



Cytogenetic Study in Females with Secondary Infertility

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ABSTRACT

Infertility affects approximately 10%-15% of couples in reproductive age. There is a complex correlation between genetics and infertility. Several factors affect on gametogenesis, from which factors that lead to chromosomal abnormalities are one of the best known. The aim of this study was to determine type and rate of chromosomal abnormalities in females suffering from secondary infertility^[1]. We studied 67 females with history of secondary infertility to find out the proportion of cytogenetic causes in them. We observed chromosomal abnormalities in 2.98% females, of which showed structural abnormalities.

Keywords:- Secondary infertility, Chromosomal abnormality, Female infertility, Karyotyping

INTRODUCTION

Infertility is the most significant human health problem of the reproductive years. Chromosome abnormalities (CA) are the major contributors to the genetic causes of reproductive disorders^[7]. The World Health Organization has described "infertility" as a health problem of global concern, one in seven couples experience infertility^[3]. Shah et al. have also reported that one in every six

couples wishing to start a family fall into this category^[4]. The prevalence of infertility is reported to be 10-15% worldwide. It is estimated that infertility affects nearly 50 to 80 million people around the world^[5]. Currently 8-10 million infertile couples are estimated to be in India. The recent National Family Health Survey estimated childlessness as 2.4% of currently married women over 40 years of age in India^[6]. Reproductive

failures include a wide variety of problems as infertility, pregnancy loss, abnormal pregnancy, and birth defects. The cause of almost any reproductive abnormality can be the result of genetic and physiological events that occur in mother, father and child [8]. Mutant genes and chromosomal disorders can disturb gamete formation and impair normal embryonic development. Besides aneuploidy segregation due to parental chromosomal aberration, a post-zygotic factor can lead to uncontrolled chromosome distribution in early cleavage stages producing mosaicism [9]. Constitutional chromosome abnormalities (CA) can account for infertility or recurrent pregnancy loss. Approximately 15%-20% of clinically recognizable pregnancies end in spontaneous abortion [10] - [11]. The incidence of chromosome abnormalities in those abortions is as high as 70% [12] - [13]. A modest but clinically important proportion of spontaneous abortions are caused by a balanced chromosomal aberration in one of the parents. This results from the production of gametes and embryos with unbalanced chromosome sets. The clinical consequences of such abnormal gametes include sterility, repeated abortions, and giving birth to malformed children [14].

MATERIAL & METHODS

Our study was conducted to evaluate the chromosomal causes in the patients referred for secondary infertility. The patients having a history of secondary infertility attending the genetic clinic were initially screened with a detailed gynaecological examination. History taking included the history of recurrent abortion (at least two), intrauterine fetal deaths or stillbirths, abnormal children. Due consent was taken and then these patients were screened for chromosomal abnormalities.

Method - Karyotyping

1-2 ml of peripheral blood of the patient was collected in sterilized, heparinized 5ml syringe by venipuncture. Planting of the culture was done on the same day. The contents of each culture vial were mixed gently and incubated for 3 days at 37°C. For each patient two vials were planted with PHA-M.

The planted cultures were shaken well after every 24 hours. This enhances better growth. Harvesting of the culture was done using colchicines (Gibco BRL). After harvesting, Giemsa banding was done and metaphases were screened. Well spread metaphases were photographed & arranged as per classification of human chromosomes recommended by International System for Human Cytogenetic Nomenclature (ISCN 1978).

Table no 1- Associated parameters studied

Sr. No	Parameter studied	No of patients	Percentage
1	Consanguinity	10	
2	Disorders of ovulation	06	
3	TORCH positive	15	
4	Anatomical abnormalities	06	
5	Hormonal imbalance	02	
6	Associate medical illness	09	

Table No 2- Proportion of Chromosomal abnormalities in Females with Secondary Infertility

Total patients	67	
Chromosomal abnormality	Structural	02
	Numerical	00

Table No 3 – Obstetric history & Structural chromosomal abnormalities found in the present study

Sr. No.	G,P,A,L	Chromosomal constitution
1.	3,0,3,0	46, XX80%/46,X,r(X)20%
2.	4,1,3,1	46,XX t(13/14)

