

January 1, 1967
 Am. J. Obst. & Gynec.

Premature menopause in XO/XX/XXX/XXXXX mosaicism

703771

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A patient with presumed XO/XX/XXX/XXXXX mosaicism presented at the age of 17 with symptoms of the menopause. Increased urinary gonadotropin titers, decreased urinary estrogen levels, and lack of ovarian follicles on histologic examination confirmed the state of ovarian insufficiency. It is suggested that chromosome studies should be carried out in patients with premature menopause so that the role of sex chromosomal abnormalities in the pathogenesis of this entity may be properly delineated.

PREMATURE menopause has been defined as the failure of normal ovarian function prior to age 40 in the presence of low urinary estrogen levels, elevated urinary gonadotropin titers, and vasomotor symptoms. In most instances, the etiology of this condition has remained unknown.^{1, 2, 3} The possibility that sex chromosomal abnormalities may be one of the factors in the pathogenesis of this entity was raised by the account which documented the finding of an XXX chromosomal pattern in a woman with premature menopause.⁴

This report describes a patient with premature menopause who was found to have an unusual form of sex chromosomal mosaicism.

Case report

N. S., a 19-year-old married female, was admitted to the United States Public Health Service Hospital in May, 1964, because of premature menopause. At the age of 12 she developed pubic and axillary hair, menses, and breast enlargement. For the next 5 years her menses occurred regularly about every 21 days. At age 17 she became amenorrheic. Within 6 months the onset of episodic hot flashes with typical flushing and sweating were noted. Subsequently, she complained of nervousness, anxiety, and generalized headaches. Cyclic diethylstilbestrol treatment relieved her symptoms and resulted in periodic vaginal bleeding. When estrogen therapy was discontinued, spontaneous menses did not return.

The patient had had a left mastoidectomy and skin graft procedure at age 17. There was no family history of abnormal pubertal development. She had two sisters (3 and 10 years of age) and three brothers (12, 17, and 24). The

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Supported in part by Grants AM-05436 and TI AM-5161, National Institutes of Health, and Grant AT(45-1)-1781, Atomic Energy Commission.

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Table I. Frequency of sex chromatin in cell nuclei from buccal and vaginal mucosa and ovarian tissue

| Origin of cells examined | No. of Barr bodies | | | | | | | | | | Total No. |
|--------------------------|--------------------|------|-----|------|-----|-----|-----|---|-----|---|-----------|
| | 0 | | 1 | | 2 | | 3 | | 4 | | |
| | No. | % | No. | % | No. | % | No. | % | No. | % | |
| Buccal mucosa | 54 | 40.6 | 69 | 51.9 | 10 | 7.5 | 0 | 0 | 0 | 0 | 133 |
| Vaginal mucosa | 89 | 44.5 | 101 | 50.5 | 8 | 4.0 | 0 | 0 | 2 | 1 | 200 |
| Ovarian tissue | 91 | 45.5 | 100 | 50.0 | 9 | 4.5 | 0 | 0 | 0 | 0 | 200 |

Table II. Chromosomal analysis of peripheral blood culture

| | No. of chromosomes | | | | | | | Total |
|--------------------------------|--------------------|----|----|-----|----|-------|-----|-------|
| | <45 | 45 | 46 | 47 | 48 | 49 | >49 | |
| Total cells counted | 3 | 16 | 59 | 12 | 0 | 1 | 0 | 91 |
| Total cells karyotyped | 1 | 13 | 12 | 10 | 0 | 1 | 0 | 37 |
| Autosomal (random) abnormality | 1 | 2 | 0 | 2 | 0 | 0 | 0 | 5 |
| X chromosome abnormality | 0 | 11 | 12 | 8 | 0 | 1 | 0 | 32 |
| Sex chromosome constitution | | XO | XX | XXX | | XXXXX | | |

oldest brother was married with 3 apparently normal children. The patient's father died of tuberculosis and her mother had a history of peptic ulcer. A maternal uncle and grandfather have diabetes mellitus.

Physical examination revealed a well-nourished, white female of average intelligence, with normal breast development associated with normal hair and fat distribution. Her height was 5 feet 3 inches and she weighed 115 pounds. Positive physical findings included multiple spider nevi on both forearms, arms, shoulders, neck, and upper chest, short fifth fingers, and the absence of the ear lobule bilaterally. Pelvic examination revealed normal external genitals and uterus. The ovaries were not palpable.

