Further Evidence of the Clinical, Hormonal and Genetic Heterogeneity of Klinefelter Syndrome: A study of 216 Infertile Egyptian Patients

Short Running Title: Heterogeneity of Klinefelter Syndrome

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ABSTRACT

Objective: This study aims to provide further insight into the phenotypic heterogeneity of Klinefelter syndrome (KS) by presenting clinical, hormonal, and genetic data from a large series of Egyptian infertile patients with KS. Patients and Methods: A retrospective case series of KS patients was studied over the period from January 2003 to April 2010. All patients underwent a complete history and physical examination; color duplex examination; semen analysis; measurement of total testosterone (T), follicle-stimulating hormone (FSH), luteinizing hormone (LH), estradiol (E2), and prolactin (PRL); and chromosomal typing. Mosaic KS diagnosis was confirmed by fluorescence in situ hybridization (FISH). Results: The series included 216 KS patients (198 non-mosaic, 16 mosaic, and 2 KS variants). Typical clinical signs of hypoandrogenism were observed in 86% of patients. Gynecomastia affected 20.8% of the patients. Eunuchoidal body proportions, with arm span exceeding height and lower segment length exceeding upper segment length, were detected in 43.9% and 64.4% of the patients, respectively. In all patients, a reduction in testicular size and azoospermia were detected. Normal levels of T, FSH, LH, E2, and PRL were detected in 44.5%, 3.7%, 3.3%, 93.5%, and 91.2% of
patients, respectively. There were no significant differences between patients with classic KS and those with mosaic KS in terms of the frequency of clinical signs of hypoandrogenism, gynecomastia, low T concentrations, or high concentrations of FSH, LH, E2 and PRL (all p > 0.05). Conclusions: The results of the current study emphasize the heterogeneous clinical, hormonal, and genetic phenotype of infertile KS patients. Our findings support the usefulness of cytogenetic studies in infertile patients showing small testicular size andazoospermia, regardless of the presence of other clinical or endocrine findings.

Key words: Klinefelter Syndrome, Clinical heterogeneity, Hormones, Genetics

INTRODUCTION
Klinefelter syndrome (KS) is the most common numerical chromosomal aberration among men, with an estimated frequency of 1:500 to 1:1000 of live births (Lanfranco et al., 2004). It is the most frequent genetic cause of infertility, occurring in 11% of azoospermic men (Foresta et al., 1999). The syndrome was first described by Klinefelter et al. in 1942. They described a cohort of 9 men characterized by gynecomastia, aspermatogenesis without a-Leydigism, and increased excretion of follicle-stimulating hormone (FSH). After the initial report, it took another 15 years before Jacobs and Strong confirmed the association between KS and an extra X chromosome, establishing it as a genetic disease (Jacobs and Strong, 1959). Although the 47,XXY karyotype is the most common chromosomal abnormality in individuals with KS, mosaic patterns (46,XY/47,XXY) and KS variants with supernumerary X and/or Y chromosomes do exist (Visootsak et al., 2001). The 47,XXY karyotype of KS arises spontaneously due to a nondisjunction event—when the paired X chromosomes fail to separate—in meiosis I or II during oogenesis or spermatogenesis (Thomas and Hassold, 2003). Less than 3% of X chromosome polysomy occurs during early divisions of the fertilized egg. Postfertilization nondisjunction is responsible for mosaicism, which is seen in around 10% of patients (Paduch et al., 2009). The classic description of men with KS emphasizes tall eunuchoid body proportions, low testosterone (T), sparse facial and pubic hair, gynecomastia, small and firm testicles, micropenis, infertility, and mild-to-moderate cognitive deficits. However, it is now well known that this original description is not accurate and that men with KS represent a broad spectrum of phenotypes as well as professions, income, and socioeconomic status (Lanfranco et al., 2004;
Smyth and Bremner, 1998). Because of variation in the severity of symptoms, a large number of
patients are never diagnosed (Ottesen et al., 2007). Only 10% of cases are detected before or
during puberty, and about two-thirds of all men with X chromosome polyploidies fail to be
identified during their lifetime (Bojesen et al., 2003; Abramsky and Chapple, 1997).
A number of reports have examined clinical and genetic factors among Egyptian patients with
KS (Girgis et al., 1969; Temtamy et al., 1980; Ismail et al., 1993). In general, these studies were
not comprehensive (i.e., they did not cover the clinical, endocrine, and genetic heterogeneity of
KS), had a relatively small sample size, or were lacking genetic or cytogenetic investigations
(Girgis et al., 1969) and hormonal investigations (Temptamy et al., 1980). To gain further insight
into the genetic heterogeneity of KS and its phenotypic manifestations, we present clinical,
hormonal, and genetic data from a large series of Egyptian infertile patients with KS.

