Identification by molecular diagnosis of mosaic Turner's syndrome in an obligate carrier female for fragile X syndrome

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Abstract

A case of mosaic Turner's syndrome with a 45,X/46,XX/47,XXX karyotype, who was also a fragile X obligate carrier as the mother of an affected boy, was identified by molecular diagnosis. Complete haplotyping and direct DNA analysis showed that the X chromosome in all metaphases was the normal X. At the age of 57, she is mentally normal. Her external appearance was typical of Turner's syndrome.

This report shows that molecular studies in conjunction with cytogenetic analysis can help in the clinical diagnosis of a rare case and can show the uniqueness of a case such as the one here described.

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It has recently been shown that the detection of chromosomal mosaicism can be substantially enhanced if at least two kinds of tissue and a greater number of cells are examined. Only 20 to 25% of women with Turner's syndrome are non-mosaic 45,X, whereas two-thirds of patients overall exhibit chromosomal mosaicism.¹ In general, patients with sex chromosome mosaicism exhibit milder signs of the typical phenotype, so that fertility in these women has been reported.²

Patients with Turner's syndrome, as well as hemizygous males, can be affected by X linked disease owing to the lack of one X chromosome. Hence, cases of Turner's syndrome and Duchenne muscular dystrophy (DMD) have been described.³ The case of a DMD carrier detected by molecular diagnosis in a female with mosaic Turner's syndrome was reported two years ago.⁴

The fragile X syndrome (fra(X)) is the most frequent cause of inherited mental retardation, with an incidence of about 1 in 1500 males and 1 in 2500 females.⁵ The case we report here was found in a family referred to us for fra(X) examination owing to mental retardation in two male members. To our knowledge it is the first case reported of a woman who had mosaic Turner's syndrome, who was a fra(X) carrier, and who was the mother of an affected male.

Case report

A woman (III.2, fig 1) was referred to us five years ago for genetic counselling because she had an uncle and a cousin who were mentally retarded. Her family lives a long way away so they could not be examined. Photographs of the family suggested fragile X syndrome, so we requested by mail samples of blood from the two retarded males. The karyotypes showed 33% (II.5) and 47% (III.1) of fra(X) cells. The karyotype of the index case proved normal.

Since not all fragile X carriers show cytogenetic expression, we wished to confirm by molecular diagnosis that she was not a carrier for fra(X). Blood samples were obtained by mail from the entire family, and complete haplotyping was carried out with eight probes (fig 1). We were then able to determine that our index case (III.2) had not inherited the fra(X) mutation.

Surprisingly, the DNA sample taken from patient II.1 appeared to be homozygous for all the polymorphic markers studied. The haplotype did not seem to be of maternal origin. In addition, the bands exhibited the characteristic

Figure 1 Pedigree of the family with the haplotypes of the fra(X) region. Proximal to the centromere: F1X, Cx55.7, 55E, 4D8, and RNI. Distal, UG.2, 1A1, and St.14.

Figure 2 Southern blot analysis with the DNA probe StB12.3 (EcoRI-EagI). * = smear in affected males. An arrow marks the unique restriction fragment in the index case (II-1) corresponding to the normal X. pm = premutation in the grandmother.

intensity of the male type. There were at that time no further data available regarding this woman.

Last year we again studied all our fra(X) pedigrees⁶ using the direct probe StB12.3 isolated and described by Oberlé *et al.*⁷ The pattern found in subject II.1 was that of a normal male (fig 2).

Having seen these results, we asked the patient to be examined. She is mentally normal, aged 57 years, with the typical appearance of Turner's syndrome: short stature (1.31 m), webbed neck, cubitus valgus, pectus excavatum, abnormal ear lobes, and very convex nails. Menarche occurred at 16 years and menopause at 30. She said that she had suffered no health problems in the past, apart from arthritic pains and renal colic suffered over the past five years. Cytogenetic study showed 83.8% 45,X/11.7% 46,XX/4.5% 47,XXX (mosaic Turner's syndrome).

Having made this diagnosis, we re-examined the Southern blots that had been carried out two years ago. With a few of the markers, bands of very low intensity could be seen. These bands correspond to the X chromosome of maternal origin (fig 3). We con-

Figure 3 Southern blot analysis with Msp + St14. An arrow marks the very low intensity allele corresponding to the maternal X chromosome involved in the Turner mosaic.

cluded that the X chromosome present in all cells is the normal X from the father, whereas the fra(X) chromosome from the mother is present only in 16.2% of the cells.

Discussion

Mosaicism of the type 45,X/46,XX/47,XXX arises from non-disjunction in a chromosomally normal female during mitosis after fertilisation. In general, mosaic Turner's syndrome shows milder signs than other cases of Turner's syndrome with 45,X only. When the proportion of 45,X cells increases, symptoms of Turner's syndrome begin to appear. Our case II.1, having 83.8% of 45,X cells, is phenotypically typical of Turner's syndrome except for fertility. The proportion of 45,X cells increases in older women⁸ because, in general, mitotic non-disjunction, as well as other chromosome aberrations, increases in older persons, who show more aberrant cells than younger ones. We thus felt that our patient, karyotyped at 57 years, should have had fewer abnormal cells before. Moreover, the karyotype was performed on a blood sample and blood cells are different in origin from gonadal ones, which are of extra-embryonic origin. Hence if there were more normal cells in gonadal tissue, this could explain this woman's 14 year fertility. Nielsen *et al*² reported 23 similar cases of mosaic Turner's syndrome in which there were 56 pregnancies.

Fertility might have given problems for this patient because she was also a carrier for fra(X) syndrome. Direct molecular diagnosis and haplotyping showed that the X chromosome carrying the fra(X) mutation was the one involved in the Turner mosaicism, or rather the one lost in many cells. We thus consider that there was a process of natural selection

working against the abnormal X, and that this selection, being incomplete, failed to preclude fertility and made possible the transmission of the fra(X) to the only son.

Initially the aim of the study was to find out whether our index case (III.2) was a carrier for fra(X) syndrome. The finding we report here emphasizes, however, that although direct DNA analysis of fra(X) syndrome is now frequent in laboratories, cytogenetics still has an important role to play in the identification of certain cases. Our conclusion is that molecular studies in conjunction with cytogenetic analyses can enhance the clinical diagnosis of rare cases and furthermore can facilitate the identification of unique cases like the one reported here.

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