Original Article

Sperm extraction in nonmosaic Klinefelter syndrome patients: A case series and literature review of sperm extraction in Klinefelter syndrome patients

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Abstract

Objectives: Klinefelter syndrome (KF) is a group of chromosomal disorders with at least one extra X chromosome in male individuals that leads to infertility and diminished hair growth in affected males. In this study, we present a case series of 16 nonmosaic KF and an extensive literature review.

Patients and Methods: This is a retrospective study including 16 nonmosaic Klinefelter Syndrome patients that underwent micro-testicular sperm extraction (m-TESE) at our center between January 2016 and December 2022. Frequencies and percentages were used to present categorical variables, whereas continuous variables were presented as the median and interquartile range (IQR). The sperm retrieval rate (SRR) was assessed using a one-sample proportions test with continuity correction. Fisher's exact test was to assess the differences between patients with negative and positive retrieval in terms of the categorical variables. A Wilcoxon rank-sum test was applied to explore the between-group differences in the numerical variables. A literature search was performed for additional publications of discussing m-TESE among KF patients.

Results: The median (IQR) age of patients was 40.0 years (34.5–47.0). All of the patients had nonobstructive azoospermia, and the majority of them (93.8%) had primary infertility. The most common histopathological findings were atrophic tubules (57.1%), followed by Sertoli cell-only (28.6%). Sperm retrieval was positive for two patients with a rate of 12.5% (95% confidence interval 2.2 to 39.6). Patients with positive sperm retrieval were significantly younger than their peers with negative retrieval (median = 28.0, IQR = 27.5 to 28.5 vs. median = 41.5, IQR = 35.8 to 47.0, $P = 0.031$). The successful conception rate was 100% ($n = 2$) using intracytoplasmic sperm injection with a birth rate of 100% ($n = 2$).

Conclusion: Our observed SRR among nonmosaic KF patients was marginally lower than the reported literature. Younger-age patients were significantly more likely to benefit from the procedure.

Keywords: Azoospermia, infertility, Klinefelter syndrome, micro testicular sperm extraction, sperm retrieval

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INTRODUCTION

In 1942, a case series of 9 men who had enlarged breasts, small-size testes, along with infertility and limited hair growth, was published by Dr. Harry Klinefelter.[1] This was initially considered a hormonal disorder but was eventually found to be a chromosomal defect later in the 1950s.[2] The disorder was named Klinefelter syndrome (KF). It is not an inherited disorder. It is distinguished as a group of chromosomal disorders with minimally one extra X chromosome compared to normal male karyotype, which results in a47, XXY karyotype. Patients with this syndrome suffer from infertility, with 90% of those patients having nonobstructive azoospermia.[3]

Patients with KF have symptoms of reduced testosterone level, increased luteinizing hormone (LH) level, high levels of estradiol, and follicle-stimulating hormone (FSH). Physical appearance reveals a tall stature with narrow shoulders, reduced muscle tone, and decreased body hair. Furthermore, those patients are more prone to autoimmune diseases, including diabetes mellitus, systemic lupus erythematosus, and neoplasms.[4] This syndrome is prevalent in 3% to 4% of men with infertility and diagnosed in more than 10% of azoospermic men.[5] Various approaches are used to retrieve the sperms in such patients, which will eventually result in successful pregnancy and live childbirth. The current review aimed to evaluate various studies concerning KF patients with respect to sperm retrieval rate (SRR), pregnancy rate and live birth rate. Further studies to minimize bias and give more reliable data in this regard are required.

PATIENTS AND METHODS

This is a retrospective study including 16 Nonmosaic Klinefelter Syndrome patients that underwent micro-testicular sperm extraction (m-TESE) at our center between January 2016 and December 2022. Data were analyzed using RStudio (R version 4.2.2.). We used frequencies and percentages to present categorical variables, whereas continuous variables were presented as the median and interquartile range (IQR). The SRR was assessed using a one-sample proportions test with continuity correction, and the estimated rate was expressed along with the respective 95% confidence intervals (95% CIs). Due to the small sample size, we used a Fisher's exact test to assess the differences between patients with negative and positive retrieval in terms of the categorical variables. In addition, a Wilcoxon rank-sum test was applied to explore the between-group differences in the numerical variables (age and body mass index). A $P < 0.05$ indicated statistical significance.

