Muscular dystrophy in an X;1 translocation female suggests that Duchenne locus is on X chromosome short arm

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SUMMARY   A unique combination of a Duchenne-like muscular dystrophy in a girl with a translocation-inversion rearrangement involving an X chromosome and a no 1 chromosome appeared as a result of both gene mutation and chromosome mutation in the mother. The X-autosome rearrangement would permit full expression of an X-linked recessive gene, such as that for Duchenne muscular dystrophy, in a female, and this would satisfactorily explain the characteristic Duchenne-like course of our patient's illness. The simultaneous *de novo* appearance of the Duchenne mutation and the X;1 rearrangement suggests possible sites for the Duchenne locus on the X chromosome short arm (at Xp1106 or Xp2107).

The classic Duchenne type of muscular dystrophy leads inevitably to early death (Stevenson, 1953). The fundamental biochemical defect is unidentified, and diagnosis is on clinical and neuropathological grounds alone. The Duchenne locus has not been mapped, but is certainly on the X chromosome: the disorder appears almost exclusively in males, and pedigrees with more than one affected generally show an X-linked pattern of inheritance (Morton and Chung, 1959). When a girl has progressive muscular dystrophy, it is usual for this to pursue a less rapidly fatal course than Duchenne in boys, and if two or more persons in the family are similarly affected, cases are limited to males and females within a single sibship, compatible with autosomal recessive inheritance (Emery, 1964; Emery and Walton, 1967; Ionasescu and Zellweger, 1974; Kakulas *et al.*, 1975).

In a few dystrophic females, a chromosome anomaly, a 45,X karyotype, makes them effectively male for X-linked recessives genes, and they show full expression of a Duchenne-like disorder (Walton, 1956; Ferrier *et al.*, 1965; Jalbert *et al.*, 1966; Berg and Conte, 1974). In normal XX females, failure of random X inactivation may lead to expression of the Duchenne gene in heterozygous carriers; slight expression is described by Moser and Emery (1974), and perhaps a more extreme case of non-random X inactivation, in one of a pair of 46,XX monozygotic

twins, by Gomez *et al.* (1977). Females who carry structural rearrangements of the X chromosome, including balanced translocations, show a non-random distribution of inactivation patterns (Therman and Pätau, 1974; Laurent *et al.*, 1975; Leisti *et al.*, 1975; Hagemeijer *et al.*, 1977). These females are also at risk for X-linked recessive disorders normally expressed only in males.

Case report

Our patient is a girl. The diagnosis of muscular dystrophy was made at 5 years of age. The development of disability and of clinical signs have run a course in every way compatible with a diagnosis of Duchenne muscular dystrophy, apart from the obvious difficulty that the patient is female.

She was born 10 days after term, weighing 3420 g, and developed well as a baby. She first walked at 15 months, but was always unsteady. At 5 years, she could hardly climb stairs alone, and rose from the floor using Gowers's manoeuvre. There was then calf muscle hypertrophy, the legs and back were more severely affected than the arms, the major areas of weakness were proximal, and tendon reflexes were present. She later developed tendo-Achilles contractures, and now, at 8 years, can move little without the help of a wheelchair. Intelligence remains normal.

Initial serum creatine kinase (CK) was grossly

raised, (10 IU/ml). Electromyography suggested a myopathy. Muscle biopsy showed degenerating muscle fibres filled with macrophages, numerous regenerating fibres, and much replacement by fat and fibrous tissue. Muscle fibres were very variable in size, usually small or atrophied, and rounded, with central nuclei. A few inflammatory cells surrounded perimysial blood vessels. Silver stains showed normal axons and end-plates, histochemistry showed involvement of both main muscle types, and electron microscopy features were of a non-specific muscle fibre degeneration. These findings are typical of Duchenne muscular dystrophy.

Of the 100 buccal mucosa cells examined, 28 carried single normal Barr bodies. Lymphocyte metaphases showed a modal 46 and no cells with X monosomy. Banding, with trypsin-Leishmann, showed a rearrangement involving the short arms of a 1 and an X (Fig. 1). The simplest interpretation is in terms of a three-break event. The distal 1, broken at 1p3400, is exchanged with distal Xp, broken at Xp2107. Proximal Xp is paracentrically inverted between Xp2107 and Xp1106 (Fig. 2).

