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RESEARCH ARTICLE

Microscopic Testicular Sperm Extraction in Patients with Klinefelter Syndrome: Long-Term Outcomes from a Single Center

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Abstract

Objective: Klinefelter syndrome (KS) represents a sex chromosome anomaly observed in approximately 1 in 500–600 phenotypic males. It is observed in 3% of infertile males and up to 11.9% of azoospermic males. KS manifests in either non-mosaic (47, XXY) or mosaic (47, XXY/46, XY) forms, with 85% of cases presenting as the non-mosaic 47, XXY karyotype. The average rate of surgical sperm retrieval in patients with KS is around 50%, ranging from 28% to 69%. In this study, we aimed to present the outcomes of microscopic testicular sperm extraction (micro-TESE) in patients with non-mosaic KS.

Material and Methods: The results of 61 patients diagnosed with KS, who presented to the Harran University Urology Clinic with azoospermia between 2017 and 2024, were retrospectively reviewed. Hormonal assessments, including follicle-stimulating hormone (FSH), luteinizing hormone (LH), estradiol, and total testosterone (TT), were conducted for all patients, and their partners underwent gynecological evaluations for infertility. Testicular dimensions were recorded via scrotal ultrasonography. Patients were categorized into TESE-positive and TESE-negative groups, and parameters were compared between these groups.

Results: The mean age of the patients was 29.0±5.1 years, and their mean infertility duration was 5.9±4.1 years. The sperm retrieval rate was 29.5% (n=18). Mean levels of FSH, LH, prolactin, estradiol, and TT were 44.9 IU/L, 23.3 IU/L, 10 nmol/L, 31.4 pmol/dL, and 219 ng/dL, respectively. Sperm was retrieved in 18 patients (29.5%), while no sperm was obtained in 43 (70.5%). No significant correlation was observed between patient age, testicular size, serum levels of FSH, LH, prolactin, estradiol, and TT, and sperm retrieval rates when comparing the TESE-positive and TESE-negative groups (P>0.005).

Conclusion: In patients with non-mosaic KS, hormonal parameters, age, and infertility duration were not found to be significant predictors of the success of micro-TESE in sperm retrieval.

Keywords: klinefelter syndrome,, microscopic testicular sperm extraction, azoospermia

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INTRODUCTION

Infertility affects approximately 15% of the general population, with azoospermia identified in about 13% of those seeking treatment for infertility (1). Genetic analysis and hormonal evaluation are essential for azoospermic males due to underlying genetic and hormonal disorders.

Klinefelter syndrome (KS) is the most common sex chromosome anomaly, and it is occurring in approximately 1 in 500–600 phenotypic males. It is observed in 3% of infertile male patients and up to 11.9% of azoospermic males (2). KS can manifest in non-mosaic (47, XXY) or mosaic (47, XXY/46, XY) forms (3). Leydig cell dysfunction is also prevalent in men with KS, leading to lower testosterone levels compared to the general population (4).

Microscopic testicular sperm extraction (micro-TESE) is the gold standard treatment for sperm retrieval in azoospermic patients. The success rate of micro-TESE in the general population without genetic anomalies is approximately 50%. In patients with KS, sperm retrieval rates with micro-TESE range from 28% to 69%, with significant variations across studies. Several studies have shown that performing micro-TESE at an early age may improve sperm retrieval success (5), although this remains a topic of debate.

In this study, we aimed to present the outcomes of micro-TESE in patients with non-mosaic KS.

MATERIAL AND METHODS

The study was initiated after approval was obtained from the Harran University Ethics Committee on 27.05.2024 with session number 07 and decision number 05. In this study, the data of 61 patients with non-mosaic KS, who presented to the Urology Clinic of Harran University with azoospermia and underwent micro-TESE between 2017 and 2024, were retrospectively screened. According to the genetic analysis patients reported as 47 XXY were included in the study. Those who had previously TESE, varicocele and undescended testicle surgery and those receiving hormonal treatment were excluded from the study.

Detailed anamnesis, physical examination, semen analysis, and hormone profiles were assessed for all patients. Semen samples were obtained through masturbation following three to five days of sexual abstinence and collected in sterile containers. The presence of azoospermia was confirmed with at least two semen samples taken two weeks apart. All samples were centrifuged at 3,000 g, and the resuspended pellet was thoroughly examined. Physical examinations included assessments of testicular size, the presence of varicocele, the development of secondary sexual characteristics, signs of orchitis, and palpation of the vas deferens. Preoperatively, follicle-stimulating hormone (FSH), luteinizing hormone (LH), and total testosterone (TT) levels were measured, karyotype analysis was performed, the presence of Y microdeletion was investigated, and the results of previous sperm extraction interventions were evaluated. Testicular dimensions were measured using scrotal ultrasonography.