Patients and Methods
A series of KS patients, who presented with infertility at our andrology clinic, were
retrospectively studied over the period from January 2003 to April 2010. The socio-demographic
and clinical characteristics of the patients are shown in Table 1. All patients enrolled in the study
underwent the usual diagnostic protocol applied to newly referred subjects at the andrology
outpatient clinic. All patients provided informed consent to participate in the study. This study
was approved by the local review board.
All patients completed a standardized history and clinical examination form, with emphasis
placed on the measurement of height, arm-span, and the upper (crown to pubis) and lower body
segments. The patients were examined for gynecomastia, which was defined as a firm, mobile,
discoid mound of tissue evident beneath the nipple that could be palpated between the thumb and
forefinger. Examination for secondary sex characteristics was performed with a focus on facial
and body hair patterns, distribution of abdominal fat, scrotal and penile development, muscle
mass, and strength.
All patients underwent a color duplex examination to evaluate testicular volume and scrotal
contents. Semen samples from the patients were collected by masturbation after 3–5 days of
sexual abstinence and examined as soon as they were liquefied at 37 °C. Ejaculate volume, pH,
concentration, and motility were evaluated according to WHO guidelines (WHO, 1999). Semen
analysis was performed 3 times over a 3 month period for each patient. At least 3 abnormal
semen analyses were required for a diagnosis of oligozoospermia. Azoospermia was verified in at least 3 ejaculates from each patient by pellet analysis after semen centrifugation (1000 × g for 20 min). Sperm counts were performed using a Neubauer counting chamber. Semen parameters presented represent the mean of 3 samples. Blood samples were taken in the morning after an overnight fast for determination of total T, prolactin (PRL), luteinizing hormone (LH), FSH, and estradiol (E2), using a chemiluminescent immunoassay system (Immulite 1000; Siemens Medical Solutions and Diagnostics, Los Angeles, CA, USA). Peripheral blood cultures confirmed abnormal male sex chromosome complement in all metaphases examined. A chromosomal analysis was performed in all cases. All samples were subjected to phytohemagglutinin (PHA)-stimulated whole blood culture in RPMI-1640 media according to standard procedures (Verma and Babu, 1995). Twenty G-banded metaphases were analyzed from each patient, and mosaic study and/or Q-banding was carried out when indicated. Karyotyping was carried out using an imaging system. Mosaic KS diagnosis was confirmed by fluorescence in situ hybridization (FISH). FISH was performed using fluorescence-labeled X- and Y-centromeric probes, according to the method of Rooney and Czepulkowski, (1997). Analysis was carried out using a fluorescent microscope (Nikon E600) equipped with the appropriate filters. Images were captured using Cytovision image analysis software (Applied Imaging 2380 Software Version 3.5, Santa Clara, CA, USA).