Translocated and normal X chromosomes are of nearly equal length. A modification of the BrdU technique of Hagemeijer *et al.* (1976) showed the normal X to be late replicating in all 75 cells examined (Fig. 3). Presumably, the translocated X is active in all or nearly all of the body cells.

There are no relatives with muscular dystrophy, though 3 distant paternal cousins suffered from forms of spinal muscle atrophy; they were well-documented, with a full necropsy report in one case.

The patient's parents and only sib, a male, are well, without signs of muscular dystrophy or weakness, and all have normal karyotypes, that is, the X;1 rearrangement arose as a mutation. The patient's mother has 4 healthy brothers and a sister who has just delivered a girl. Pedigree analysis, combined with CK testing of all close female relatives, makes it unlikely that our patient's mother is a Duchenne carrier (probability of being a carrier = 0·01).

Segregation of the X-linked blood group Xg^a suggests that the translocation is maternal in origin: the father is Xg^a negative, his wife and both children Xg^a positive. Other blood groups and biochemical polymorphisms showed no anomalies of segregation.
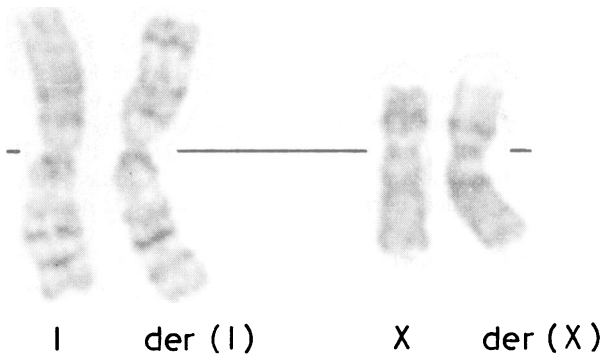

I　　der (I)　　　　X　　der (X)

Fig. 1　*Partial karyotype (trypsin-Leishmann). der, derived chromosome, ie, involved in translocation/translocation + inversion.*
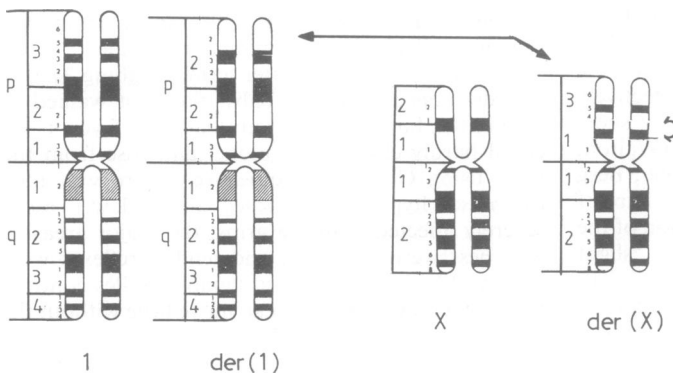

Fig. 2　*Diagrammatical representation of rearrangement, based on Paris Conference (1971).*
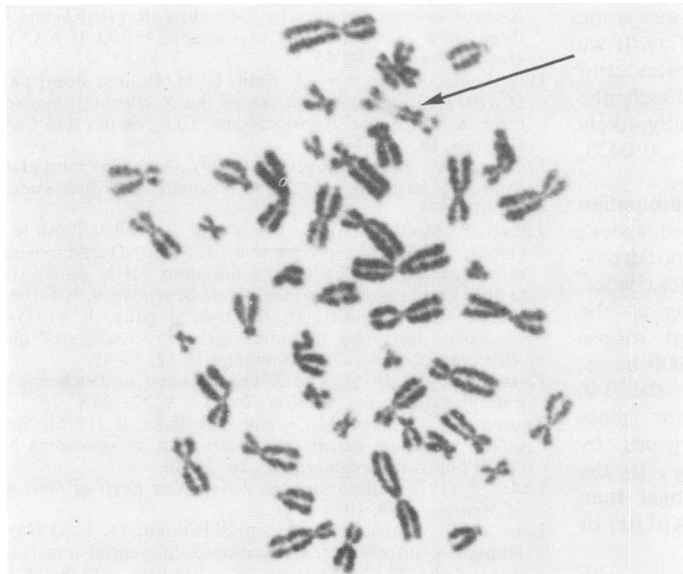
Fig. 3 *Metaphase, BrdU technique. Arrow indicates late replicating X (normal pattern).*

Examination for polymorphisms of the qh region of chromosome 1 was uninformative, as was colour vision testing. Two of the mother's brothers are protanomalopes, but the proband, sib, and parents have normal vision.

