Multiple Abnormalities Due to Possible Genetic Inactivation in an X/Autosome Translocation

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INTRODUCTION

There have been several reports of translocations in man that involve the X chromosome. Lie et al. [1] reported a young woman with a karyotype in which a chromosomal segment of undetermined origin had been translocated to the short arm of the late-replicating X chromosome (46,XX,p+). Various abnormalities displayed by the young woman were interpreted as due to the loss of a portion of the short arm of the X involved in the translocation. Mukerjee and Burdette [2] described a child with multiple congenital abnormalities associated with a ring 3 and a late-replicating translocated 3/X chromosome (46,XX,p+,3r). The abnormalities in this case were due apparently to the loss of genetic material during the formation of the ring and the translocation, and to the subsequent instability of the ring itself. Neuhauser and Back [3] described a child with multiple abnormalities who had a translocation that involved a C group autosome and the late-replicating X chromosome (45,XX,p+,C−). The portion of the C group autosome not translocated to the X was lost, and again the abnormalities were attributed to the loss of genetic material. German [4] reported a malformed infant with a translocation involving a chromosomal segment of undetermined origin and the long arm of the late-replicating X (46,XX,q+). Her abnormalities were thought to be the result of genetic duplication.

The child described in this report also has a translocation that involves the X chromosome and also displays a variety of abnormalities. Unlike the previously mentioned patients, however, she displays abnormalities that are due not to a loss or gain of genetic material, but apparently to genetic inactivation.

CASE REPORT

The proband, a white female infant, was the offspring of an 18-year-old unmarried mother and a 16-year-old father. Born May 29, 1968, she was the product of an uncom-

Received November 25, 1970; revised January 27, 1971.

This investigation was supported in part by a grant from the National Foundation—March of Dimes.

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plicated full-term first pregnancy. The delivery was uncomplicated and the birth weight was 3,480 g. *Talipes equinovalgus* found at birth on the right side was subsequently treated with a series of plaster casts. She had no history of hyperbilirubinemia. According to her mother, she was hypotonic and sometimes constipated but usually ate well and cried little during the newborn period. Strabismus has been noted since five months of age. She has had repeated myoclonic convulsions since birth (at least five times with elevated fever) although she had been receiving phenylbarbital regularly. Notwithstanding generally good health, her motor and mental development has been very slow. She could not sit up unaided until 17 months of age. She could transfer objects from one hand to another at 26 months of age, but could not walk even with assistance. The only words she could articulate were "mama" and "dada" without reference to her parents.

On her last visit, at 26 months of age, her weight was 16.7 kg (above the ninetieth percentile); her height, 93 cm (above the eighty-fourth percentile); her occipitofrontal circumference, 46 cm (−2 standard deviations); and her chest circumference, 57.5 cm (above the ninetieth percentile). Her anterior fontanelle was closed. Her posterior hairline was low and her neck normal. Her mid-face was hypoplastic and depressed. Her eyes were almond-shaped, the interpupillary distance being 4 cm. The sclera in both eyes was normal. She could follow moving objects with both eyes. Her nose was broad and the antitragus on both ears prominent. Her mouth resembled that of a carp. Her chest was shieldlike, and the nipples were widely spaced and underdeveloped (fig. 1). There were

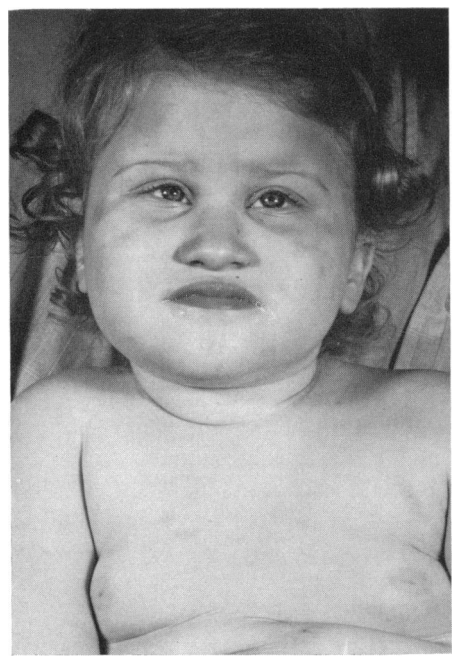

FIG. 1.—The patient at 26 months of age

pits below the acromion. Her external genitalia were normal. No heart murmur was heard. Her hands were puffy and had short tapered fingers. Each palm had a single transverse crease and two triradii, one in the t′ position and one in the t″ position. The atd angles of the right hand were 138° and 62°; of the left hand, 133° and 68°. There

