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Cytogenetic Abnormalities Found in Patients with Reproductive Problems

Mirela Mackic-Djurovic[1], Sabaheta Hasic[2], Emina Kiseljakovic[2], Dunja Rukavina[3], Slavka Ibrulj[1]

[1]Center for Genetic, Faculty of Medicine, University of Sarajevo, Sarajevo, Bosnia and Herzegovina

[2]Department of Medical Biochemistry, Faculty of Medicine, University of Sarajevo, Sarajevo, Bosnia and Herzegovina

[3]Veterinary Faculty, University of Sarajevo, Sarajevo, Bosnia and Herzegovina

Corresponding author: Mirela Mackic-Djurovic, PhD. Faculty of Medicine. University of Sarajevo, Čekaluša 90, 71000 Sarajevo, Bosnia and Herzegovina. Phone:+387 33 226 472 lok.159. ORCID iD: http://www.orcid.org: 0000-0001-7198-5642. E-mail:mirelamd@yahoo.com

ABSTRACT

Introduction: One of the important causes of male infertility is aberration at the chromosomes. **Aim:** The main purpose of this study was to determine the frequency and types of chromosomal aberration in infertile/sterile men whose samples were analyzed in the Center for Cytogenetics of Faculty of Medicine University of Sarajevo in the last four years. **Methods:** A total of 353 infertile/sterile men, between the ages of 22-55 years, referred for cytogenetic analysis to the Center for Genetics of Faculty of Medicine during the period 2013-2016. Karyotyping was performed on peripheral blood lymphocytes by using the Giemsa trypsin banding (GTG) technique. **Results:** The structural and numerical chromosomal aberration in infertility/ sterility of men found with the incidence of 6% (20/353). Out of the 20 patients with abnormal cytogenetic diagnosis, structural chromosome abnormalities were observed in 17 (85%) patients and 3 (15%) with numerical aberrations. The type of aberrations mostly found were Robertsonian and reciprocal translocations (35%, 35%, respectively). **Conclusions:** The incidence of chromosomal abnormalities in infertile/sterile males suggests that the cytogenetics analysis is an important in male infertility, especially if it will be used for the purpose of assisted reproduction techniques.

Keywords: Cytogenetic analysis, structural and numerical chromosomal aberration, infertility.

1. INTRODUCTION

The World Health Organization has described infertility as a health problem of global concern. Infertility is defined as inability to conceive in period of 12 months of normal intercourse (without contraception) (1). Infertility affects about 10%-15% of couples around the world (2). Genetic factors have only 10% of influence at infertility at men. There are many and diverse causes of male infertility, including accidental causes, hormonal imbalance, erectile dysfunction, infections, antisperm antibodies, environmental pollutants, or genetic factors. (3, 4). However, infertility causes are unknown in 12%-41% of men (5). Chromosomal anomalies are considered as one of the most important causes of male infertility. The incidence of chromosomal abnormalities is about 1:1000, half of them are numerical, and half of them are structural aberrations, but also, both normalities can occurs together. Infertility caused by numerical and structural aberrations occurs more often in male than in female. The rate of chromosomal abnormality in infertile men is 2.1-19.48% (6).Chromosomal abnormalities are not the only genetic causes of male infertility. Deletion of the Y chromosome region containing the azoospermia factor (AZF) is considered the most common genetic cause of male infertility (7).

The main purpose of this study was to investigate the frequency and types of major cytogenetic abnormalities among infertile/sterile man, analyzed in Center for Genetics, Faculty of Medicine University of Sarajevo in the period 2013-2016.

2. MATERIAL AND METHODS

Sample

The cross-sectional study included cytogenetic analysis of 353 male that has been performed in Center for Genetics of Faculty of Medicine, University of Sarajevo. The mean age of patients was 35 years (22-55). Participants recruited in the study had already been considered as an infertile/sterile or whose spouse had more than one spontaneous abortion. The present study was performed in accordance with the ethical standards and the Declaration of Helsinki.