Discussion

It is thought that normal 46,XX females randomly inactivate an X in all somatic cells at an early stage of development (Lyon, 1961, 1962). Should they carry a deleterious X-linked recessive gene, this results in mosaicism, the harmful gene being fully expressed in some cells and inactivated in others. Duchenne heterozygotes may show calf hypertrophy, moderately raised CK levels, and suffer muscle cramps and mild weakness (Moser and Emery, 1974).

Structural rearrangements of the X chromosome upset the process, or effects, of random inactivation, and in most or all somatic cells the pattern found is that which minimally upsets the active genome. When the rearrangement involves only the X (ring, isochromosome, or deletion), this chromosome is the inactive late replicating one, and the normal X is active in the somatic cells.

A different pattern is found in persons with X-autosome translocations. In 20 of 40 families with such a translocation, one or more females carry a balanced karyotype, and 22 of 27 balanced females inactivate the normal X, leaving active the full complement of a single X chromosome, which is divided between the two translocation chromosomes

(Therman and Pätau, 1974; Laurent *et al.*, 1975; Leisti *et al.*, 1975; Hagemeijer *et al.*, 1977). This generally results in a normal phenotype, but as the translocated X is consistently the active one, should it carry a deleterious recessive gene this will be expressed in all body cells. In this sense, the phenotype resembles that of a hemizygous male and not that of a heterozygous female. This situation has been found in a female with an X;9 translocation and the X-linked recessive type of anhidrotic ectodermal dysplasia (P. J. L. Cook, 1978, personal communication). It also appears in the case we describe. The alternative explanation, that our case suffers from an autosomal recessive dystrophy, and that the translocation is coincidental, fits the clinical course less satisfactorily and is overall less probable.

Gene and chromosome mutations appeared *de novo*, and necessarily involve the same X chromosome. They could have arisen independently; alternatively, one of the breaks on the Xp produced the Duchenne mutation as well as the translocation-inversion. These mutually exclusive situations have the following relative probabilities. For the former, the Duchenne mutation rate is $4·6 - 10·5 \times 10^{-5}$ (Katsantoni, 1976); for the latter, it is twice the length of the Duchenne locus divided by the total length of the X 'available' as a site for Duchenne. Linkage studies suggest at least one third of Xq, the long arm, is excluded (Zatz *et al.*, 1974; Pearson *et al.*, 1975). There are about $3·2 \times 10^9$ base pairs in the haploid human genome (Comings, 1972), and

'available' X is 0·0423 of the total haploid autosome length (derived from Paris Conference, 1971). If we use the lower of the quoted mutation rates, and write D for the number of base pairs at the Duchenne locus, the alternative situations are equally likely when $4·6 \times 10^{-5} = D \times 2/3·2 \times 10^9 \times 0·0423$, which solves as $\underline{D} = 3114$ base pairs.

Recombinant DNA techniques show mammalian structural genes to carry 'intervening sequences', interrupting the codon series with bases not translating to amino-acids in the protein product (Leder, 1978). These sequences may be as long as the structural sequences themselves, so that mouse β-globin must comprise at least 900 to 1000 bases, rather than the 438 corresponding to the 146 structural codons. Furthermore, regulator genes might be separated from structural genes by sequences several thousand bases long. If the Duchenne gene complex is effectively longer than 3114 bases, it is probably sited at either Xp1106 or Xp2107.

For two further X-autosome translocations, with breaks in Xp21, associated with Duchenne, see Sanki et al. (1979), Annales de Génétique, 22, 33–39.

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References

Berg, G., and Conte, F. (1974). Duchenne dystrophy in a female with a structurally abnormal X chromosome. *Neurology*, **24**, 356.

Comings, D. E. (1972). The structure and function of chromatin. In *Advances in Human Genetics*, Vol. 3, pp. 237–431. Ed. by H. Harris and K. Hirschhorn. Plenum, New York and London.

Emery, A. E. H. (1964). Hereditary myopathies. *Clinical Orthopaedics*, **33**, 164–173.