DISCUSSION

Infertility is a common clinical problem. It affects approximately 10%-15% of couples in reproductive age. [15]. The prevalence varies widely, being less in developed countries and more in developing countries where limited resources for investigation and treatment are available [16]. Age alone impacts on fertility; aging of the reproductive system plays a role and spontaneous abortion provides another factor [21]. The risk of clinically recognized

spontaneous abortion was 10% under age thirty, 18% in the late Thirties, and 34% in the early Forties. In our present study we found that out of the 7 women referred for secondary infertility 67 (47.76%) were in the age group 21-25.

Among the factors, which are known to increase the risk of chromosomal abnormalities in offspring, consanguinity has a significant role [22]. In the present study of 67 females 10 females had a history of consanguinity.

Disorders of ovulation account for about 30% to 40% of all cases of female infertility [23]. Out of the 67 females referred for secondary infertility, 6 females (8.95%) showed menstrual irregularities. Some however feel that there is definite association of toxoplasma infection with pregnancy loss. [24]-[25] found association of toxoplasma with spontaneous abortion [26]- [27] slated that herpes simplex virus and human cytomegalovirus directly infect the placenta and fetus. The resulting villitis and related tissue destruction may lead to pregnancy disruption. In the present study it was found that 15 (22.39%) females out of the 67 referred for secondary infertility had positive TORCH titers. These results were subjected to statistical analysis and were found to be significant. Anatomical abnormalities of both the uterine cervix and the uterine body have been associated with recurrent pregnancy loss. Acquired anatomic anomalies have likewise been linked to both isolated and recurrent pregnancy losses. These abnormalities include such disparate conditions as intrauterine adhesions, uterine fibroids and endometriosis. In our present study anatomical abnormalities were found in 6 out of 67 (8.96%) women referred for secondary infertility. Hormonal studies – Spontaneous pregnancy losses occurring before 10 weeks of gestation may result from a number of alterations in normal progesterone production or use. Hypothyroidism has been associated with spontaneous and recurrent pregnancy loss [23]. In our present study 2 out of 67 (2.99%) with secondary infertility had hormonal problems.

Associated Medical illness – Any severe systemic illness such as tuberculosis, diabetes mellitus, renal failure, liver failure, metastatic cancer etc can lead

to disruption of hypothalamic-pituitary-ovarian axis and cause infertility [23]. In our country tuberculosis is an important cause of pelvic inflammatory diseases [28]. In our study we found that 9 out of 67 (13.43%) females referred for secondary infertility had an associated medical illness.

There is a complex correlation between genetics and infertility. Several factors affect gametogenesis, from which, factors that lead to chromosomal abnormalities are one of the best known. Some chromosomal aberrations are inherited, while others arise de novo [17].

In the present study, we evaluated 67 females with history of secondary infertility. Chromosomal abnormalities were found in 2 patients i.e. 2.98%. There were no numerical abnormalities found in the present study group i.e. 0%. Structural abnormalities were found in 2 patients i.e. 2.98%. The structural abnormalities were 46, XX80%/46,X,r(X)20% & 45, t (13/14). In a study conducted by Duzcan et al (2003) they found mosaicism as a cause of infertility in 3 out of 354 (1.97%) patients. In our study we found 3 out of 200 (1.5%) patients. Palka et al 1990 discussed the significance of mosaicism XO/XX, and suggested this chromosome picture may be associated with fertility [18]. Chromosomal aberrations in recurrent abortions are mostly structural ones and those in female infertility mosaicism of sex chromosomes [19]. Our present study shows a single case of 46, XX t (13/14). In females the children may have a normal or an abnormal chromosome. Balanced chromosomal translocations lead to 50% risk of spontaneous abortions in phenotypically normal adult carriers [20].

CONCLUSION

Chromosomal abnormalities form the important contributors to the causes of infertility. It is therefore important to do thorough cytogenetic investigation of patients with infertility as it forms the basis of genetic evaluation of the patient. Further counselling will depend on the type of abnormality detected.

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