The micro-TESE procedure was performed under spinal anesthesia via a midline scrotal raphe incision. After passing through the dartos and tunica vaginalis, a longitudinal incision was made in the avascular area of the tunica albuginea. Testicular parenchymal dissection was conducted under a microscope with 18–22x magnification, selecting enlarged and opaque tubules. The testicular tissues obtained were examined by an embryologist under an inverted microscope for the presence of spermatozoa. The tunica albuginea was closed with continuous 5-0 nylon sutures. Following hemostasis, layers were closed anatomically, and patients were discharged on the postoperative first day.

The retrieved sperm cells were cryopreserved. Intracytoplasmic sperm injection (ICSI) was performed in a separate session using the obtained sperm cells. Pathological sampling was conducted on TESE-negative patients.

In the descriptive statistics of the data, mean, standard deviation, median lowest, highest, frequency and ratio values were used. The distribution of variables was measured by Kolmogorov-Smirnov, Shapiro-Wilk test. Independent sample t test was used in the analysis of quantitative independent data with normal distribution. Mann-Whitney U test was used in the analysis of quantitative independent data with non-normal distribution. Chi-square test was used in the analysis of qualitative independent data, and Fischer test was used when chi-square test conditions were not met. The effect level was investigated with univariate and multivariate logistic regression. SPSS 28.0 program was used in the analyses.

RESULTS

A total of 61 patients were included in the study. The mean age was 29.0 ± 5.1 years, and the mean duration of infertility was 5.9 ± 4.1 years. The sperm retrieval rate was 29.5% (n =

18). Mean levels of FSH, LH, prolactin, estradiol, and TT were 44.9 IU/L, 23.3 IU/L, 10 nmol/L, 31.4 pmol/dL, and 219 ng/dL, respectively (Table 1). Sperm was retrieved in 18 patients (29.5%), while no sperm was obtained in 43 patients (70.5%). No significant correlation was observed between patient age, testicular size, serum levels of FSH, LH, prolactin, estradiol, and TT, and sperm retrieval rates when comparing the TESE-positive and TESE-negative groups (P > 0.005).

Among the 43 patients reported as TESE-negative, 23 had Sertoli cell-only syndrome, 12 exhibited complete hyalinization with no seminiferous tubules observed, five had maturation arrest, and three showed hypospermatogenesis (Table 3).

Postoperative complications included wound infection in three patients and scrotal hematoma in one patient, all of which were managed with medical treatment.

Table 3. Pathology results of TESE-negative patients

	n (%)
Hypospermatogenesis	3 (6.9%)
Maturation arrest	5 (11.6%)
Sertoli cell-only syndrome	23 (53.4%)
Complete fibrosis	12 (27.9%)

Table 1. Demographic and clinical characteristics of participants

		Min-Max			Median	Mean ± SD/n, %		
Age		21.0	-	43.0	29.0	29.0	±	5.1
Infertility duration		1.0	-	25.0	5.0	5.9	±	4.1
FSH		30.1	-	90.6	43.0	44.9	±	17.5
LH		12.6	-	39.5	23.4	23.3	±	8.0
Testosterone		38.4	-	471.3	201.9	219.0	±	111.5
Prolactin		4.7	-	19.8	9.4	10.0	±	3.2
Estradiol		7.5	-	63.0	31.3	31.4	±	11.8
Testicular volume (cc)		10.0	-	23.0	17.0	16.3	±	3.6
TESE	(-)					43		70.5%
	(+)					18		29.5%

SD: standard deviation, FSH: follicle-stimulating hormone, LH: luteinizing hormone, TESE: testicular sperm extraction

Table 2. Comparison of demographic and clinical characteristic between the study groups

	TESE (-)					TESE (+)				p	
	Mean ± SD n = 43 (70.5%)			Median		Mean ± SD n = 18 (29.5%)			Median		
Age	29.0	±	5.3	29.0		29.2	±	4.7	28.5	0.680	m
Infertility duration	5.9	±	4.6	5.0		5.8	±	2.6	6.0	0.609	m
FSH	46.4	±	19.0	44.0		41.1	±	13.1	37.6	0.217	m
LH	23.9	±	8.5	23.4		21.9	±	6.7	23.4	0.390	t
Testosterone	278.0	±	123.5	269.2		279.2	±	97.6	269.9	0.712	m
Prolactin	10.2	±	3.4	9.8		9.5	±	2.8	9.0	0.704	m
Estradiol	30.9	±	11.9	30.6		32.6	±	11.7	35.0	0.605	t
Testicular volume	16.2	±	3.7	16.0		16.7	±	3.3	17.5	0.617	m