Emery, A. E. H., and Walton, J. N. (1967). The genetics of muscular dystrophy. In *Progress in Medical Genetics*, Vol. 5, pp. 116–145. Ed. by A. G. Steinberg and A. G. Bearn. Heinemann, London.

Ferrier, P., Bamatter, F., and Klein, D. (1965). Muscular dystrophy (Duchenne) in a girl with Turner's syndrome. *Journal of Medical Genetics*, **2**, 38–46.

Gomez, M. R., Engel, A. G., Dewald, G., and Peterson, H. A. (1977). Failure of inactivation of Duchenne dystrophy X-chromosome in one of female identical twins. *Neurology*, **27**, 537–541.

Hagemeijer, A., Hoovers, J., Hasper-Voogt, I., von Ruhe-Zurcher, T., and Bootsma, D. (1976). Late-replicating ring X-chromosomes identified by R-banding after BrdU-pulse: three new examples of mosaicism 45,XO/46,Xr(X). *Human Genetics*, **34**, 45–52.

Hagemeijer, A., Hoovers, J., Smit, E. M. E., and Bootsma, D. (1977). Replication pattern of the X chromosomes in three X/ autosomal translocations. *Cytogenetics and Cell Genetics*, **18**, 333–348.

Ionasescu, V., and Zellweger, H. (1974). Duchenne muscular dystrophy in young girls? *Acta Neurologica Scandinavica*, **50**, 619–630.

Jalbert, P., Mouriquand, C., Beaudoing, A., and Jaillard, M. (1966). Myopathie progressive de type Duchenne et mosaïque XO/XX/XXX; considérations sur la genèse de la fibre musculaire striée. *Annales de Génétique*, **9**, 104–108.

Kakulas, B. A., Cullity, P. E., and Maguire, P. (1975). Muscular dystrophy in young girls. *Proceedings of the Australian Association of Neurologists*, **12**, 75–79.

Katsantoni, A. (1976). The X chromosome in Duchenne's muscular dystrophy. *Clinical Genetics*, **9**, 371–373.

Laurent, C., Biemont, M-Cl. and Dutrillaux, B. (1975). Sur quatre nouveaux cas de translocation du chromosome X chez l'homme. *Humangenetik*, **26**, 35–46.

Leder, P. (1978). Discontinuous genes. *New England Journal of Medicine*, **298**, 1079–1081.

Leisti, J. T., Kaback, M. M., and Rimoin, D. L. (1975). Human X-autosome translocations: differential inactivation of the X chromosome in a kindred with an X-9 translocation. *American Journal of Human Genetics*, **27**, 441–453.

Lyon, M. F. (1961). Gene action in the X-chromosomes of the mouse (Mus musculus L.). *Nature*, **190**, 372–373.

Lyon, M. F. (1962). Sex chromatin and gene action in the mammalian X-chromosome. *American Journal of Human Genetics*, **14**, 135–148.

Morton, N. E., and Chung, C. S. (1959). Formal genetics of muscular dystrophy. *American Journal of Human Genetics*, **11**, 360–379.

Moser, H., and Emery, A. E. H. (1974). The manifesting carrier in Duchenne muscular dystrophy. *Clinical Genetics*, **5**, 271–284.

Paris Conference (1971). Standardisation in human cytogenetics. *Birth Defects: Original Article Series*, **8**, No. 7, 1972. The National Foundation—March of Dimes, New York.

Pearson, P. L., Sanger, R., and Brown, J. A. (1975). Report of the committee on the genetic constitution of the X chromosome. *Cytogenetics and Cell Genetics*, **14**, 190–195.

Stevenson, A. C. (1953). Muscular dystrophy in Northern Ireland. I. An account of the condition in 51 families. *Annals of Eugenics*, **18**, 50–93.

Therman, E., and Pätau, K. (1974). Abnormal X chromosomes in man: origin, behaviour and effects. *Humangenetik*, **25**, 1–16.

Walton, J. N. (1956). The inheritance of muscular dystrophy: further observations. *Annals of Human Genetics*, **21**, 40–58.

Zatz, M., Itskan, S. B., Sanger, R., Frota-Pessôa, O., and Saldanha, P. H. (1974). New linkage data for the X-linked types of muscular dystrophy and G6PD variants, colour blindness, and Xg blood groups. *Journal of Medical Genetics*, **11**, 321–327.

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