were ulnar loops on all but the second finger of each hand. There was a radial loop on the second finger of the right hand and a whorl on the second finger of the left hand, as well as on the toes of both feet. Both feet were puffy and had hypoplastic nails.

Roentgenograms of her chest and vertebrae were normal. Roentgenograms of her skull revealed a shortened anterior-posterior diameter and an increase in the vertical dimension that resulted in a dolichocephalic skull. Roentgenograms of one wrist at 21 months of age revealed metacarpals typical of a 36-month-old and a radius typical of a 30-month-old. Metabolic screening of her urine revealed the absence of sulfur-containing amino acids, indican, mucopolysaccharides, ketostix, ferric chloride, and reduced dinitrophenol. The results of the IgA were 21 mg%; of the IgM, 131 mg%; and of the IgG, 460 mg%. According to audiological tests, she responded to speech as well as cessation of activity and could hear sounds at the thirtieth decibel. She could not be conditioned to respond to pure tones; therefore, the possibility of hearing deficit could not be ruled out. Electroencephalographic records obtained without sedation were mildly abnormal, showing an occasional spike of the left frontal region and a lower amplitude than expected for her age. Psychomotor evaluation at 21 months of age resulted, according to the Bailey Scales of Infant Development, in a mental development index of 45 and a psychomotor development index of 38. Thus, her mental performance was at the level of a nine-and-one-half-month-old and her motor performance at the level of an eight-month-old. Both her father and her mother are in good health. There is no history of mental retardation or physical malformation in the family of either parent.

CYTOGENETIC STUDIES

Chromosomes were prepared from peripheral blood of the patient according to a modification of the method of Moorhead et al. [5]. All of the cells examined lacked a C group and a no. 18 chromosome, but had an additional large metacentric chromosome and a small metacentric marker. The large metacentric was morphologically similar to the no. 3 chromosomes, although it was slightly shorter in most metaphases. The small marker chromosome was less metacentric. Its short arm was essentially the same length as the short arm of the remaining no. 18 chromosome. The combined length of the large metacentric and the marker was essentially the same as that expected for the combined length of an X and the no. 18 chromosome.

Autoradiographic studies were performed by following a technique essentially like that of Schmid [6]. Sixty-five metaphases displaying a single very heavily labeled chromosome were photographed for analysis prior to removal of the autoradiographic film. In 51 of these 65 metaphases, the large metacentric was identified (after removal of the film) as the heavily labeled (late-replicating) chromosome (fig. 2). Labeling was consistently heavy throughout the entire length of both chromosome arms in these cells. In the remaining 14 metaphases, a chromosome assumed to be the normal X was the late-replicating chromosome, while the large metacentric showed little or no labeling (fig. 3). The labeling of the small marker was quite varied; in some cells it was relatively heavy, in others light or even lacking.

From both the clinical and the cytogenetic findings, the karyotype of the patient was interpreted to be the result of a reciprocal translocation involving the interchange of a large segment of the long arm of a no. 18 chromosome with

