56. Chromosome Studies in Twelve Sexually Abnormal Patients^{*)}

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Following the reports on chromosome anomalies of Turner's and Klinefelter's syndromes by Ford *et al.* (1959 a) and by Jacobs and Strong (1959), many cases of sexual anomalies were subjected to chromosomal studies; most of those anomalies have been shown to be associated with abnormalities of sex chromosomes (Harnden and Jacobs 1961). In the present communication are reported chromosome conditions of 12 patients with sex abnormalities. The diagnostic and clinical data of each patient are tabulated in Table I. Chromosome observations were carried out on cells from short-term cultures of leucocytes separated from the heparinized peripheral blood (for technical details, see Makino and Kikuchi 1962). Chromosome analyses are described in accordance with the Denver system of nomenclature for the classification of individual chromosomes. Twelve cases of sex abnormalities under study included four cases of Klinefelter's syndrome (nos. 1, 2, 3, and 4), two cases of hypogonadism (nos. 8 and 9), two cases of hypospadias (nos. 10 and 11), one each of gonadism (no. 5), atresia vagina (no. 6), testicular feminization (no. 7) and monorchism (no. 12). The results of chromosome counts in those patients are presented in Table II.

Two cases of Klinefelter's syndrome (nos. 1 and 2) showed each a consistent chromosome count of 47 (Table II). The karyotype analyses showed the occurrence of five small acrocentric elements in the complement which is suggestive of the existence of a Y chromosome, while 16 medium-sized submedian chromosomes indicated the presence of two X elements among them (Fig. 1). The above findings adequately justify the conclusion that the cells with 47 chromosomes contain two X chromosomes and one Y chromosome, in addition to 44 autosomes. It is then evident that two Klinefelter's syndrome cases here considered have an XXY sex-chromosome constitution characteristic to that syndrome.

In contrast to the above two cases, the other two (nos. 3 and 4), though they were clinically diagnosed as Klinefelter's syndrome, showed in each a normal complement of chromosomes, 46 in number.

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Table I. Clinical and diagnostic features in twelve cases of sexual abnormalities under study

Case No.	Age and sex	Diagnosis	Clinical feature
1	30 yr. ♂	Klinefelter's syndrome	Underdeveloped external genitalia, particularly of testes; right testis: $1.8 \times 0.9 \times 1.2$ cm; left testis: $1.6 \times 1.2 \times 1.1$ cm; impotent; no child during four years of married life; I.Q.: 92; 17-KS: 3.86 mg/day; gonadotropin: 80 muu/day.
2	25 yr. ♂	Klinefelter's syndrome	Gynecomastia; penis normal in size; atrophic testes: $2.2 \times 2.0 \times 2.0$ cm; hypoplastic prostate; normal pubic hairs.
3	32 yr. ♂	Klinefelter's syndrome	Atrophic hyalinized seminiferous tubules; arrest of spermatogenesis; aspermia; normal history of development and puberty; normal penis and scrotum; 17-KS: 3.6 mg/day; gonadotropin: 16 muu/day.
4	10 yr. ♂	Klinefelter's syndrome	Underdeveloped external genitalia, together with small sized testes; 17-KS: 3.6 mg/day; gonadotropin: 4 muu/day.
5	3 yr. —	Agonadism	Tiny clitoris-like protrusion; no gonad and internal genital organ except for bilateral small tubular structures (Wolffian remnants); sex chromatin negative.
6	23 yr. ♀	Atresia vaginae	Primary amenorrhea; defect of vagina; underdevelopment of uterus; small sized ovaries.
7	17 yr. ♀	Testicular feminization	Female type external genitalia; well-developed breast without pubic hairs and axillary hairs; internal organs of male type except for the remnant of vagina; ovaries, oviducts and uterus being not detected; undescended testes; 17-KS: 3.85 mg/day.
8	18 yr. ♂	Hypogonadism	Hypoplastic scrotum and small penis (about 4 cm in length); intra-abdominal testes with atrophic structure; prepubertal mentality; semi-high-pitched voice; no beard; scanty pubic hairing; 17-KS: 41 mg/day; gonadotropin: 32 muu/day; 17-OHCS: 6.1 mg/day.
9	27 yr. ♂	Hypogonadism	Hypoplastic scrotum and penis; eunuchoid proportions; obesity; no pubic hairs; 17-KS: 4.1 mg/day; gonadotropin: 64 muu/day; 17-OHCS: 6.1 mg/day; hypergonadotropic hypogonadism.
10	22 yr. ♂	Hypospadia penoscrotalis	Urethral groove failed to close completely and opened at the base of crooked penis.
11	14 yr. ♂	Hypospadia	Hypospadia; testes being palpable in hypoplastic scrotum; small crooked penis (about 2.5 cm in length); prostate being not digitally palpated.
12	52 yr. ♂	Monorchism (Retentio testis sinistra)	Absence of right testis; retentio left testis; other external genitalia being of normal male; 17-KS: 3.85 mg/day.

Karyotype analyses revealed that they possessed an apparently normal male complement, consisting of 44 autosomes and an XY complex. A similar feature was reported by Ford *et al.* (1959 b) in a case of chromatin-negative Klinefelter's syndrome.

