Chromosome Aberration in Azoospermias

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Abstrak

Enam (11,3%) dari 53 penderita azoospermia sekresi ternyata menunjukkan aberasi kromosom. Aberasi ditemukan pada pemeriksaan sitogenetik (analisis kromosom dan pemeriksaan kromatin seks). Sebagian besar (4 dari 6) penderita dengan aberasi kromosom, ternyata telah dapat dideteksi dengan pemeriksaan kromatin seks. Pemeriksaan tersebut relatif murah, sederhana tetapi cukup andal untuk menyingkirkan kasus azoospermia karena aberasi kromosom, yang bila diobati memerlukan waktu lama dan biaya mahal tetapi biasanya tanpa memberikan hasil.

Abstract

Six (11.3%) out of 53 secretory azoospermic patients had chromosome aberrations. The aberrations were found from cytogenetic examinations (chromosome analysis and sex chromatin examination). Most (4 out of 6) of the aberrations could already be detected by sex chromatin examination. The simple, inexpensive and reliable examination has the capability to eliminate the uncurable, chromosome aberration azoospermia, from a time consuming and expensive but usually with no-result treatment.

***Keywords** : Azoopsermia, Chromosome aberration.*

INTRODUCTION

Azoospermia is a basic failure in reproduction, in which there is no sperm in the ejaculated semen of the patients. This abnormality can be divided into secretory and obstructive azoospermia[1]. In obstructive azoospermia, sperm production in the seminiferous tubules is still in progress. Due to the obstruction, the sperms can not be expelled from the testes. The testes usualy has a normal consistency on palpation. In secretory azoospermia, the germinal epithelium of the seminiferous tubules fails to perform spermatogenesis. In contrast to obstructive azoospermia, the testes in secretory azoospermia has a rather soft consistency. Both conditions can be diagnosed by the fructose test. In obstructive azoospermia the fructose test is negative, while secretory azoospermia has a normal, positive fructose test.

Chromosome aberration can be found in and considered as the cause of azoospermia.[2] Klinefelter syn-drome (47,XXY) and their variations are good examples and are mostly the cause of azoospermia (Table 1). Many investigators proposed chromosome examination as a routine procedure in the management of azoospermia.[3]

Table 1. Chromosome aberrations found in Klinefelter syndrome

Chromosome aberration	Type of syndrome	Percentage*
47,XXY	Classical or primary	80
47,XXY/46,XY	Mosaic	
47,XXY/46,XX	„	
47,XXY/46,XY/45,XO	„	
47,XXY/46,XX/45,XO	„	
47,XXY/46,XY/46,XX	„	
		20
48,XXYY	Variants of syndrome	
48,XXXY	„	
49,XXXXY	„	

* from de Grouchy & Turleau, 1984.

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The aberrations found were very varied, either in the types of chromosomes involved or in the percentage of aberrations. If the aberration includes Klinefelter syndrome (47,XXY) the percentage of this aberration in azoospermia varied from 34%[3] to 14.1%.[4] If subjects with this syndrome were excluded, the figures decreased to 7.4%[5] and even to 5.3%.[6]

Structural aberration of one of the sex chromosomes in 46,X (pseudodiploid) were reported repeatedly.[7,8,9,10] Concomitant aberrations of the Y chromo- some and one autosome were also reported, such as in Y and chromosome no.14,[11] in Y and no.15.[12] A familial case with aberration of the X and chromo-some no.3 in 3 generations which gave result to azoospermia, but without any reproductive disturbances in carier women.[13] A mosaic case of 45,XO/46,XY was reported by Werner et al.[14] Azoospermia with structural aberration of an autosome but with normal sex chromosome was also reported, such as inversion of the chromosome no. 1.[15,16] Balanced rearrangement involving 4 chromosomes, no. 1, 5, 10 and 10 and 12 was reported.[17] Balanced translocation involving chromosome 10 and 14 was reported,[8] and involving chromosome 1 and 15[18] and ring chromosome 21 were also reported.[19,20] There was also a case report of azoospermia with normal 46,XY but with a decreased frequency of spermatogenetic meiotic chiasma.[21]

The first objective of the study was to obtain the percentage and the types of chromosome aberrations in azoospermias. The other objective was to find any possibility to develop a more efficient procedure for the management of azoospermia, especially for country like Indonesia. This study will also show the role of cytogenetics in andrology.