Complete blood count, urinalysis, VDRL, BSP, alkaline phosphatase, serum cholesterol, protein-bound iodine, and roentgenograms of the chest and skull were all within normal limits. Total urinary estrogens measured by the immature rat uterine weight method⁶ on three different occasions were < 0.3 μ Eq of estradiol benzoate per 24 hours. (Normal values range from 0.3 to 10.0 μ Eq in ovulatory women.) Total urinary gonadotropins determined by the immature rat ovarian weight method⁶ on three different occasions were 4.9, 9.0, and 9.6 mEq. UPM-1*

*UPM-1, a postmenopausal gonadotrophin preparation, supplied by the Endocrine Study Section of the National Institutes of Health, was used as our bioassay standard.

per 24 hours. (Normal values range from 0.15 to 2.3 mEq. in ovulatory women.) Roentgenograms of the lumbar spine revealed an anomalous fifth lumbar vertebral body and enlarged right articular transverse body. Roentgenograms of the hands and wrists revealed a small fifth middle phalanx bilaterally. The metacarpals were normal. The carpal angle was normal at 127 degrees.⁷ Seven and one-half per cent of cells on buccal smear contained two sex chromatin masses (Table I).

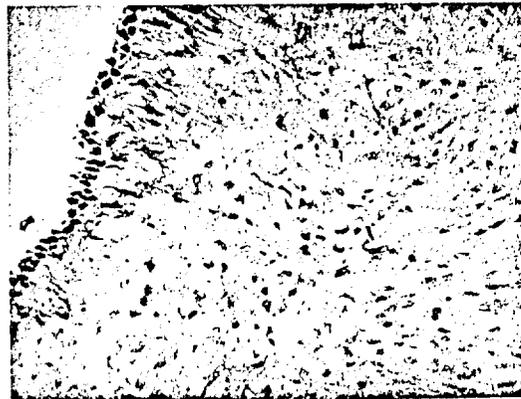


Fig. 1. Photomicrograph of ovarian biopsy specimen. Note the absence of follicles and corpus albicans. ($\times 160$.)

Figs. 2A-2D. Karyotype analysis from peripheral blood of Patient N. S.

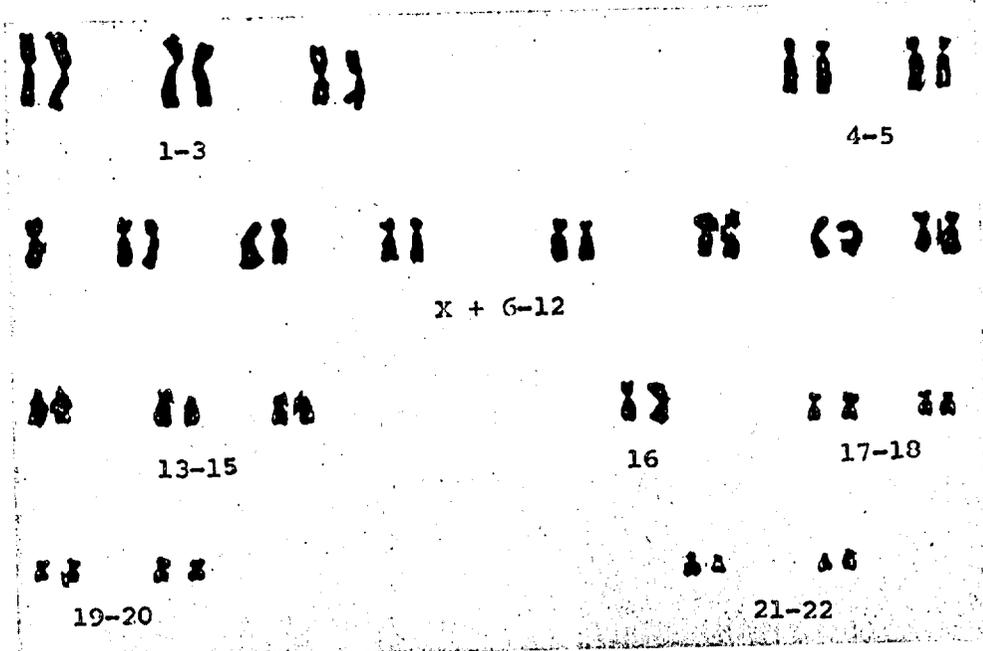


Fig. 2A. There are 45 chromosomes—XO configuration.