[t]*Independent-samples t-test* / [m]*Mann-Whitney U test*

TESE: testicular sperm extraction, SD: standard deviation

DISCUSSION

KS is the most prevalent sex chromosome anomaly (6). Due to fibrotic testes, patients with KS typically exhibit low testosterone levels and elevated serum FSH and LH levels, consistent with primary testicular failure. Approximately 11–14% of azoospermic patients are diagnosed with KS. Due to testicular atrophy and fibrosis, the micro-TESE method is employed to retrieve sperm in azoospermic patients with KS. The first positive TESE procedure in patients with KS was described by Tournaye et al. in 1996, followed by TESE + ICSI and the subsequent report of the first successful pregnancy (7-10).

The literature reports a wide range of sperm retrieval rates via micro-TESE in patients with KS, ranging from 28% to 69% (11-13). In the current study, the TESE positivity rate in patients with non-mosaic KS was 29.5% (n = 18/61). Many studies have administered preoperative hormone therapy to patients before performing the TESE procedure, which contributes to varying sperm retrieval rates. In a comparative study by Guo et al., patients received preoperative human chorionic gonadotropin (hCG) therapy. The authors found no statistical difference between the treated and untreated groups in terms of sperm retrieval rates (44% vs. 43.3%) (14). In another study, Majzoub et al. reported that sperm retrieval was achieved in 27.8% of patients given aromatase inhibitors and 12.5% of those given clomiphene citrate plus hCG, while no sperm was retrieved in the untreated group. However, that study had a limited sample size, which is a notable limitation (15). Ramasay et al. found that patients who responded to hormone therapy with testosterone levels exceeding 250 ng/dL had sperm retrieval rates of 77%, compared to 55% in those who did not respond to therapy (16).

Several parameters are investigated to predict sperm retrieval success in patients with KS, but no definitive predictors have been established in the literature. Nevertheless, studies suggest that performing TESE at an earlier age yields more successful outcomes in this patient population (17,18). Liu et al. determined that testicular volume, FSH, LH, and testosterone were not predictors of sperm retrieval success in patients with KS. Instead, patient age and anti-Müllerian hormone levels showed some predictive value (19). Another study reported a micro-TESE success rate of 37.8% in patients with KS, with preoperative testosterone levels being the most significant predictor when comparing successful and unsuccessful groups (20). A study from Turkey involving 67 patients with KS found that early-age TESE increased sperm retrieval success, while other parameters, such as FSH, LH, prolactin, and TT levels, did not show any significant differences (21). In our study, no significant differences were observed in hormone levels, age, infertility duration, or testicular volume between the TESE-positive and TESE-negative patients. Sperm retrieval rates vary in the literature and in our study, sperm retrieval rates were close to the lower limit. The main factors that can cause and this can be interpreted as the older age of our patient population and the lack of any preoperative hormonal treatment. However, according to the guidelines, still there is currently no hormonal treatment method that can be applied before TESE to increase the sperm retrieval rates (22).

The primary factors negatively affecting sperm retrieval in patients with KS are testicular hyalinization and fibrosis. Histopathological examinations in these patients frequently reveal sclerosis, hyalinization in seminiferous tubules, and Sertoli cell-only syndrome (23). Studies have shown that testicular hyalinization increases with age, which decreases sperm retrieval rates in patients with KS, suggesting that earlier micro-TESE may have higher success rates (18). In the current study, no significant differences were found between the TESE-positive and TESE-negative groups. We found that 53.4% of the patients had Sertoli cell-only syndrome, and 27.9% had complete fibrosis, which is consistent with previous studies.

The limitations of our study include its retrospective nature, the limited number of patients, and the absence of an evaluation of ICSI, fertilization, and live birth outcomes.

CONCLUSION

The success rate of micro-TESE in patients with non-mosaic KS varies widely in the literature. The current study reveals that a rate of 29.5%. Hormonal parameters, age, and infertility duration were not found to be significant predictors of sperm retrieval success.

Ethics Committee Report: The study was initiated after approval was obtained from the Harran University Ethics Committee on 27.05.2024 with session number 07 and decision number 05.

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