletions, duplications, and insertions). These abnormalities cannot be easily defined by conventional cytogenetic analysis or chromosome paint (Hu et al., 2006; Weise et al., 2008). In our case the use of MCB enables an appreciation, at first glance, that X(pter→q22) regions are duplicated and inverted, and that another part of Xq is deleted.

Our study demonstrated that the MCB technique allows a powerful focus on the mechanism underlying several types of rearrangements and is a useful complementary technique for the analysis of complex intrachromosome abnormalities.

## ACKNOWLEDGMENTS

The authors thank the parents and clinicians for the data. Research supported by the Fundação de Amparo à Ciência e Tecnologia do Estado de Pernambuco (FACEPE - APQ-0335-2.02/06), Brazil, and the IZKF Jena, Germany.

## REFERENCES

- Armstrong L, McGowan-Jordan J, Brierley K and Allanson JE (2003). *De novo dup(X)(q22.3q26)* in a girl with evidence that functional disomy of X material is the cause of her abnormal phenotype. *Am. J. Med. Genet. A* 116A: 71-76.
- Bondy CA (2005). New issues in the diagnosis and management of Turner syndrome. *Rev. Endocr. Metab. Disord.* 6: 269-280.
- Cheng SF, Rauen KA, Pinkel D, Albertson DG, et al. (2005). Xq chromosome duplication in males: clinical, cytogenetic and array CGH characterization of a new case and review. *Am. J. Med. Genet. A* 135: 308-313.
- Gorlin RJ, Cohen MM Jr and Hennekam RCM (2001). *Syndromes of the Head and Neck*. 4th edn. Oxford University Press, New York.
- Hanson L, Bryman I, Barrenas ML, Janson PO, et al. (2001). Genetic analysis of mosaicism in 53 women with Turner syndrome. *Hereditas* 134: 153-159.
- Hu J, Sathanoori M, Kochmar SJ and Surti U (2006). Application of multicolor banding for identification of complex chromosome 18 rearrangements. *J. Mol. Diagn.* 8: 521-525.

- Kotzot D, Martinez MJ, Bagci G, Basaran S, et al. (2000). Parental origin and mechanisms of formation of cytogenetically recognizable *de novo* direct and inverted duplications. *J. Med. Genet.* 37: 281-286.
- Liehr T, Heller A, Starke H, Rubtsov N, et al. (2002). Microdissection based high resolution multicolor banding for all 24 human chromosomes. *Int. J. Mol. Med.* 9: 335-339.
- Petkovic I, Barisic I and Bago R (2003). Cytogenetic evaluation, fluorescence *in situ* hybridization, and molecular study of psu idic(X)(pter→q22.3::q22.3→pter) chromosome aberration in a girl with moderate growth retardation. *Croat. Med. J.* 44: 494-499.
- Stankiewicz P, Thiele H, Schlicker M, Cseke-Friedrich A, et al. (2005). Duplication of Xq26.2-q27.1, including SOX3, in a mother and daughter with short stature and dyslalia. *Am. J. Med. Genet. A* 138: 11-17.
- Taylor KM, Francke U, Brown MG, George DL, et al. (1977). Inverted tandem ("mirror") duplications in human chromosomes: -nv dup 8p, 4q, 22q. *Am. J. Med. Genet.* 1: 3-19.
- Van Dyke DL, Miller MJ and Weiss L (1983). The origin of inverted tandem duplications, and phenotypic effects of tandem duplication of the X chromosome long arm. *Am. J. Med. Genet.* 15: 441-450.
- Weise A, Mrasek K, Fickelscher I, Claussen U, et al. (2008). Molecular definition of high-resolution multicolor banding probes: first within the human DNA sequence anchored FISH banding probe set. *J. Histochem. Cytochem.* 56: 487-493.