An extensive search was conducted online for available literature regarding KF and associated outcomes such as SRR, pregnancy rate, and live birth rate in patients suffering from KF. The research included English articles and studies conducted on humans. Studies from three different databases, including MEDLINE, Cochrane, and EMBASE, were evaluated from 2005 to 2022. Only studies that examined the three outcomes of interest were included. In total, 15 studies were included in this review. Our primary sources for information for the current review were published articles.

The study was approved by the Institutional Review Board of King Abdullah International Medical Research Center, Ministry of National Guard-Health Affairs, Riyadh, Kingdom of Saudi Arabia (approval number NRC21R/218/04). Serial numbers were used instead of the medical record number to ensure confidentiality. Due to the retrospective nature and the use of anonymized patient data, the requirement for consent was waived.

RESULTS

Demographic characteristics and clinical history of patients

Data from a total of 16 patients with nonmosaic KF were analyzed in the current study. The median (IQR) age of patients was 40.0 years (34.5–47.0). All of the patients had nonobstructive azoospermia, and the majority of them (93.8%) had primary infertility. Only one patient had hypogonadism (6.2%) and undescended testicles (6.2%). A history of previous testicular sperm extraction (TESE) attempts was positive for one patient [6.2%, Table 1].

Preoperative and operative characteristics

Regarding the laboratory parameters, the majority of patients had a preoperative FSH of >7.6 (93.8%) and preoperative testosterone of >3 (73.3%), and all the patients had preoperative LH of >7 (100%). Approximately one-third of patients had a testicular volume of ≥4.6 (31.2% in the right and left testicles). Ultrasound findings revealed varicocele among five patients (31.2%). Surgeries were performed for 62.5%, and bilateral surgeries were carried out in 75.0% of them [Table 2]. The most common histopathological findings were atrophic tubules with (57.1%) followed by Sertoli cells only [28.6%, Figure 1].

Outcomes of surgeries

Sperm retrieval was positive for two patients with a rate of 12.5% [95% CI 2.2 to 39.6, Figure 2]. Patients with positive sperm retrieval were significantly younger than their peers with negative retrieval (median = 28.0,

Table 1: Demographic characteristics and clinical history of patients

Parameter	Category	*n*=16, *n* (%)
Age	Median (IQR)	40.0 (34.5–47.0)
BMI	Median (IQR)	27.0 (23.8–33.0)
Type of infertility	Primary	15 (93.8)
	Secondary	1 (6.2)
Type of azoospermia	Obstructive	0
	Nonobstructive	16 (100.0)
Medications	No	9 (56.2)
	Clomid	4 (25.0)
	Arimidex	2 (12.5)
	Testosterone	1 (6.2)
Related history	No	14 (87.5)
	Epididymitis and/or orchitis	0
	Hypogonadism	1 (6.2)
	UDT	1 (6.2)
Family history	No	15 (93.8)
	Brother	1 (6.2)
Past surgical history	No	11 (68.8)
	TESA	2 (12.5)
	TESE	0
	Varicocelectomy	0
	Orchiopexy	3 (18.8)
Previous m-TESE attempts	No	15 (93.8)
	One	1 (6.2)
	Two	0
	More than two	0
Previous m-TESE	Yes	1 (6.2)

BMI: Body mass index, IQR: Interquartile range, TESE: Testicular sperm extraction, TESA: Testicular sperm aspiration, UDT: Undescended testicle, m-TESE: Micro-TESE