MATERIALS AND METHODS

The subjects were husbands of infertile couples, who underwent semen analysis at the Department of Medical Biology, Faculty of Medicine, University of Indonesia and were diagnosed as having azoospermia. They were then suggested to undergo cytogenetic examination, and on agreement a second semen analysis was performed. Only azoospermia with no signs of classical Klinefelter syndrome and with a positive fructose test were accepted as subjects. Fifty three patients with azoospermia, conform to these criteria were studied.

Cytogenetic examinations were sex-chromatin examination and chromosome analysis. Sex-chromatin examination used the modified routine techniques of "Barr body" and "drumstick" for X-chromatin, and "F-body" for Y-chromatin. The specimens were peripheral blood smears and buccal mucosa smears. The chromosome analyses were done with the modified routine microculture technique. Chromosome identification were done morphologically according to the Denver standard. All these methods are used as routine cytogenetic examinations in our laboratory.

RESULTS

Six patients (11.32%) out of 53, had abnormal results of the cytogenetic examination. Two of them had normal female chromosomal pattern (46,XX). The other four had mosaicsm, e.i. 46,XY/45,XO in 2 patients, mosaic 47,XXY/46,XY in one and 46,XX/47,XXY in one patient. Sex chromatin results were in accordance to the chromosomal results. Normal cytogenetic results were found in the remaining, 47 subjects. The results are tabulated in Table 2.

Table 2. Result of cytogenetic examinations of 53 patients with azoospermia

Sex chromatine			
X-chromatine	Y-chromatine	Chromosome	Number
-	+	46,XY	47
+	-	46,XX	2
-	+	46,XY/45,XO	2
+	+	47,XXY/46,XY	1
+	+	46,XX/47,XXY	1

DISCUSSION

This study showed that 11.32% of the azoospermia patients examined in our laboratory, had chromo-some aberration. This figure is rather far below the figures reported by others, who included patients with Klinefelter syndrome in their studies. A percentage of 34[3] was regarded far above the percentage (11.32%) in this study. The difference can be understood very clearly, due to the inclusion of patients with Klinefelter syndrome in their studies. Only Retief et al[4] had a rather similar result (14,1%) with this study.

Compared to reports which also excluded patients with Klinefelter syndrome as in this study, percentages of 7.4%[5] and 5.3%[6] were found. The percentage found in this study was rather high. The difference might be explained as follows : not all infertile

couples in developed countries seek help. Due to their modern culture, children are not very important for their lives. In Indonesia a childless couple is regarded as an incomplete family. If there were no differences in regardin g a child in the family between the societies, the different percentage of chromosome aberration can be due to racial factors. From a theoretical standpoint a difference in the genetic pool can be understood. But anyhow, the difference still can be regarded as a normal variation in the human race.

Chromosome aberrations found in this study were only of the sex chromosome. From a certain point of view, this result is in accordance to the general assumption that the sex chromosome has more influence on normal sexual function than autosomes. Rather astonishing was the detection of 46,XX in 2 patients among the 53 subjects with azoospermia. The first 46,XX male was found by La Chapelle et al in 1964 (cited by de Grouchy and Turleau[22]). They explained that mosaicsm with 46,XY (46,XX/46,XY) actually was present, but the XY component was not detected or has disappeared after the differentiation and development of the testis.

Another explanation proposed was that a part of the Y chromosom which carries the gene for male determination, was translocated to the X-chromosome. RFLP analysis has succeeded in revealing the translocated specific parts of the Y-chromosome to another chromosome.[22] Explanation based on gene mutation was also proposed (La Chapelle 1976 cited by de Grouchy and Turleau[22]). The mutation transforms the 46,XX female to 46,XX male, just as a mutation transforms the 46,XY male to 46,XY female (testicular feminization syndrome).

From the clinical point of view, 46,XX male is rather similar to Klinefelter syndrome, especially in the testis and hormonal condition and sometimes in gynecomastia.[22] The height and body configuration of a 46,XX male is not as high as and not as gynecoid as Klinefelter syndrome. Mental retardation which usually is found in the Klinefelter syndrome, does not exist in a 46,XX male (does not increase compared to normal population). Usually a 46,XX male expresses the H-Y antigen, which explains the paradox of its cytogenetic finding and the genotype.[23]

In this study 2 patients were found to have Klinefelter syndrome mosaicsm. One patient had 47,XXY/46,XY and the other 46,XX/47,XXY. A normal 46,XY zygote which experienced non-disjunction mitosis (cleavage) followed by dying of unviable cell (45,YO) will then end to 47,XXY/46,XY (Figure 1). Those mosaicsm could also originate from a 47,XXY zygote. Loss of an X or a Y chromosome then, during the cleavage, yield to 47,XXY/46,XY or 47,XXY/46,XX mosaic respectively (Figure 2).