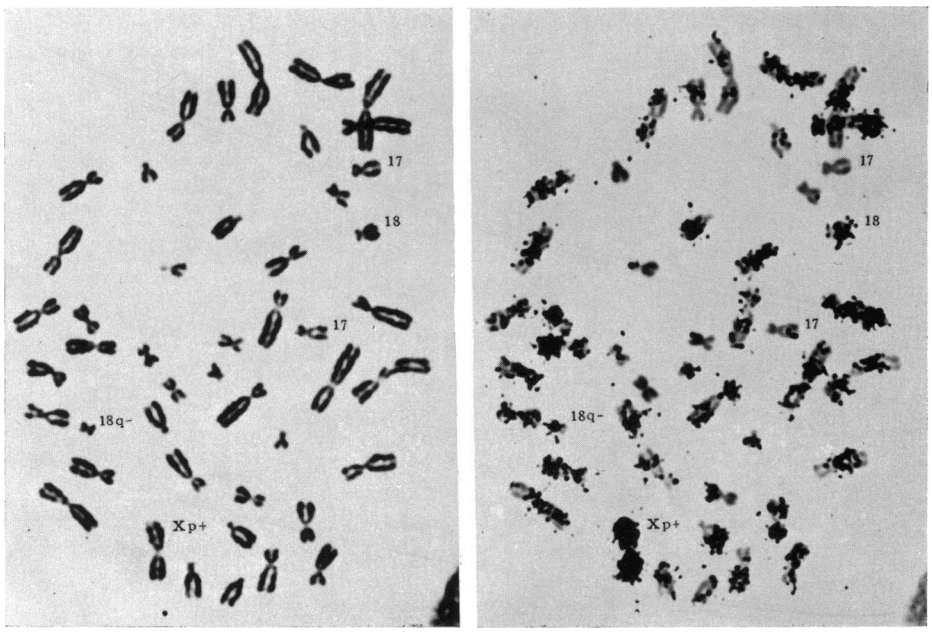

FIG. 2.—Leukocyte metaphase plate from the patient showing heavy labeling of the abnormal X chromosome (Xp+).

a small segment of the short arm of an X chromosome [46,Xt(Xp+;18q—)]. Chromosome preparations of the parents were normal.

DISCUSSION

Balanced carriers for translocations between autosomes are usually phenotypically normal, since little or no change has occurred in either the amount of genetic material or the pattern of genetic activity. This does not appear to be the case, however, for translocations that involve the X chromosome. For example, Russell [7] has shown that in mice, balanced carrier females of X/autosome translocations regularly display variegated-type position effects, and that these position effects are due to genetic inactivation of the translocated autosomal segment. The inactivation does not involve the entire translocated piece of the autosome, but spreads along a gradient to limited distances. Similar position effects have also been found [8] in mice heterozygous for Cattanach's translocation.

Phenotypic effects might also be expected in human female carriers, for there is evidence that genetic material translocated to the X chromosome can be inactivated. In both the patient reported by Lie et al. [1] and the patient reported by Mukerjee and Burdette [2], the segment translocated to the X assumes the same late-replicating pattern as the X chromosome to which it became attached (the inactivated X). Little if any of the segment translocated to the X is late replicating, however, in the patient reported by Neuhauser and Back [3] or the