**Statistical Analysis**

All statistical analyses were performed using Statistical Package for the Social Sciences (SPSS, Chicago, USA) for Windows 17.0. All variables were checked for normal distribution using the Kolmogorov-Smirnov one-sample goodness-of-fit test. Data were expressed as mean ± SD when normally distributed and as median [range] when parameters had a non-normal distribution. Unpaired two-sided Student’s t-tests were used for comparison of the means of normally distributed parameters. In all other cases, the Mann–Whitney U-test was used for comparisons between groups. The relationship between individual variables was determined by non-parametric Spearman rank correlation tests. Proportions were analyzed by chi-squared test, chi-squared test with Yates continuity correction, and Fisher exact test when appropriate. Differences were considered statistically significant when the p value was less than 0.05.
Results
The series investigated in the current study included 216 Egyptians patients who were suffering from primary infertility. All patients displayed features of non-obstructive azoospermia. There was no family history of male infertility for all patients studied. None of the patients in the series presented with maldescended testes. Table 1 shows the clinical and laboratory characteristics of the patients, with comparison between non-mosaic and mosaic forms of KS.

Karyotyping
Cytogenetic investigations diagnosed non-mosaic, mosaic, and KS variants in 198 (91.6%), 16 (7.4%), and 2 (0.09%) patients, respectively. Table 2 shows the results of karyotyping in patients with mosaic KS and KS variants. Figure 1 shows the 3 stages of interphase detected by FISH analysis, confirming the mosaic form XXY/YYX/XY.

Clinical and hormonal phenotypes
Eighty-six percent of the patients presented with the typical clinical signs of hypoandrogenism (absent temporal recession of hair, decreased facial hair, decreased pubic and body hair, accumulation of abdominal fat, decreased scrotal and penile development, and decreased muscle mass and strength) [Figure 2]. The remaining 14% of patients were phenotypically normal, and all appeared to be normally androgenized [Figure 2]. The testicular size was reduced in all patients. Table 3 shows the frequency of different clinical and laboratory abnormalities in the KS patients. There were no differences between patients with mosaic and classic KS in terms of the frequency of clinical signs of hypogonadism, gynecomastia, arm span exceeding height by 5.3 cm, lower segment length exceeding upper segment length by 5.3 cm, low total T, high FSH, high LH, high PRL, or high E2 (all p > 0.05).

Relationship between clinical and laboratory phenotypes
There were significant negative correlations between age and arm span, height, upper segment length, ejaculate volume, LH, and FSH levels (all p < 0.05). However, a significant positive correlation was found between age and T levels (p < 0.05). There were significant positive correlations between testicular size and the duration of infertility, lower segment length, T, PRL, and E2 levels (all p < 0.05). Additionally, significant negative correlations were found between testicular size and LH and FSH levels (both p < 0.05). Significant positive correlations were observed between ejaculate volume and testicular size, LH, T, and E2 levels (all p < 0.05). In
contrast, a significant negative correlation was found between ejaculate volume and FSH levels (p < 0.05). Arm span was negatively correlated with PRL levels and positively correlated with LH and E2 levels (all p < 0.05). Significant positive correlations were established between height and LH and PRL levels (both p < 0.05), and a significant negative correlation was found between height and E2 levels (p < 0.05). There were no significant differences between KS patients who presented with the typical signs of hypogonadism, eunuchoidal body proportions, or gynecomastia and those without these signs in terms of FSH, LH, PRL, and E2 concentrations (all p > 0.05, data not shown). However, there was a significant decrease in T concentrations (z = -5.95, p < 0.001) among patients with gynecomastia compared to those without. There were no significant differences in ejaculate volume among patients with gynecomastia when compared to those without (p > 0.05).