Not all patients with Klinefelter syndrome mosaicsm exhibit azoospermia. This can be explained by the possibility that a cell with normal chromosome (46,XY) complement exist in the seminiferous tubules (spermatogenesis epithelium). The result of the spermatogenesis might be a wide range from azoospermia to normospermia. If there is no cell with 46,XY, of course it will result in azoospermia.

Two subjects in this study were found to have a mosaic chromosome complement, e.i. 46,XY/45,XO. From the cytogenetic aspect, this kind of mosaicsm could be derived from a normal zygote, 46,XY. Disturbances in mitosis cause loss of a Y chromosome in a daughter cell, leaving a 45,XO cell (Figure 3). Those cells persisted through the development process, together with the normal 46,XY daughter cell, which then end up as a mosaic newborn. The theoretical fertility status of this mosaicsm is the same as the Klinefelter syndrome mosaicsm. This means that azoospermia is not the rule. A mosaic of 46,XY/45,XO could also have a normal spermatogenesis, if all the germinal cells of the seminiferous tubules were normal cells of 46,XY. On the other hand it could also happen that all the cells of the spermatogenesis were 45,XO which then result in azoospermia, as shown by both cases in this study.

No structural aberration was found in this study. This study used the Denver classification of individual chromosomes by the morphological identification method. Only major structural chromosome aberrations can be identified with this method. Minor aberrations can not be identified with this method. If there were structural aberrations found in this study, the percentage of chromosome aberration would increase (become more than 11.32%).

As shown in Table 2, four out of 6 patients with azoospermia could be regarded as having abnormal cytogenetic results, mainly from the sex chromatin examination. They exhibited different results from the normal 46,XY. Nearly 70% (66.66%) of patients with an aberration of the sex chromosome, can be detected mainly by sex chromatin examination. Only patients with mosaicsm of 46,XY/45,XO have the same results as the normal 46,XY. This means, that they could be erronously regarded as having a normal 46,XY chromosome, without chromosome analysis. Considering that sex chromatin examination need only hours and is relatively inexpensive, this examination could be regarded as an efficient procedure to eliminate other and further examination in the management of azoospermia. As is already known,

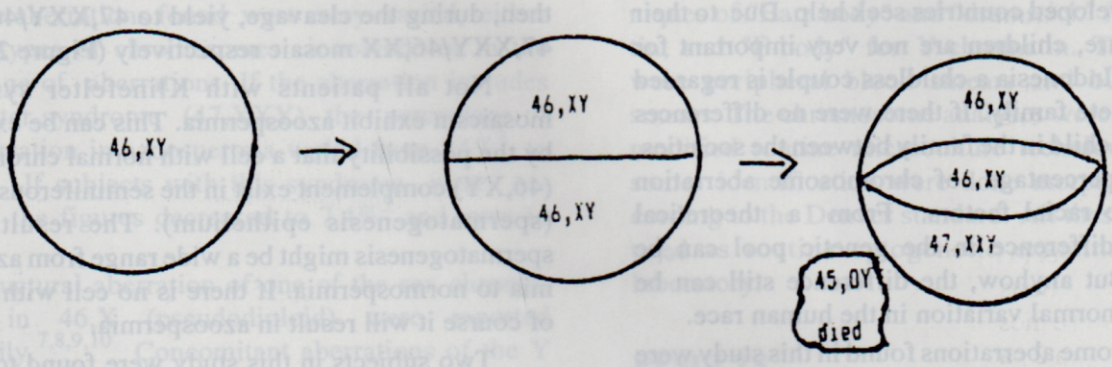

Figure 1. A 46,XY zygote with non-disjunction cleavage yield to 47,XXY/46,XY mosaic.