Methods

Cultivation of peripheral blood sample at patients' is performed by

short-term cultivation methods (8). Cytogenetic analysis is performed under International System of Human Chromosomal Nomenclature (ISCN) regulations (9). With every karyotype analysis 25 cells per sample is used by G-bending technique (10). C-bend is used to confirm heterochromatic regions.

Statistical analysis was done using -SPSS version 19.0. (Chicago, IL, USA) program. Data were expressed in term of frequency such as total number of cases and percentage of total number. The average age of the subjects age was presented as median with the range of minimum and maximum of the variable.

3. RESULTS

Cytogenetic analysis showed that among the 353 patients included in the study, 333 men (94%) had a normal karyotype (46; XY), and 20 men (6%) had various chromosomal abnormalities. Analysis of chromosomal abnormalities in 20 patients (the mean age was 34 (27-43) years) revealed (85%) abnormality of autosomal chromosomes and (15%) of sex chromosomes. They were dominantly infertile and only 1% sterile. Average life expectancy at men with chromosomal aberrations is 34 years and without chromosomal aberrations 35 years.

The most frequent structural aberrations were Robertsonian translocation (Figure1) and reciprocal translocation (7, 7 cases, respectively) followed by inversion of chromosome 9 detected in three cases. Numerical aberrations were observed in 3 patients: one patient with Klinefelter syndrome, one patient with 47, XY, + mar chromosome and one with mosaic forms 46,XY/45,X. Characteristics of the patient's detected chromosomal abnormality were summarized in Table 1.

Type of aberrations	Karyotype	Frequency(n)	Percentage(%)
Structural aberrations	47,XY+marker	1	5%
	46,XY/45,X	1	5%
	47,XXY	1	5%
Numerical aberrations	46,XY, inv9(p11;q13)	3	15%
	45,XY;rob(13;14)(q10;q10)	6	30%
	46,XY;t(6;7)(q25;q36)	1	5%
	46,XY, der5,t(5;19)(q35;p13.22)	1	5%
	46,XY; t(11;17)(q13.3;q25)	1	5%
	46,XY;t(5;9)(p15;q22)	1	5%
	46,XY;t(14pq?;11q)	1	5%
	46,XY, der 12, t(12q;18q)	1	5%
	45,XY,rob21,t(21;21)(q10;q10)	1	5%
	46,XY; der20, t(13q14-34;20q13)	1	5%

Table 1. Type and frequency of chromosomal aberration in infertile/sterile man. Legend: Data are presented as absolute number and percentage of total number of cases affected by a particular chromosome aberration. n-number of cases

4. DISCUSSION

The present study results implicate the contribution of structural and numerical chromosomal aberration in fertility/sterility of men. According to updated and published studies, the usual cause of infertility is aneuploidy

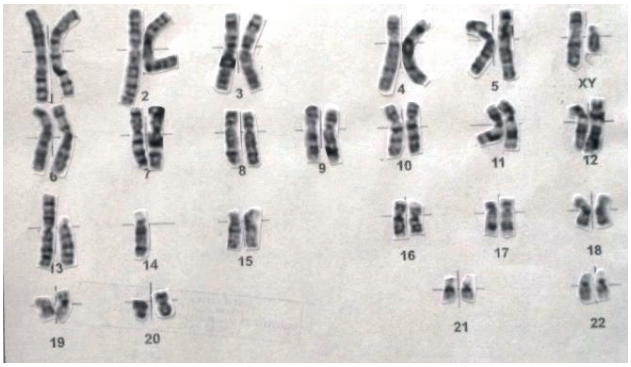

Figure 1: Karyotype of a patient with Robertsonian translocation: 45,XY,t(13;14) (q10;q10)

of sex chromosomes (11), while in our case there were more changes on autosomes (85% vs. 15%).

The most common autosomal abnormality detected in infertile men was the Robertsonian translocation karyotype. Among them the most frequent was 45,XY,t (13;14) karyotype and only one with 45,XY,t (21;21) karyotype were detected in our study.