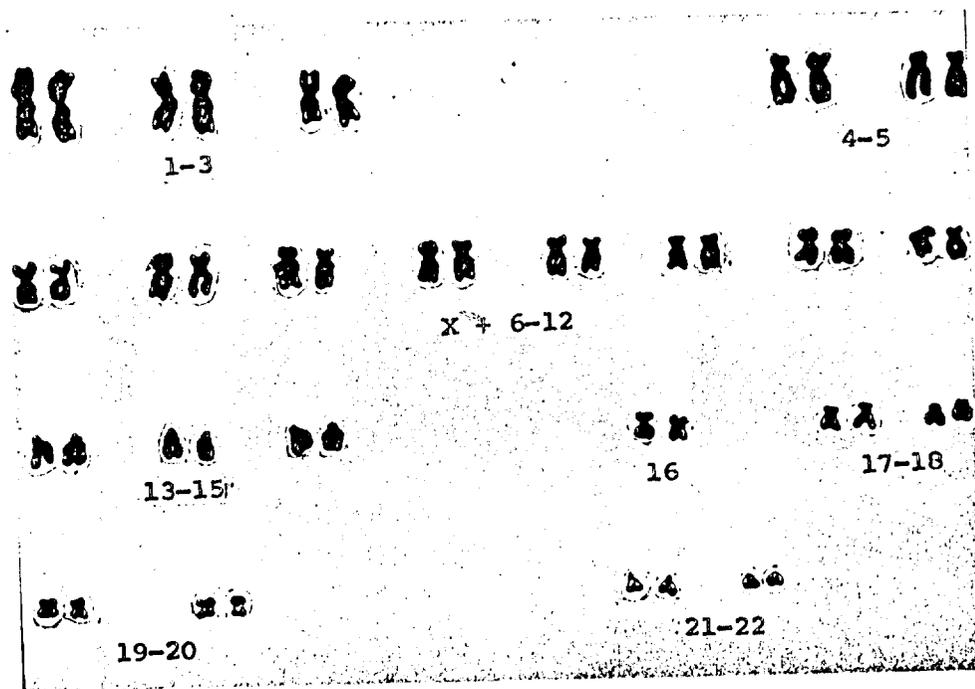


Fig. 2B. There are 46 chromosomes—XX configuration.

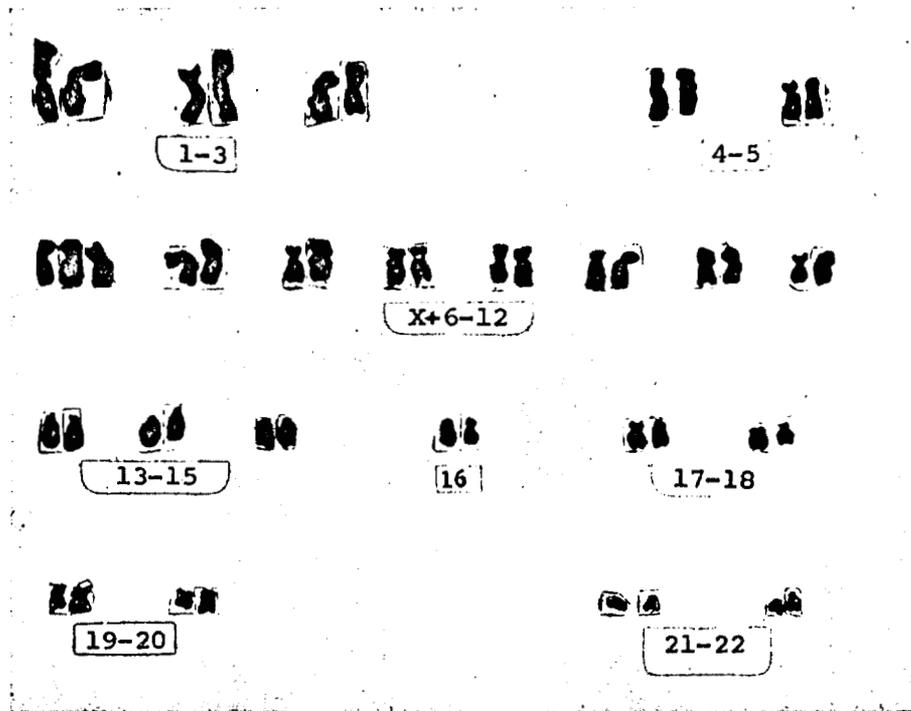


Fig. 2C. There are 47 chromosomes—XXX configuration.