Table 2: Preoperative and operative characteristics

Parameter	Category	*n*=16, *n* (%)
Preoperative FSH	Median (IQR)	32.5 (24.2–38.5)
Preoperative FSH	≤7.6	1 (6.2)
	>7.6	15 (93.8)
Preoperative LH	Median (IQR)	16.0 (15.0–19.0)
Preoperative LH	≤7	0
	>7	16 (100.0)
Preoperative testosterone*	Median (IQR)	8.0 (4.0–12.5)
Preoperative testosterone*	<2	2 (13.3)
	2–3	2 (13.3)
	>3	11 (73.3)
Right testicular volume preoperative	Median (IQR)	3.5 (2.0–6.5)
Right testicular volume preoperative	<4.6	11 (68.8)
	≥4.6	5 (31.2)
Left testicular volume preoperative	Median (IQR)	3.0 (2.0–6.0)
Left testicular volume preoperative	<4.6	11 (68.8)
	≥4.6	5 (31.2)
US findings	Not done	2 (12.5)
	Normal	9 (56.2)
	Left varicocele	4 (25.0)
	Bilateral varicocele	1 (6.2)
	Others	0
Hormonal therapy preoperative	Yes	7 (43.8)
Hormonal therapy postoperstive	Yes	1 (6.2)
Site	Right	1 (6.2)
	Left	3 (18.8)
	Bilateral	12 (75.0)

*The variable has one missing value. IQR: Interquartile range, FSH: Follicle-stimulating hormone, LH: Luteinizing hormone, US: Ultrasound

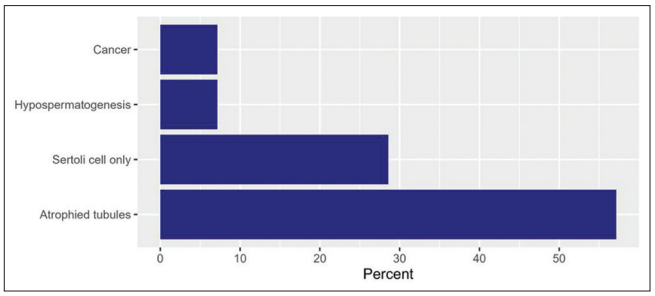

Figure 1: The proportions of histopathological findings for patients under study

IQR = 27.5 to 28.5 vs. median = 41.5, IQR = 35.8 to 47.0, $P = 0.031$). No other characteristics differed between patients with positive and negative retrieval, including the demographic, clinical, preoperative, and operative characteristics [Table 3]. Successful conception rate was 100% ($n = 2$) using intracytoplasmic sperm injection with a birth rate of 100% ($n = 2$).

Fifteen studies, which included SRR, pregnancy rate, and live birth rate, were included in the review. Out of 15 studies, 11 studies were retrospective and 4 studies were prospective. Information collected from these studies included the number of patients in each study, number of total procedures performed, surgical procedure used (SRR), pregnancy rate, live birth, and mean age of patients included in each study. Studies of both procedures, i.e., mTESE and conventional-TESE (c-TESE) or a mixture of both of these procedures, were included. Out of those 15 studies, 8 (53.3%) studies used the mTESE technique, 3 studies (20%) used the cTESE technique, and 4 studies (26.7%) used both of these techniques for sperm retrieval. The outcomes of all these studies are summarized in Table 4.

In our findings, sperm retrieval was positive with a rate of 12.5% (95% CI 2.2 to 39.6). Patients with positive sperm retrieval were significantly younger than their peers with negative retrieval. The total number of patients in the 15 studies included in the current review was 734, of which sperm retrieval was successfully achieved in 327 patients, giving a SRR of 44.55%. Depending on the procedure adopted for sperm extraction, SRR was observed as 47.13% using the mTESE approach, 37.75% using the cTESE approach, and 42.21% using the mixed approach for retrieval of sperms. Similarly, the overall pregnancy rate for all studies included was 24.38%, with specific approach pregnancy rates of 22.42%, 27.13%, and 27.55% for mTESE, mixed, and cTESE sperm extraction techniques, respectively. Live childbirth rates observed were 24.42% overall, with 21.51%, 29.14%, and 28.57% for mTESE, mixed, and cTESE sperm extraction techniques, respectively.