The most principal role in male infertility is performed by numerical and structural chromosome abnormalities.

Normal chromosome pairing and segregation at meiosis I (formation of unbalanced gametes and subsequent unbalanced abnormal offspring) is influenced by autosomal abnormalities (12). Low sperm concentrations and abnormal sperms with male infertility and increased miscarriages can be caused by structural chromosome autosomal abnormalities (13).

Both the chromosomes involved in translocation and the location of the breakpoints are likely to be determining factors for the fertility status of the patient. In addition, Robertsonian translocation can result in offspring with Down syndrome or Patau's syndrome or in gestational loss of concepts with monosomy of chromosome 13, 14 or 21, or trisomy of chromosome 14, which are lethal (14). Male infertility and chromosomal anomalies are often closely related and reciprocal translocations are the most frequent (1 in 600) structural chromosomal anomalies in humans (12). Carriers of these translocations normally do not have any abnormal phenotype which leads to having normal offspring, recurrent miscarriage also chromosomally abnormal offspring and in some cases infertility (15).

Pericentric inversion found in the participants is one type of structural changes, chromosomal rearrangement that is considered to be just benign chromosomal polymorphism and is not expected to correlate with abnormal phenotype (16). Pericentric inversion of chromosome 9 can cause clinical problems on offspring of the inversion carrier, as well as at infertility of unknown etiology sex linked and it be a cause of the disturbances of spermatogenesis (17, 18).

In the present study, we found balanced translocation to be the most frequent structural aberration among these patients. A balanced translocations rarely cause an abortion, their determination is very important. Also, it is cause of offspring conception with unbalanced karyo-

type. Offspring with some kinds of unbalanced rearrangements can survive gestation age, but have multiple congenital malformations and mental retardation (19).

The clinical situation where the development of the chromosomal, gonadal or anatomical sex is atypical is generally considered to be disorder of sex development(DSD) (20).

In our sample two forms of the DSD sex caused by chromosome aneuploidy were observed: 47; XYY and 45; X/46; XY. The mosaic form are described in many studies and the phenotype can range from female phenotype and ambiguous to male phenotype with infertility (21). The formation of mosaic 45,X/46,XY leads to spermatogenic defects. These males carry an increased risk of developing gonadal tumors and must be followed closely (22). In our study, we found one case with Klinefelter syndrome (47,XXY karyotype). According to published studies, this syndrome is the most common abnormality of sexual differentiation, and occurs in approximately 1 in 500 live births. (23, 24). It is a form of primary testicular failure with testicular hypotrophy and elevated gonadotropin plasma levels, and represents the most common form of male hypogonadism (25). Klinefelter syndrome is often not diagnosed until adulthood. Most men with the syndrome produces little or no sperm. But assisted reproductive technology may allow some men with this syndrome to become parent. Low number of numerical abnormalities is not in a line with published data concerning the worldwide prevalence of Klinefelter syndrome which implicates necessity of chromosomal abnormalities screening among population with history of family fertility problems. Possible reasons of low frequency of Klinefelter syndrome detection in Center of Genetics may be due to inadequate education and unconsciousness the chromosomal aberration as cause of infertility/sterility so many of them are undiagnosed.

Our results (5,67%) of chromosomal abnormalities in infertile males indicate that the chromosomal abnormalities are one of the important causes of male infertility and a risk for transmission of chromosomal abnormalities to their offspring. Therefore, it is necessary to do karyotype and genetic counseling. And as one of the solutions for healthy offspring is preimplantation genetic diagnosis (PGD).

5. CONCLUSION

This cytogenetic study on infertile or sterile men confirmed previous reports that chromosomal abnormalities might be one of the most common causes of infertility and a risk for transmission of these genetic defects to the future generations. Therefore routine cytogenetic analysis are necessary prior to reproductive techniques.

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- Author Contribution: All authors participated in each step of research. All authors gave final approval for final version to be submitted.

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