The XXXXY Sex Chromosome Abnormality

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Over the past few years several cases have been reported concerning phenotypical males with hypoplastic genitalia, skeletal abnormalities, and mental retardation, who have also had one or more sex chromatin bodies in the cells of the buccal mucosa and in other tissue cells, as well as other less constant features. They have been grouped under the heading of Klinefelter's syndrome.

Jacobs and Strong (1959) first described the common form of the XXY-karyotype. Since then other variants have been encountered, e.g. XXXY, XXYY, and XXXXY aneuploids, in addition to various mosaic complements such as XY/XXY, XXXY/XXXXY.

Fraccaro, Kaijser, and Lindsten (1960) described a 7-year-old boy with small testicles and ambiguous genitalia, who was mentally retarded and had a modal chromosomal count of 49. Later Fraccaro, Klinger, and Schutt (1962a) and Fraccaro, Lindsten, and Kaijser (1962b) interpreted the karyotype as sex chromosome abnormality XXXXY. That this chromosomal abnormality is the basis of a specific syndrome was suggested by Fraser, Boyd, Lennox, and Dennison (1961).

Similarly affected males were described by Anders, Prader, Hauschteck, Schärer, Siebenmann, and Heller (1960), in whom the bone-marrow cells showed 3, 4, and 5 X chromosomes, the majority possessing 4 X chromosomes. Miller, Breg, Schmickel, and Tretter (1961) also presented a patient with a sex chromosome complement: 4 X and 1 Y. Further cases have been reported by Maclean, Mitchell, Harnden, Williams, Jacobs, Buckton, Baikie, Court Brown, McBride, Strong, Close, and Jones (1962), Fraccaro *et al.* (1962a, b), Barr, Carr, Pozsonyi, Wilson, Dunn, Jacobson, Miller, Lewis, and Chown (1962), Turpin, Lafourcarde, Cruveiller, Lejeune, Bocquet, Hoppeler, and Guibert (1962), Pfeifer (1962), Lamy, Aussannaire, de Grouchy, and Lalande (1963), Schade, Schöller, and Töberg (1963), Atkins, Böök, Gustavson, Hansson, and Hjelm (1963), and Scherz and Roedel (1963).

In addition, the chromosome abnormalities were

discussed by Ferguson-Smith, Lennox, Mack, and Stewart (1957), Ferguson-Smith (1958), Bergman, Reitalu, Nowakowski, and Lenz (1960), and Day, Levinson, Larson, and Wright (1963).

Case Report

C.P. was born on April 18, 1950, at full term, weighing 5 lb. 2 oz. (2,324 g.), by uncomplicated forceps delivery. The mother was said to have been admitted to hospital for treatment of carbon monoxide poisoning during the third month of pregnancy, when she lost consciousness but recovered rapidly and apparently showed no residual symptoms.

The child first sat alone at the age of 9 months and cut his first tooth at 13 months. At 2 years of age he had pneumonia, and, during the doctor's visit, a diagnosis of mental retardation was made.

At 3 years he could not speak or walk alone; he was not toilet-trained nor could he dress himself. The clinical impression at this time was 'mental retardation, severe'. He was at a neurology clinic the following year, at which time the clinical impression was 'cerebral palsy', probably a mixture of pyramidal and extrapyramidal involvement, the feet being particularly involved. Mental deficiency was obvious but the level of intellectual defect could not be estimated.

He did not walk unsupported until the age of 4 years. At about this time he had measles and chicken-pox complicated by bronchopneumonia, and at the age of 6 years he had a bout of bronchitis. At the age of 7 the IQ was 54.

From 1961 to 1963 he appeared to regress somewhat, and another IQ test showed a slight relative decrease in his intelligence. He was reported to be uncooperative and a disrupting influence to the rest of the class, being apprehensive, tense, and explosive, and he was considered incapable of continuing in school. In January 1964 his IQ was below 45 and an institution was recommended.

He was seen at the Pineland Hospital and Training Centre where a diagnosis of 'Encephalopathy, due to unknown or uncertain cause with structural reaction manifest', was made, and at this time he was placed in the group of Klinefelter's syndrome. His Standard Binet (LM) IQ was 43. (He was 14 years of age.)

Physical examination revealed a thin white male with disproportionately long lower extremities and long tapering fingers. His height was 61.5 in. (156 cm.); he had a moderate degree of prognathism, a short 5th finger

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FIG. 1.—Note prognathism, kyphosis, and disproportionately long extremities.