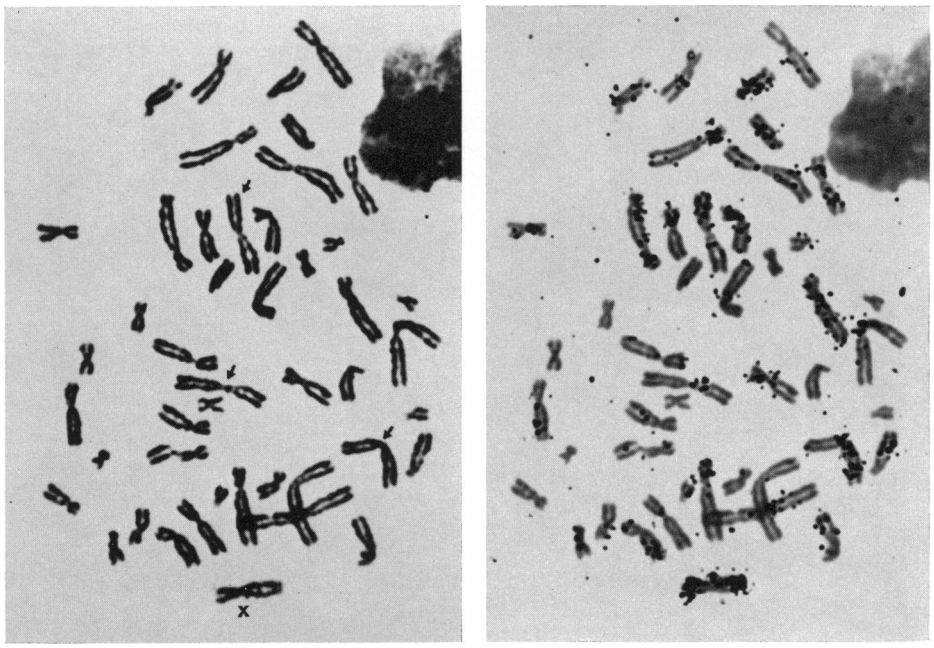

FIG. 3.—Leukocyte metaphase plate from the patient showing heavy labeling of the normal X. The abnormal X (not distinguishable from the no. 3 chromosomes) shows light labeling.

patient reported by German [4]. None of these four patients is a balanced carrier. Furthermore, their abnormalities are not a consequence of inactivation.

Since there was no apparent loss of genetic material in the translocation, we believe that our patient is a balanced carrier—in fact, the first reported case of a balanced X/autosome translocation carrier in man. We also believe that her abnormalities are the result of inactivation of that portion of the long arm of chromosome 18 translocated to the X chromosome. Although proof of inactivation at the genetic level cannot be provided, the pattern of late replication throughout the entire abnormal X suggests that inactivation has occurred. Additional evidence of inactivation can be found by comparing the abnormalities in our patient with those in patients with the 18q— syndrome (table 1). As can be seen in the table, many defects characterizing the deletion of the long arm of chromosome 18 are found in our patient.

Interpretations other than that of a reciprocal translocation are possible. The karyotype could be the result of a nonreciprocal translocation involving the transposition of much of the long arm of the no. 18 chromosome onto the short arm of one of the X chromosomes, together with the subsequent loss of a short terminal fragment of the X which arose as a result of the translocational process. Although arm-length measurement studies did not provide evidence to support this interpretation, the loss of such a small chromosomal segment (the terminal fragment of the X) would be difficult to detect (only if the translocation had been

TABLE 1

COMPARISON OF THE ABNORMALITIES FOUND IN OUR PATIENT WITH THOSE REPORTED MOST FREQUENTLY IN 18q— PATIENTS REVIEWED BY CENANI ET AL. [9] AND LAFOURCADE AND LEJEUNE [10]

Abnormality	Frequency of Trait in 18q— Patients Reviewed by Cenani et al. [9]*	Frequency of Trait in 18q— Patients Reviewed by Lafourcade and Lejeune [10]*	Our Patient
Mental retardation	21/23	18/19	+
Somatic hypotrophy	20/24	16/18	—
Microcephaly	20/21	15/17	+
Hypotonia	19/23	...	+
Mid-face dysplasia	18/19	12/15	+
Hypertelorism	15/15	...	+
Epicanthus	11/15	...	—
Carp-shaped mouth	14/16	9/12	+
Low-set ears	12/17	...	—
Prominent antihelix and/or antitragus	15/17	19/19	+
Hearing defects	12/20	...	?
Heart defects	10/21	...	—
Cox valga	7/8	...	—
Spindle fingers	7/9	11/11	+
Defects of the palate	9/15	...	+
Ophthalmological defects	18/23	14/21	+
Axial triradius t'	9/9	...	+
Multiple digital whorls	14/18	15/16	—
Genital abnormalities	7/12	...	—
Pits below the acromion	...	8/10	+

* Some patients are included in both reviews.

carried by the father would one be able to rule out this sort of a nonreciprocal event). In this interpretation, the autoradiographic and clinical evidence still suggests that inactivation, rather than the loss of the terminal fragment of the X, is responsible for the phenotypic abnormalities.