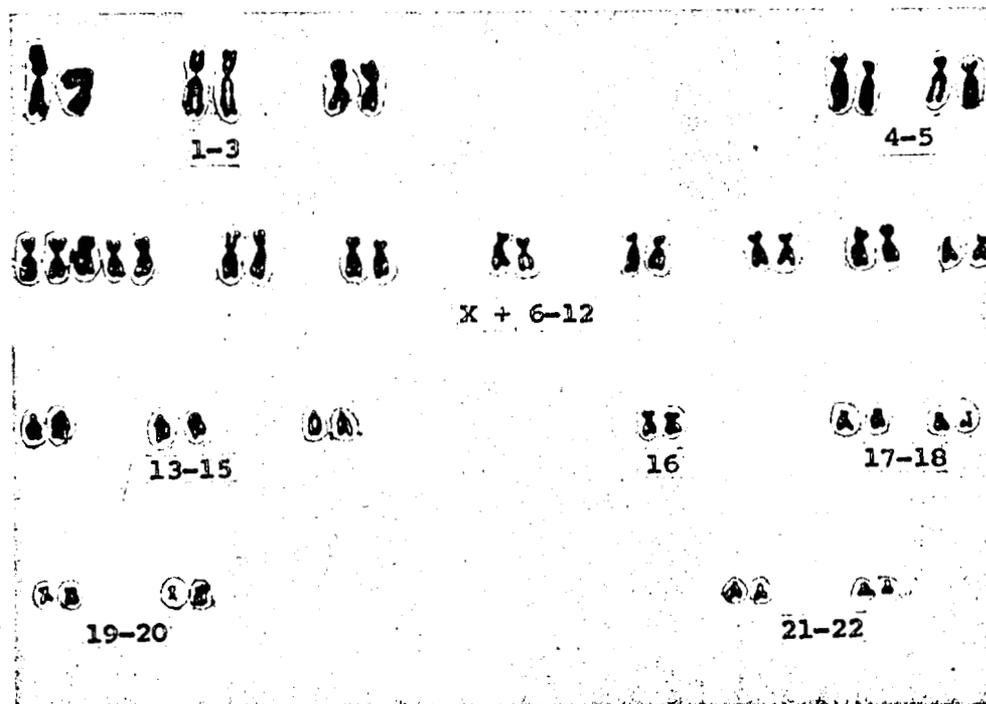


Fig. 2D. There are 49 chromosomes—XXXXX configuration. Short arm of chromosome No. 12 is overlapping the centromere and a long arm.

On May 6, 1964, an exploratory laparotomy was performed because the patient desired knowledge as to the possibility of becoming pregnant. The right ovary measured $1 \times 1 \times 1$ cm. and contained a small cyst. The left ovary measured $3 \times 0.5 \times 0.5$ cm. The uterus was normal in size and appearance. Tissue was obtained by incisional biopsy from each ovary for microscopic examination and chromosome analysis. Histologically, the ovarian stroma was hypocellular, with a rare corpus albicans and no maturing follicles. The sections were similar histologically from each gonad and were interpreted as representing an involutinal ovary (Fig. 1). Unfortunately, the ovarian cells did not grow in tissue culture so that chromosomal analysis was not possible.

Results of sex chromatin (Barr body) counts from cells of the buccal mucosa, vagina, and ovary are depicted in Table I. Karyotype analysis of the white blood cells grown in tissue culture by a modification of the method of Moorehead and associates⁸ is shown in Table II and Figs. 2A through 2D. These data suggest the presence of XO/XX/XXX/XXXXXX mosaicism.

The patient had an uneventful postoperative course. She was placed on diethylstilbestrol, 0.5 mg. daily, and was subsequently lost to follow-up.