FIG. 3.—Both hands show long tapering fingers.

on both hands, with incurving of the fingers towards the midline, and both big toes were clubbed (Fig. 1-3). He was short-sighted, his temperature 98.6° F. (37° C.), blood pressure 100/70 mm. Hg.

Language consisted of short sentences which were spoken with repeated omissions and distortions, partly due to a double row of teeth in the mandible and to his prognathism.

He was dolichocephalic, had a few scars from a dermal infection in the buttocks and legs, and the hair appeared normally implanted. The pupils were round and equal and reacted to light and to accommodation. The jaw was protruding and showed a double ridge of teeth.

FIG. 2.—'Clubbing' of the large toes.

FIG. 4.—Genitalia: the uncircumcised penis is small, as is the right testicle within the scrotum. The left testicle is undescended.

The penis was quite small and uncircumcised (Fig. 4). The right testicle was in the scrotum, approximately bean-sized, the left testicle was not palpable.

Girth measurements were symmetrical, but the lower limbs were quite thin and long, and the left lower extremity was shorter than the other by 0.25 in. (0.6 cm.). When walking, he tended to bear weight on the right leg during the stance phase of ambulation with the trunk listing to the right.

Extension of the elbow was normal; the flexion, however, lacked 20 degrees bilaterally. There was hypermotility in supination of the forearm bilaterally, more on the right side. Hand grips were equal and muscle

strength was within normal limits. Pronation of the forearms was limited, more marked on the left side than on the right. The limitation of elbow flexion and forearm pronation made it difficult for him to approximate fingers to shoulder. Even though his elbow extension was normal, the arms were held in the position of abduction of 35 degrees at the shoulder and 35 degrees flexion at the elbow joints. However, radiograph of both elbows showed no evidence of radio-ulnar synostosis.

There was a severe lordosis and rotary scoliosis of the lumbar spine with convexity directed to the right, associated anterior pelvic tilt, protrusion of the abdomen, and winging of the scapulae. On walking, the arms continued to be held in an abducted position with slight flexion, though arm swinging was present. There was a bilateral genu valgum, more severe on the right.

There was a noticeable tremor in all extremities, the trunk, the facial muscles, and especially the lips. This tremor occurred in a rhythmic fashion and became slightly exaggerated in the hands when fine movements were attempted.

The electroencephalogram revealed a very high voltage, 9 per second alpha, a little more prominent occipitally; however, there were symmetrical bursts of high voltage sharp spikes. The impression was of a seizure or interseizural tracing with a suggestion of abortive *petit mal* (Dr. Maltby).

The chest radiograph showed the presence of bilateral cervical ribs, and the heart shadow was described as borderline, possibly slightly enlarged. Radiological examination of the skull showed normal development of the calvarium with some occipital asymmetry. The sella turcica appeared quite large and somewhat elongated. There was premature eruption of teeth with a double row in the mandible. The palatine arch was flat and splayed.

Urinanalysis and blood count were within normal limits, the pituitary gonadotropins were 164 HMGU/24 hr. The normal prepubertal level is less than 3 HMGU.

The electrocardiogram showed high voltage in leads II, III, AVF, V1, V2, V4, of the QRS complex, which raised the possibility of left ventricular hypertrophy (Dr. Davidsohn).

The serum electrophoresis showed a normal pattern; total protein 7.7 g./100 ml., albumin 5.16/100 ml.; α_1 0.06, α_2 0.78, β 0.70, γ globulins 1.00 g./100 ml. Sodium 145 mEq/l., potassium 4.4 mEq/l., CO_2 26 mEq/l., phosphorus 5.2 mg./100 ml., calcium 10.5 mg./100 ml., alkaline phosphatase 14 KA units; blood group ACcDEeMN; Fy^a negative, Kell negative.

Buccal mucosa smears were studied in fresh preparations stained with aceto-orcin and in fixed preparations stained with Biebrich red-fast green, thionin tartaric acid, and cresyl violet. Only heterochromatic bodies immediately adjacent to the nuclear membrane were counted as Barr bodies. Approximately 60% of the cells were sex chromatin positive; of these 52% showed 1 body, 39% 2 bodies, and 9% 3 bodies (Fig. 5).

A chromosomal analysis of metaphase cells from peripheral blood leucocytes was carried out using a slight modification of the technique described by Moorhead,

FIG. 5.--Buccal mucosa nucleus with 3 sex chromatin bodies.