The karyotype could also be the result of an insertion of a segment of the long arm of chromosome 18 into the short arm of an X. Although a greater number of chromosome breaks are required to have an insertion, this interpretation is somewhat attractive in that there need not be a loss of genetic material. Again, in this interpretation the evidence suggests that the phenotypic abnormalities are the result of genetic inactivation.

Finally, the karyotype could be interpreted as the result of both a deletion of the long arm of chromosome 18 and the formation of an isochromosome (46,XX,qi,18q—). The clinical findings, however, do not support this interpretation. For example, in contrast to all of the iso-X patients reported by both Lindsten [11] and Engel and Forbes [12], our patient is not excessively short (her height is above the eighty-fourth percentile) nor does she have an increased number of pigmented nevi. Furthermore, other associated characteristics such as webbing of the neck, cubitus valgus, low-set ears, and blue sclerae are absent. The autoradiographic findings also fail to support this interpretation. In patients

with a true X isochromosome, the late-replicating chromosome is the same in all cells—always the metacentric (the iso-X) and never the normal X [13–16]. In our patient the normal X is the late-replicating chromosome in 14 of 65 selected cells.

The fact that the normal X was inactivated in some cells was unexpected in view of the findings concerning other patients with structurally abnormal X chromosomes. In patients with ring X chromosomes [17–19], deletions of the X [20–24], translocation-bearing X's [1–4], and X isochromosomes [13–16], only the abnormal X is late replicating. Occasionally, neither X is late replicating, as in patients with small ring X's [25, 26] or fragmented X chromosomes [27, 28]. Rarely, however, is the normal X late replicating in even a small percentage of cells containing a structurally altered or abnormal X chromosome. Our patient and a phenotypically normal woman with a satellited X reported by Lubs [29] are apparently exceptions.

Although late replication of the normal X in man is unusual, it is not totally unexpected when one considers the findings in other mammals. In mice heterozygous for Cattanach's translocation (an insertion of an autosomal segment into the X), the normal and the translocation-bearing X are genetically inactivated in approximately the same proportion of cells [30]. Autoradiographic studies have verified this essentially random inactivation [31]. In mice heterozygous for Searle's translocation (a translocation of a portion of the X to an autosome), the opposite of that normally occurring in man is realized, for in these mice only the normal X is inactivated [32]. Essentially the same situation has been found in a domesticated fertile cow carrying an X/autosome translocation [33]. In all bovine cells examined, only the normal X was the "hot" or late-replicating chromosome. Seemingly, the particular X which is inactivated and the number of cells it is inactivated in depend upon both the type of change occurring and the species in which the change occurs.

If only the normal X were inactivated in man (as in mice heterozygous for Searle's translocation), many translocations involving the X could not be distinguished from those which involve other C group chromosomes, since identification of the X depends on its being late replicating. Some previously reported C group translocations may involve an X chromosome. If the normal X had been inactivated in all the cells of our patient, it is unlikely that abnormalities would have been present, and therefore that she would have come to our attention. Other X/autosome balanced carriers may soon be found who are phenotypically normal.

The prognosis for our patient is obviously poor, although it may be somewhat better than that for a patient with a deletion of the long arm of chromosome 18; for, at least in some cells (those with an inactive normal X), the translocated autosomal segment is not inactivated. If the inactivated autosomal segment were reactivated in the remaining cells, the prognosis would undoubtedly improve. Occasional reactivation does occur in mice heterozygous for Cattanach's translocation [8]. However, judging from the limited degree of reactivation in these mice, it seems unlikely that such an effect will be of much benefit to our patient.

SUMMARY

Chromosome studies of a child with abnormalities suggestive of the 18q— syndrome were carried out. The studies showed that the long arm of a no. 18 chromosome had not been deleted but had been translocated to a chromosome later identified autoradiographically as an X. Because of its position, the translocated segment apparently was inactivated along with the X to which it had attached. The abnormalities exhibited by the child were interpreted to be the result of this inactivation.

While the late-replicating chromosome was identified as the translocation-bearing X in most cells selected from the patient, in some cells the normal X was late replicating. Although inactivation of either the normal or the abnormal X occurs in mice, such an occurrence in man is quite unique.

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