Figs. 1-5. Somatic chromosome complements based on leucocyte cultures. 1, a patient with Klinefelter's syndrome (no. 2). 2, a patient with hypogonadism (no. 8). 3, a patient with agonadism (no. 5). 4, a patient with atresia vaginae (no. 6). 5, a patient with testicular feminization (no. 7).

Patient (no. 5) aged 3 years was clinically diagnosed as agonadism (Table I). Cells from buccal mucosa revealed no sex-chromatin, and drumstic count was negative in polymorphonuclear neutrophil leucocytes. Chromosome counts made in 35 ideal cells showed that all of them had 46 chromosomes (Table II). Karyotype analyses made in two cells identified 5 small acrocentric chromosomes, one of which is the Y, and 15 medium-sized submedian chromosomes involving an X (Fig. 3). It is then evident that this patient has a normal male complement of chromosomes, consisting of 44 autosomal elements and an XY sex-mechanism, and is genetically male.

Clinical diagnosis of patient (no. 6) was atresia vagina (Table I). Exact chromosome counts made in 24 cells showed that 21 cells had 46 chromosomes, while the remaining three cells possessed 44, 45, and

47 chromosomes, respectively (Table II). Karyotype analyses of chromosomes in cells having 46 chromosomes showed that the cells, having in each 4 small acrocentric chromosomes, proved the absence of the Y, while there occurred 16 medium-sized submedian chromosomes with a sign of the presence of two X elements (Fig. 4). A possible conclusion is that the present case has a normal female chromosome constitution, 44 autosomes and two X's.

Table II. Chromosome constitution in twelve cases of sexual abnormalities

Pat. No.	Chromosome counts					Chrom. constitution	No. of cells obs.
	44	45	46	47	48		
1		1	2	15	1	47/2A+XXY	19
2			1	46	1	47/2A+XXY	48
3			9	1		46/2A+XY	10
4		4	24	1		46/2A+XY	29
5			35			46/2A+XY	35
6	1	1	21	1		46/2A+XX	24
7		5	28	3		46/2A+XY	36
8		1	36	2		46/2A+XY	39
9		1	41	1		46/2A+XY	43
10			7			46/2A+XY	7
11	1	2	11			46/2A+XY	14
12		2	19			46/2A+XY	21

A phenotypically female patient (no. 7) was clinically diagnosed as testicular feminization. In 28 out of 36 cells observed a normal diploid number, 46, was found (Table II). Chromosome analysis showed that 46-chromosome cells had in each a normal male complement because of the occurrence of 15 medium-sized submedian chromosomes including an X element and 5 small acrocentric elements involving the Y. This patient is therefore genetically male.

Two cases diagnosed as hypogonadism (nos. 8 and 9) indicated after careful chromosome counting that they invariably had 46 chromosomes. There were no detectable changes or variation of chromosomes from a normal complement either morphologically or numerically in these cases (Fig. 2): each case comprised 44 autosomes superficially identical to those of a normal complex, and X and Y sex chromosomes.

Examinations of the chromosomes in two cases of hypospadias (nos. 10 and 11) yielded consistent counts of 46 chromosomes. Karyotype analyses made it clear that they had a normal chromosome complement consisting of 44 autosomes and an XY sex-constitution. It is self-evident that they are genetically males.

One case of monorchism (no. 12) was found to have a diploid

complement of 46 chromosomes with an XY sex-mechanism. By means of morphological analysis the karyotype was found to be superficially of a normal male pattern.

Remarks: During the past several years, the chromosomes of a number of human syndrome subjects have been investigated; many instances have been provided of association between chromosome abnormalities and various disease states. Recently, various types of sexual aberrations in man have been found associated with abnormalities of the sex-chromosomes.

The great majority of phenotypic males classified under the general term of Klinefelter's syndrome were characterized by an XXY sex-mechanism. In four cases of Klinefelter's syndrome herein dealt with, two cases were found to have 47 chromosomes and an XXY sex-constitution, in complete agreement with those usually known in the vast majority of cases of this syndrome; the other two cases were found to have 46 chromosomes showing a normal male sex-mechanism. A similar feature has been reported in cases of chromatin-negative Klinefelter's syndrome, or pseudo Klinefelter's syndrome.

To date no studies have been reported of the chromosomes of any patient with agonadism. The clinical and diagnostic features of the patient dealt with in this paper were previously reported by Tsuruta (1961). Cytological examinations made on leucocyte cultures have revealed that this patient possesses a normal male chromosome complement, his XY sex-constitution suggesting evidence of a sex chromatin-negative condition.

A case of atresia vagina under study was found to be a genetically female individual by the fact that she has 46 chromosomes with an XX sex-determining mechanism. A patient whose defect was clinically diagnosed as testicular feminization was recognized cytologically as male on account of his 46 chromosomes including an XY sex-complex. Two cases of hypogonadism studied showed that they were males having 46 chromosomes with an XY sex-mechanism. Two cases of sexually abnormal individuals with hypospadias and one case of monorchism, all being phenotypically males, were also recognized genetically as males.

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