**Discussion**

It has been proposed that the clinical picture of KS gained during evaluation of infertility is a biased one and shows only the extreme case, whereas patients with more unobtrusive phenotypes can lead relatively normal lives (Zitzmann et al., 2004). Nevertheless, because spermatogenesis is affected by problems with meiosis in all KS patients, they are usually diagnosed with KS at infertility centers (Vernaeve et al., 2004). Our study indicates that even those presenting to infertility clinics may be heterogeneous in terms of karyotype and phenotype. Mosaicism is a state in which there is more than 1 population of cells within an individual, and these populations have different genotypes. In KS, this can include individuals with 46,XY/47,XXY, 47,XXY/48,XXXY, or other variations (Boada et al., 2009). It has been suggested that 46,XY/47,XXY patients with an increased incidence of XY cells show very few phenotypic features; the testis may be normal in size, endocrine abnormalities are less severe, and gynecomastia and azoospermia are less common (Lanfranco et al., 2004; Lenz et al., 2005; Seo et al., 2004). However, our results showed no significant differences in the frequency of gynecomastia, typical clinical signs of hypogonadism, or in the body proportions, testicular size, semen volume, and hormonal levels between patients with non-mosaic KS and mosaic KS. These results may reflect either the small sample size of mosaic KS patients studied, which is due to the lower prevalence of 46,XY/47,XXY mosaicism compared to nonmosaic 47,XXY. Alternatively, they might indicate that the degree of mosaicism measured in blood leukocytes may not be equivalent to that present in the testis, breast, or other tissues.
Variations in genetic polymorphisms of specific genes on the X chromosome, parent-of-origin of the extra X chromosome, genes that escape X inactivation, and the pattern of X inactivation may be common and could explain the wide range of phenotypic abnormalities observed in KS (Iitsuka et al., 2001). For example, androgen receptor (AR) CAGₜ repeats were found to be associated with increased height, decreased testicular volume, decreased bone density, and the presence of gynecomastia in 1 study (Zitzmann et al., 2004). However, this association has not been confirmed in other studies (Seo et al., 2004). In addition, the parental origin of the supernumerary X chromosome was not found to be associated with the phenotypes of penile length, testicular volume, or height (Seo et al., 2004). Unfortunately, our series was not subjected to analysis of AR CAGₜ repeat length, parent-of-origin of the extra X chromosome, genes that escape X inactivation, or the pattern of X inactivation.

It has been suggested that the phenotype of a patient diagnosed with KS in childhood or at puberty is different from that of a patient diagnosed with KS in adulthood (Zinn et al., 2005; Bojesen and Gravholt, 2007). Our results indicate that even in adulthood, phenotypes may vary, especially in those patients visiting infertility clinics. The current study shows that gynecomastia was present in nearly 20% of patients and that there was a significant decrease in T concentrations among those with gynecomastia compared to those without. However, 56.5% of patients had low total T. These findings indicate that T deficiency is not the only factor that induces gynecomastia in KS. Numerous pathological mechanisms could account for the estrogen-androgen imbalance that is proposed to cause gynecomastia. The complexities of estrogen and androgen metabolism, together with the extraglandular formation of estrogens that occurs via the enzymatic aromatization of androgens, suggest that several factors contribute simultaneously to this hormone imbalance (Braunstein, 2007). The frequency of gynecomastia in our study was less than that reported in other studies. For example, Vorona et al. (2007) reported a frequency of 38.6% in the patients that they studied. This difference could be due to racial and/or genetic factors. For example, an increase in aromatase activity has been reported in KS patients (Paduch et al., 2009). In addition, aromatase cytochrome P450arom polymorphisms might contribute to the incidence of gynecomastia (Czajka-Oraniec et al., 2008).

Although 56.5% of our patients had low total T, the frequency of the typical clinical signs of hypogonadism among the patients was 86%. This indicates that 14% of KS patients showed no typical clinical signs of hypogonadism, and at least 30% of patients showed typical clinical signs
of hypogonadism in spite of normal total T levels. These findings could be explained by the fact that serum total T may not reflect the true androgenic status of KS patients, for 2 reasons. First, total T levels may appear to be deceptively high, as serum hormone-binding globulin levels in patients with KS are elevated, and free T levels, which may provide a more accurate assessment of the androgen status in KS, are low (Wang et al., 1975). Second, individuals with hormone levels that are 2 SD below the population mean can still have adequate hormone concentrations to meet their metabolic needs (Harman et al., 2001).

