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Chromosomal Role in the Aetiology of Amenorrhoea, Sterility and Reproductive Failure

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Summary

Chromosomes play an important role in the aetiology of amenorrhoea, sterility and reproductive failure. We report the prevalence data obtained from screening 3010 cases over a period of 20 years. The frequency of chromosomal defects was detected in 8.34% of the cases investigated. The type of the defects identified and the possibilities of their origin are described.

Introduction

Cytogenetic investigations play a vital role in the diagnosis of genetic disorders, for proper management and genetic counselling. Hansteen et al (1982) identified chromosomal anomalies in 2% of consecutive new borns. However, in cases suspected for chromosomal abnormalities the percentage ranged from 16-24% (Kitano et al 1986, Mitra et al 1988). Chromosomal analysis has become a useful tool in speculating the aetiology of cases presenting amenorrhoea, sterility and reproductive failure.

Delayed initiation of menses at the time of puberty or even the interruption of an established menstrual pattern essentially represents amenorrhoea. Sterility in its broadest context includes both sub-fertility and absolute sterility. Individuals with reproductive failure include repeated abortions, still births, neonatal deaths and congenital anomalies.

The present study was carried out to find the possible role and distribution of chromosomal anomalies in predisposing these conditions.

Materials and methods

The study is comprised of 3010 cases presenting with a history of amenorrhoea (n=1144), sterility (n=228) and reproductive failure (n=1638), over a period of 20 years (Jan 1979 – Jan 1999). The cases were referred to cytogenetic clinic at the Institute of Genetics, O.U., Hyderabad for confirmation and counseling from various hospitals of Andhra Pradesh, South India. A complete clinical assessment and information pertaining to age, region, religion, habits, health status, pedigree, medical histories etc. were recorded in special case proformas. The age of the subjects ranged from 13 to 46 years.

Chromosomal investigations were carried out

using lymphocyte microcultures according to a modified method of Moorhead et al (1960). G-banding was performed according to Seabright (1971). Wherever necessary C-banding (Arrighi and Hsu, 1971) and Ag NOR banding (Howell and Black, 1978) techniques were also employed. Buccal mucosal cells stained with 2% aqueous toluidine blue were scored for X and Y chromatin (Borgaonkar and Hollander, 1971). 20 to 25 well spread metaphases were scored in each case and in case of mosaicism 50 or more metaphases were analysed.

Results and Discussion

The analysis of the karyotypes are presented in Table-I. 21.34% of the cases presenting with primary amenorrhoea were found to have chromosomal abnormalities. The most common chromosome anomaly associated with primary amenorrhoea is 45X0. The typical phenotypic changes observed were short stature, webbing of neck, shield chest, cubitus valgi and absence of secondary sex characters. On ultrasonographic examination majority of cases revealed absence of uterus and a few showed hypoplastic uterus. The reason for the origin of 45X0 could be either anaphase lag or mitotic non-disjunction in the early embryonic stage.

45X 46XX was the most frequent X-chromosome mosaicism. Secondary sex characters were normal or poorly developed in the cases studied. The possible reason for this chromosome pattern may be the consequence of elimination of one of the X-chromosomes during the subsequent divisions of a zygote starting with a normal female chromosome complement. Other types of sex chromosome mosaicism observed by us include 2 cases with 45X/47XXX, 2 cases of 46XX/47XXX and 2 cases of 45X/46XX/47XXX.

We observed rare variants of 46X, fragment (2 cases), 45X 46X, fragment (1 case), 46 XX/46X, fragment (2 cases). The clinical findings, late replication studies of the X-chromosome and the absence of Y fluorescent body in the buccal epithelial cells, clearly showed the fragments to be a portion of the X-chromosome.

47 XXX karyotype was seen in 0.21% of the women ascertained. One rare case with 48 XXXX was found to have mental retardation and primary amenorrhoea.

Patients with mixed gonadal dysgenesis are often mosaics with a 45X/46XY karyotype. Turner's stigma may be present and masculinization is common in this group with a streak gonad on one side and a testis on the other. Ten such cases were observed in the present study. The typical characteristic features observed included ambiguous external genitalia, clitoromegaly, hypoplastic uterus, absence of secondary sex characters. Knudtzon and Aarshog (1987) summarized the wide clinical spectrum of these mosaics that range from female with turner like phenotypic males and females with mixed gonadal sysgenesis, male pseudohermaphrodites to almost normal.

Six cases with XY and XX mosaic forms of pure gonadal dysgenesis were identified. One of the six cases of mosaic type 46XX/46XY dyagenesis showed a hypoplastic uterus, palpable testis, not well formed scrotal sacs and urethral opening below scrotal sacs. This may be the product of double fertilization of an already divided ovum with two sperms, one bearing X and the other bearing Y chromosome or due to the fusion of two embryos, one bearing XX and the other XY (De la Chapalle et al, 1974). Another female subject in this category also had bilateral palpable testes, hypospadias and absence of vaginal opening. One case with 46XX 47XXY was also observed.