Comment

Gonadal damage is frequently found in subjects with X chromosomal abnormalities. In patients with an XO sex chromosomal pattern the pathology generally occurs during the development of the gonad before differentiation has occurred.⁹ The clinical result is a phenotypic female who lacks pubertal development. This differs from the triploid X syndrome in which the phenotypic female may have normal ovarian function with normal fertility.¹⁰⁻¹³ When gonadal pathology is present in these subjects, it occurs after ovarian differentiation, with some degree of ovarian function still possible.^{4, 14} Therefore, these patients may develop secondary sexual characteristics and later present with secondary amenorrhea or premature menopause. Indeed, this is the clinical picture of the subject of this report. Ovarian damage was inferred by the history of menopausal symptoms at age 17 and the

laboratory finding of elevated urinary gonadotropin titers. It was confirmed on histologic examination of the ovary which demonstrated an absence of follicles and only a rare corpus albicans. Although the cells of the ovary did not grow in tissue culture, sex chromatin analysis of the nuclei from the histologic sections revealed a 4.5 per cent incidence of ovarian cells containing two Barr bodies. Since no nuclei with more than two sex chromatin masses were observed in the ovary, it is presumed that a population of cells with no more than three X chromosomes was present. Since karyotype analysis of ovarian tissue chromosomes was not possible, we cannot define the exact X-chromosomal configuration present in her ovarian tissue.

Such an analysis on the patient's white blood cells revealed three major sex chromosomal patterns, i.e., XO, XX, XXX. In addition, one cell contained 49 chromosomes with 19 of these belonging to the X + 6-12 group. This was interpreted to be consistent with the presence of five X chromosomes. Since two cells on Papanicolaou smear contained four Barr bodies (Fig. 3), this is additional evidence for the presence of a small population of cells with five X chromosomes.

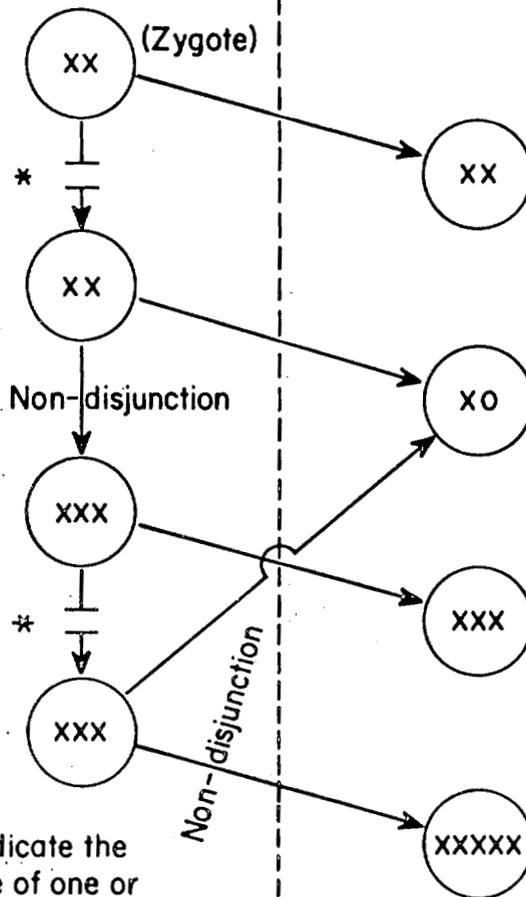
Several possibilities exist to explain the cytologic events that resulted in this type of sex chromosomal mosaicism. The simplest mechanism takes into account two separate episodes of nondisjunction (Fig. 4) in the postzygotic period, so that the end result is four stem-cell lines (XO/XX/XXX/XXXXXX). This is the first reported case of this type of mosaicism.



Fig. 3. Photomicrograph of cell from buccal mucosa of Patient N. S. Arrows depict the four Barr bodies.

INITIATION OF ABNORMAL STEM CELL LINES

CONTINUOUS PROPAGATION OF CELL LINE



* Brackets indicate the occurrence of one or more cell divisions.

Fig. 4. Suggested derivation of the XO/XX/XXX/XXXXX mosaicism in Patient N. S.

We urge that chromosome analysis be performed in patients with "spontaneous premature menopause" in order to clarify the role of sex chromosomal abnormalities in pathogenesis of this entity.

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