A lack of androgens may contribute to fundamental deviations in body proportions in patients presenting with androgen deficiency before puberty (Schibler et al., 1974). The literature indicates either no difference in arm span to body height, or relatively long or relatively short arms among KS patients (Zitzmann et al., 2004; Varrela et al., 1984; Visootsak et al., 2001; Graner et al., 1982). Eunuchoid proportions, with arm span exceeding body height and lower body segment length exceeding the upper body segment length, were noted in 43.9% and 64.4% of patients, respectively. In addition, there was a significant negative correlation between total T and lower body segment length. Moreover, there were significant positive correlations between E2 and both arm span and lower segment length. There were also significant positive correlations between LH and arm span and height. These findings may indicate that androgen deficiency was present before puberty in a significant number of our patients. Relevantly, the role of estrogens in epiphyseal closure and skeletal proportions is crucial not only in women but also in men (Rochira et al., 2001).

In agreement with other reports (Paduch et al., 2009; Smyth and Bremner, 1998; Seo et al., 2004; Hsueh et al., 1978), the constant features of KS observed in our patients were small testes and azoospermia. These findings may be due to germ cell loss and interactions between gonadotropins and sex hormones. Our results showed evidence of positive correlations of these features with T, E2, and PRL and negative correlations with FSH and LH. Moreover, testicular size was positively correlated with semen volume. Almost all men with KS are azoospermic, but rarely, spontaneous paternity is reported (Terzoli et al., 1992). Intriguingly, whilst male secondary sexual deficiency is normally reversible by androgen replacement therapy, infertility in KS patients is intractable—at least it was until the advent of intracytoplasmic sperm injection (ICSI) (Tournaye et al., 1996). The success achieved with ICSI constituted a major breakthrough, to the extent that in the modern era of assisted reproductive technology, KS
patients need no longer be considered infertile (Fullerton et al., 2010). Surgical sperm retrieval has succeeded in recovering spermatozoa in up to 50% of patients with classic KS (Friedler et al., 2001). This recovery rate is similar to that in all men with non-obstructive azoospermia (Vernaeve et al., 2004).

In the current study, concentrations of LH and FSH were high in most cases. However, 3.7% and 3.3% of patients showed levels within the normal range for FSH and LH, respectively. Raised gonadotropin levels indicate germ cell damage, hypogonadism, and increased pituitary drive and may be a cause of Leydig cell hyperplasia. Although 45.5% of our patients had normal circulatory total T, there was inappropriate elevation of LH. In addition, there was an inverse correlation between total T levels and LH. These findings indicate that substances other than T are involved in the control of LH secretion. One of these substances may be E2, as evidenced by the inverse correlation between E2 levels and LH. However, the hypothesis that positive estrogen feedback occurs in KS as a result of removal of, or reductions in, testosterone has been refuted (Goh and Lee, 1998). On the other hand, normal serum gonadotropin levels in association with KS have been reported in the literature (Carter et al., 1977; Advani et al., 1991). This discrepancy could be explained by exhaustion of gonadotropin levels as a result of previous prolonged hypersecretion (Goh and Lee, 1998), or the finding may be a simple association. Alternatively, the response to LH-releasing hormone (LHRH) may be normal in these patients (Carter et al., 1977).

Although PRL-secreting tumors have been reported in KS patients (Kumanov et al., 1995; Pinto et al., 1996), hyperprolactinemia was of mild severity in our cases, and computed tomography scans of the pituitary were found to be normal. It is reasonable to assume that because of the loss of negative feedback effect, chronic stimulation of lactotroph cell type would result in hyperplasia, which in turn may transform into adenoma (Samaan et al., 1979). Therefore, it remains to be elucidated whether hyperplasia of the pituitary occurs secondary to gonadal failure (Samaan et al., 1979; Pinto et al., 1996) or is caused by a genetic abnormality linked to KS (Scheithauer et al., 2005). Although unlikely, the hyperplasia observed in KS patients may have been an incidental finding, having no causal relationship with hypogonadism.