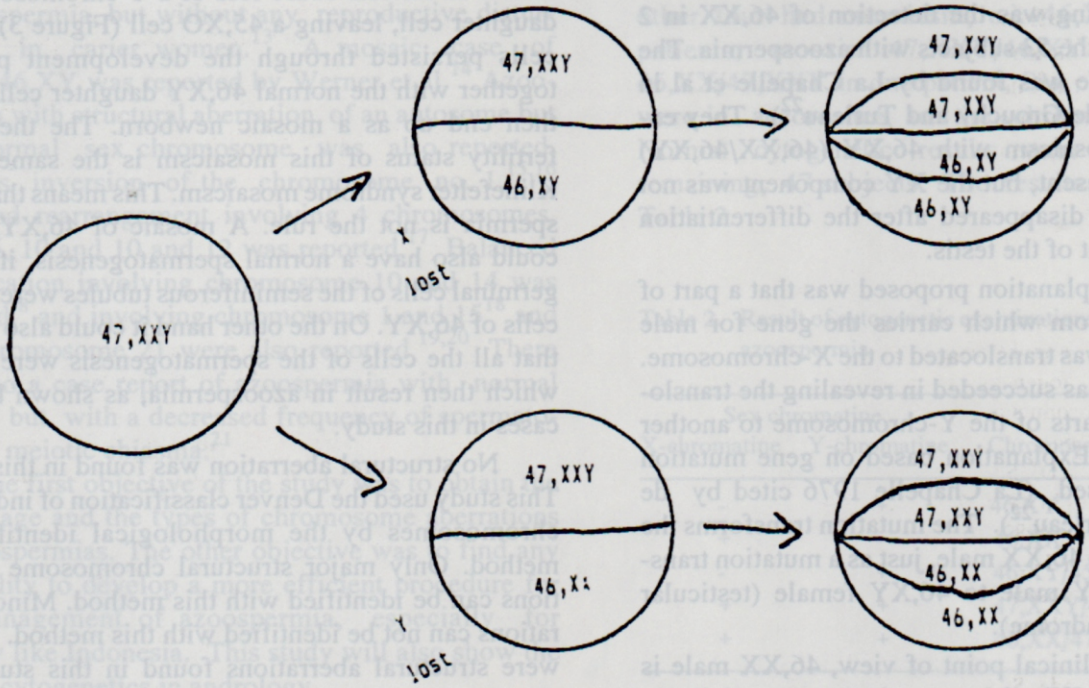

Figure 2. A 47,XXY zygote with lost of an X or a Y chromosome in cleavage then gave result to 46,XY/47,XXY mosaic (upper part) or 46,XX/47,XXY mosaic (lower part).

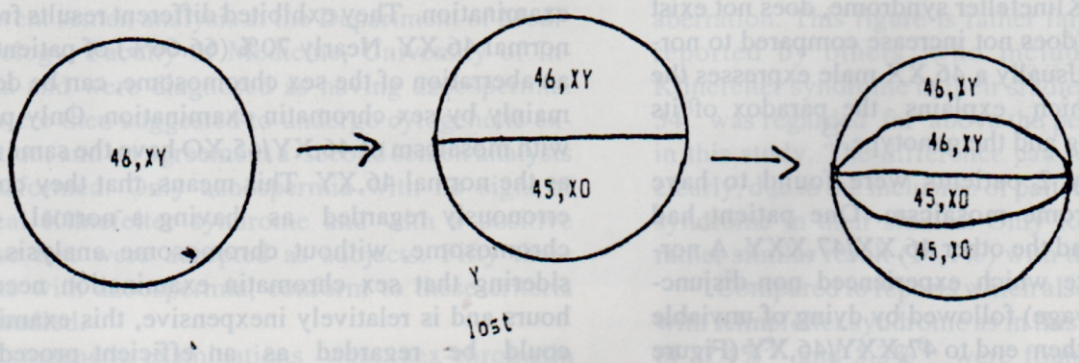

Figure 3. A 46,XY zygote with disturbance in cleavage which give result to 46,XY/45,XO mosaic.

azoospermia with chromosome aberration can not be corrected. Psychological counselling should be born in mind, in dealing with azoospermia patients with chromosome aberrations.

CONCLUSION

Six out of 53 patients (11.3%) with azoospermia have chromosome aberrations. Four of those azoospermics with aberration were mosaic with normal complements i.e. 46,XY/45,XO in 2 patients, 47,XXY/46,XY in one and 46,XX/47,XXY in the second patient, and 46,XX in the other two patients.

Four (2 with 46,XX and 2 with mosaic Klinefelter syndrome) could already be detected as having chromosome aberrations from the chromatin examination only. It is known, that azoospermia due to chromosome aberrations are uncorrectable. The sex chromatin examination could be used as an inexpensive and reliable method to eliminate the uncorrectable azoospermics.

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