Testicular feminization with 46XY karyotype was observed in 13 cases. The cause of primary amenorrhoea in these cases is due to a mutant gene in the X-linked Tfm (Testicular feminization locus) that causes individuals with 46XY karyotype to become phenotypic females. The normal allele at the Tfm locus has been assumed to act as a major regulatory gene in secondary sex development. The other abnormalities observed by us include a case of 46XX, 13p+, a case of 46XX/46Xi (Xq) and two cases with 45XX t (21 ; 21).

Cases with secondary amenorrhoea exhibited abnormal karyotype in 6.9% of the cases. A rare case with 45X/46XX/47XXX was detected. The other abnormalities included were one case with 45X0, 7 cases with 45X0/46XX, one case with 45X0/47XXX, one case with 46XX/47XXX, 2 cases with 46XY and one case with 46XX 13 p+.

The association of specific sex chromosome aneuploids with particular syndromes led to the screening of subjects with sterility to determine the frequency of such abnormalities. The abnormalities observed in the female partners were two cases with 45X/46XX, one case with 46XX/45XX t(21 ; 21), one case with 46XY and one case with 47XXX.

Cytogenic studies often forms important parameters for the medical evaluation of subjects presenting a bad obstetric history (BOH). Earlier studies revealed that about 5-10% of cases of BOH were due to chromosomal anomalies (Simpson, 1980). It is well known fact that chromosomal rearrangements

Table – I
Frequency of chromosome abnormalities in Amenorrhoea, sterility and reproductive failure.

Type of Disorder	Total No. of cases Investigated	Chromosome Constitution	No. of cases	Percentage of chromosome abnormalities
Primary amenorrhoea	942	45, X0	84	8.92
		45, X0/46, XX	68	7.22
		45, X0/46, XX/47, XXX	2	0.21
		45, X0/47, XXX	2	0.21
		46, XX/47, XXX	2	0.21
		47, XXX	2	0.21
		48, XXX	1	0.11
		46, X, fragment	2	0.21
		45, X0/46, X, fragment	1	0.11
		46, XX/46, X, fragment	2	0.21
		45, X0/46, XY	10	0.06
		46, XX/46, XY	6	0.63
		46, XX/47, XXY	1	0.11
		46, XY	13	1.38
		46, XX/47, XX +21	1	0.11
		46, XX/46, Xi (Xq)	1	0.11
		46, XX, 13p+	1	0.11
		45, XX t(21:21)	2	0.21
		46, XX	741	78.66
Secondary amenorrhoea	202	45, XO	1	0.49
		45, X0/46, XX	7	3.47
		45, X0/47, XXX	1	0.49
		46, XX/47, XXX	1	0.49
		45, X0/46, XX/47, XXX	1	0.49
		46, XY	2	0.99
		46, XX 13p+	1	0.49
		46, XX	188	93.07
Sterility	228	45, X0/46, XX	2	0.53
		46, XX/45, XX t (21 : 21)	1	0.26
		46, XX/47, XX +21	1	0.26
		46, XY	1	0.26
		47, XXX	1	0.26
		46, XX	222	98.42
Reproductive failure	1638	45, X/46, XX	18	1.1
		47, XXX	2	0.12
		47, XX + mar	1	0.06
		46, XX/47, XX + mar	1	0.06
		45, X0/46, XX/47, XXX	1	0.06
		46, XX t (7:13)	1	0.06
		45, XX t (13:15)	1	0.06
		45, XX t (14:15)	1	0.06
		45, XX t (14:21)	1	0.06
		45, XX t (21:21)	1	0.06
		46, XX 16p+	1	0.06
		46, XX Gp+	1	0.06
		46, XX	1608	98.17

especially balanced translocations constitute a recognized cause for repetitive early spontaneous abortions. Individuals with balanced translocations are phenotypically normal, but as a result of normal meiotic segregation a proportion of their gametes are chromosomally unbalanced and may predispose to abnormal offspring with chromosomal imbalance (Simpson, 1989). The translocations observed in our study include one case with 46XX rcpt(7,13), one case with t(13:15), one case with t(14:15), one case with t(14:21) and one case with t (21:21).

In our study the frequency of translocation was 0.30%. This frequency was higher than the percentage of translocation reported in cytogenetic analysis of consecutive new borns (De Braekeleer and Dao, 1990). Notably, 2 women in whom a robertsonian translocation was detected were found to have a Down's syndrome child in the past.

Supernumerary (marker) chromosomes were observed in 2 women, the complement were 47XX + mar and 46XX/47XX + mar. The accessory chromosomes interface with normal meiosis leading to hypo or hyper haploid gametes and their participation in fertilization can result in the formation of zygotes with monosomy or trisomy.

The X chromosome mosaicism was observed in 19 women. In 18 women we observed 45X/46XX and in one woman it was 45X/46XX/47XXX. 47XXX was observed in 2 women. The women with 47,XXX karyotype are at a higher risk for meiotic nondisjunction leading to hypo or hyper haploid gametes. Other abnormalities included are one case with 46XX 16p+ and one case with 46XX 6p+.

In our study on the role of chromosome in the aetiology of amenorrhoea, sterility and reproductive failure we observed chromosomal abnormalities in 8.34%. The present report of 3010 cases investigated over a period of two decades helps to establish the prevalence of chromosomal abnormalities in women suffering from the above anomalies. In our opinion chromosomal studies form on integral component in the evaluation of these cases, for the prognosis and proper genetic counseling.

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