The varied but relatively mild physical abnormalities observed in some patients explain why they do not receive clinical attention until adulthood, when they seek medical advice for small testes
or infertility. Diagnosis is also hindered by a low awareness of the disease among health professionals. Early diagnosis of KS improves a patient’s quality of life and enables better medical treatment, because these patients have a high risk of developing osteoporosis (van den Bergh et al., 2001), metabolic syndrome (Bojesen et al., 2010), and diabetes mellitus. KS has even been associated with mortality (Bojesen et al., 2004).

The major research limitation in this study was the failure to collect adequate cases of mosaic KS. This was due to ascertainment bias during the recruitment of patients. In addition, although 216 patients were enrolled in our study, all had presented with infertility. These factors might explain the lack of significant differences seen between mosaic and non-mosaic KS patients. To circumvent this problem, larger cohorts of unbiased patients should be investigated in the future. Lack of verification of hypogonadism through measurement of free T is another limitation of this study. Our study is also retrospective in nature and so includes all the well-known limitations of such studies. To limit the potential for bias, we used a standardized history and clinical examination form for all study participants. The retrospective design of the study did not allow us to examine any missed data. Patients with 2 or more missed variables were excluded from the study.

In conclusion, the results emphasize the heterogeneous clinical, endocrine, and genetic phenotype of infertile KS patients. Our findings support the usefulness of cytogenetic studies in infertile patients showing small testicular size and azoospermia, regardless of the presence of other clinical or endocrine findings. In addition, future studies on a sufficient number of infertile patients with mosaic KS are required. Further prospective genetic studies are required to delineate the molecular factors underlying the phenotypic variability in infertile KS patients; this may enable determination of an accurate prognosis in individual patients in the future.

**Declaration of Interest**

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of this report.

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References


Forestà, C., Galeazzi, C., Bettella, A., Marin P, Rossato M, Garolla A, Ferlin A. Analysis of


Zitzmann M, Depenbusch M, Gromoll J, Nieschlag E. X-chromosome inactivation patterns and androgen receptor functionality influence phenotype and social characteristics as well as
Figure 1 Shows the interphases 1, 2 and 3 from FISH analysis, there are two A signals of the two X chromosomes and one B signal of the Y chromosome in interphase number 1, two B signals of the two Y chromosomes and one A signal of the X chromosome in interphase number 2 and one A signal of X and one B signal of the Y chromosome in interphase number 3, confirming the mosaic form XXY/YYX/XY.

Figure 2 shows the clinical features in two patients with classic KS. A) It shows normal facial and body hair distribution with masculine features; B) The same patient in figure 3A shows normal pubic hair distribution, normal penile development but the testicular size is small. C) Another patient shows sparse facial and body hair with gynaecomastia ;D) The same patient in figure 2C shows sparse stumps of pubic hair and small testicular size.
### Table 1: Clinical and Laboratory Data of KS Patients