Nowell, Mellman, Battips, and Hungerford (1960). The modification consisted in treatment of the cell suspension with 1% trypsin for 15 minutes just before application of the hypotonic solution. 50 cells were counted under the microscope, 46 cells with 49 chromosomes, 2 with 46, and 2 with 47 chromosomes. Since, in all of the latter, 4 different autosomes were found missing in each of these cells, they were taken to represent artefacts. 10 well-spread groups of chromosomes, each from a single cell, were photographed and enlarged, and the count of 49 chromosomes was verified in each instance. Of the chromosomal spreads, 2 were enlarged and karyotyped, and in both instances the additional 3 chromosomes were found to be about the size of those of the 6-12 group of the Denver classification, this point being verified by an independent investigator (Fig. 6). These supernumerary chromosomes were taken to represent X chromosomes and were thus labelled. Although final confirmation with tritiated thymidine was not carried out, the presence of 1, 2, and 3 Barr bodies in the buccal mucosa cells tends to support this interpretation. Although the pattern of distribution of the Barr bodies in the epithelial cells would be suggestive of mosaicism, we were unable to demonstrate it in the cultures of peripheral blood.

The leucocyte culture from the father of the proband revealed a normal count of 46 chromosomes without abnormal autosomes or sex chromosomes. We were unable to persuade the mother to submit to an identical examination.

Discussion

There have been only 16 other reported cases of the XXXXY chromosome complex. The origin of this abnormal chromosomal constitution still remains a matter of conjecture, though several possibilities have been postulated.

The individuals afflicted with this chromosomal aberration have all been mentally retarded, most of them with an IQ below 50, as in our case. They have had associated skeletal abnormalities, among

FIG. 6.—XXXXY karyotype.

which the most commonly encountered have been disproportionately long lower extremities, radio-ulnar synostosis, incurved phalanges, cleft palate, elongation of the radii, abnormal ossification centres, scoliosis, prognathism, etc. Congenital heart disease, myopia, pes planus, epicanthic folds, slanting eyes, and deformities of the chest have also been described, but these features are not encountered frequently enough to be considered of diagnostic significance.

Histological study of testicular biopsies in the cases of Barr *et al.* (1962) and Atkins *et al.* (1963) revealed dysgenesis of testicular germ cells and tubules which led to fibrosis.

The most constant and significant laboratory finding has been the increase in pituitary gonadotropins, which in normal subjects is found around the time of puberty, but which in the condition under discussion is not an unexpected finding, in view of the previously cited pathological changes seen in the testicles.

The protein-bound iodine and other laboratory findings have been normal as have the electroencephalographic tracings, though, the latter were abnormal in our case.

Thus far no correlation has been established between the occurrence of the syndrome and the age of the parents, though such a suggestion is mentioned by Atkins *et al.* (1963), in whose case the mother of the propositus was 39 years old at the time of delivery.

The mechanism invoked for the production of the XXXXY complex can only be one of non-disjunction and several types of non-disjunction have been proposed.

(a) Double non-disjunction in maternal and paternal gametogenesis, resulting in either an XX ovum and XXY sperm, or XXX ovum and XY sperm.

(b) The occurrence of a single non-disjunction in a fertile female with an XXX sex chromosome pattern.

(c) If by non-disjunction a fertilized egg XXY were produced, the X chromosomes would undergo mitotic non-disjunction in the first cleavage division, and one of the two resulting daughter cells would contain the XXXXY complex while the other containing the Y chromosome only would be lethal and would not contribute to the somatic constitution of the embryo.

(d) Non-disjunction in both the first and the second meiotic division during the mother's gametogenesis, resulting in an XXXX ovum which would be fertilized by a Y sperm (Fraser *et al.*, 1961).

Until now, there is lack of support for any of the above-mentioned possibilities; however, the possibility of B seems the least likely since no triple X mother of an XXXXY son has been described. We agree with Scherz and Roeckel (1963) that maternal double non-disjunction resulting in an XXXX ovum is the most likely.

Lyon (1962) postulated the inactivation of one of the two X chromosomes of normal females, occurring early in development, and that the sex chromatin body in interphase nuclei is precisely this inactivated X. She further suggested that, in cases of anomalies concerning the number of sex chromosomes, all the X chromosomes in excess of one would be inactivated. However, the fact that multiple X karyotypes, particularly the quadruple X, are always associated with several somatic anomalies and testicular atrophy, suggests that inactivation is only partial, as has been suggested by Grumbach (1963), and there is some influence at work of chromosomal environment on the individual's somatic constitution.

Summary

A 14-year-old phenotypical male with small genitalia, long slender extremities, prognathism, and clubbed large toes is described. The buccal mucosa cells showed 1, 2, and 3 sex chromatin bodies, and the chromosomal count was 49, with the karyotype XXXXY and without evidence of mosaicism.

The clinical symptomatology and genesis of this sex chromosome anomaly are discussed.

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