<table>
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<tr>
<th></th>
<th>Classic KS No = 198</th>
<th>Mosaic KS and variants No = 18</th>
<th>Significant test</th>
<th>P value</th>
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<tr>
<td><strong>Patient's age (year)</strong></td>
<td>30. (18-54)</td>
<td>37 (19-47)</td>
<td>Z=-2.195</td>
<td>.028</td>
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<tr>
<td><strong>Duration of infertility (year)</strong></td>
<td>3.0 (1-24)</td>
<td>5.0 (1-19)</td>
<td>Z=-1.21</td>
<td>.23</td>
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<td><strong>Age of the wife (year)</strong></td>
<td>24.0 (14.0-39.0)</td>
<td>27.50 (18.0-37.0)</td>
<td>Z=2.1</td>
<td>.038</td>
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<td><strong>Arm-span at examination (cm)</strong></td>
<td>184.0 (159.0-197)</td>
<td>180.0 (135-196)</td>
<td>Z=-1.526</td>
<td>.127</td>
</tr>
<tr>
<td><strong>Height at examination(cm)</strong></td>
<td>180.50 (156.0-194.0)</td>
<td>179.0 (144.0-184.0)</td>
<td>Z=-1.588</td>
<td>.112</td>
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<td><strong>Upper segment(cm)</strong></td>
<td>87.0 (76.0-103.0)</td>
<td>85.0 (70.0-90.0)</td>
<td>Z=-1.411</td>
<td>.158</td>
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<tr>
<td><strong>Lower segment (cm)</strong></td>
<td>92.0 (75.0-106.0)</td>
<td>92.0 (74.0-100.0)</td>
<td>Z=-.963</td>
<td>.335</td>
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<td><strong>Testicular size (ml)</strong></td>
<td>1.0 (.25-5)</td>
<td>1.0 (.25-2.5)</td>
<td>Z=-.952</td>
<td>.341</td>
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<td><strong>Ejaculate volume (ml)</strong></td>
<td>2 (0.2-7)</td>
<td>2 (.7-6)</td>
<td>Z=-.855</td>
<td>.393</td>
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<td><strong>Spermatozoa concentration (million /ml)</strong></td>
<td>Azoospermia</td>
<td>Azoospermia</td>
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<td>-</td>
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<td><strong>FSH (miu/ml)</strong></td>
<td>32.5 (3.3-84)</td>
<td>32.5 (5.5-47)</td>
<td>Z=-.74</td>
<td>.46</td>
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<td><strong>LH (miu/ml)</strong></td>
<td>16.0 (5.3-58)</td>
<td>16.5 (8.6-27)</td>
<td>Z=-.820</td>
<td>.412</td>
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<td><strong>Prolactin</strong></td>
<td>8.0 (2-44)</td>
<td>7.0 (3.6-22)</td>
<td>Z=-1.388</td>
<td>.165</td>
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<td><strong>T (ng/dl)</strong></td>
<td>225 (13-990)</td>
<td>195 (47-500)</td>
<td>Z=-.660</td>
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<td><strong>E2</strong></td>
<td>24.0 (7-207)</td>
<td>23 (8.5-200)</td>
<td>Z=-.173</td>
<td>.86</td>
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Normal ranges for Testosterone = 245-1600 ng/dl ; LH= 0.8-7.6 miu/ml ; Estradiol = 0.0-56 pg/ml and prolactin = 2.5-17 ng/ml.
Table 2: Karyotyping of patients with mosaic KS and KS variants

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<tr>
<td>47xxy/47xxy/46xy</td>
<td>1</td>
<td>17</td>
<td>68:21:11</td>
</tr>
</tbody>
</table>
Table 3: Frequency of different clinical and laboratory abnormalities in KS patients

<table>
<thead>
<tr>
<th>Clinical sign / Laboratory Abnormality</th>
<th>Total No= 216</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clinical signs of hypogonadism</td>
<td>186 (86%)</td>
</tr>
<tr>
<td>Gynecomastia</td>
<td>45 (20.8%)</td>
</tr>
<tr>
<td>Arm-span exceeds height by 5.3 cm (2.1 inch)</td>
<td>95 (43.9%)</td>
</tr>
<tr>
<td>Lower segment exceeds Upper segment by 5.3 cm (2.1 inch)</td>
<td>139 (64.4%)</td>
</tr>
<tr>
<td>Low total testosterone</td>
<td>122 (56.5%)</td>
</tr>
<tr>
<td>High FSH (miu/ml)</td>
<td>208 (96.3%)</td>
</tr>
<tr>
<td>High LH</td>
<td>209 (96.7%)</td>
</tr>
<tr>
<td>High Prolactin</td>
<td>19 (8.8%)</td>
</tr>
<tr>
<td>High E2</td>
<td>14 (6.5%)</td>
</tr>
</tbody>
</table>

Normal ranges for T = 245-1600 ng/dl; LH = 0.8-7.6 miu/ml; FSH = 0.7-11.1 miu/ml; Estradiol = 0.0-56 pg/ml and prolactin = 2.5-17 ng/ml.