Table 3: Statistical differences between patients with positive and negative sperm retrieval

Parameter	Category	Sperm retrieval		
		Negative (*n*=14), *n* (%)	Positive (*n*=2), *n* (%)	*P*
Age	Median (IQR)	41.5 (35.8–47.0)	28.0 (27.5–28.5)	0.031
BMI	Median (IQR)	25.5 (23.2–32.5)	31.5 (29.2–33.8)	0.299
Type of infertility	Primary	13 (92.9)	2 (100.0)	>0.999
	Secondary	1 (7.1)	0	
Medications	No	8 (57.1)	1 (50.0)	0.400
	Clomid	4 (28.6)	0	
	Arimidex	1 (7.1)	1 (50.0)	
	Testosterone	1 (7.1)	0	
Related history	No	12 (85.7)	2 (100.0)	>0.999
	Epididymitis and/or orchitis	0	0	
	Hypogonadism	1 (7.1)	0	
	UDT	1 (7.1)	0	
Family history	No	13 (92.9)	2 (100.0)	>0.999
	Brother	1 (7.1)	0	
Past surgical history	No	9 (64.3)	2 (100.0)	>0.999
	TESA	2 (14.3)	0	
	TESE	0	0	
	Varicocelectomy	0	0	
	Orchiopexy	3 (21.4)	0	
	Previous m-TESE	1 (7.1)	0	
Preoperative FSH	≤7.6	0	1 (50.0)	0.125
	>7.6	14 (100.0)	1 (50.0)	
Preoperative LH	≤7	0	0	>0.999
	>7	14 (100.0)	2 (100.0)	
Preoperative testosterone	<2	2 (15.4)	0	>0.999
	2–3	2 (15.4)	0	
	>3	9 (69.2)	2 (100.0)	
Preoperative right testicular volume	<4.6	10 (71.4)	1 (50.0)	>0.999
	≥4.6	4 (28.6)	1 (50.0)	
Preoperative left testicular volume	<4.6	10 (71.4)	1 (50.0)	>0.999
	≥4.6	4 (28.6)	1 (50.0)	
US findings	Not done	2 (14.3)	0	>0.999
	Normal	7 (50.0)	2 (100.0)	
	Left varicocele	4 (28.6)	0	
	Bilateral varicocele	1 (7.1)	0	
	Others	0	0	
Preoperative hormonal therapy	Yes	6 (42.9)	1 (50.0)	>0.999
Postoperative hormonal therapy	Yes	1 (7.1)	0	>0.999
Site	Right	1 (7.1)	0	0.450
	Left	2 (14.3)	1 (50.0)	
	Bilateral	11 (78.6)	1 (50.0)	

IQR: Interquartile range, TESE: Testicular sperm extraction, TESA: Testicular sperm aspiration, UDT: Undescended testicle, FSH: Follicle-stimulating hormone, LH: Luteinizing hormone, US: Ultrasound, m-TESE: Micro-TESE

DISCUSSION

In our findings, sperm retrieval was positive with a rate of 12.5% (95% CI 2.2 to 39.6). Patients with positive sperm retrieval were significantly younger than their peers with negative retrieval. In our literature review, a 44.55% successful sperm extraction rate was reported in this population irrespective of clinical parameters, which is higher than what we reported in our cohort. We hypothesize that the lower SRR is attributed to two factors: First, our patients were nonmosaic KF. Second, the median age of our study population, 40.0 (34.5–47.0), is marginally higher than other studies reporting in the literature. This is also an indication that most of our patients have a late presentation making outcomes less optimal.

The first successful extraction was reported in 1996 through c-TESE approach by Tournaye *et al*.[21] After that, the first pregnancy after TESE in KF was reported in 1998.[22] Prediction of a successful m-TESE approach is still controversial even after 22 years. Depending upon the hyalinization of seminiferous tubules in the testes of patients suffering from this syndrome, it is hypothesized by various studies that early TESE approach adaptation may lead to better results of sperm extraction and thus lead to pregnancy with live childbirth.[23,24] There are various factors that interfere with sperm extraction in patients with KF. Those factors may include age, hormonal factors, and testicular volume. In our cohort of 16 patients, younger patients had a significantly higher chance of a successful m-TESE. These factors may aid in the prognosis of SRR in patients of KF.[23,25,26]

Table 4: Outcomes of studies included in the review

Study	Study type	Number of patients	Number of total procedures	Surgical procedure	SRR (%)	Pregnancy rate (*n*)	Live birth (*n*)	Mean age (year)
[6]	Prospective	10	10	m-TESE	6	4	3	-
[7]	Retrospective	51	51	Mixed (m-TESE and c-TESE)	26	12	12	34.4
[8]	Retrospective	42	54	m-TESE	29	19	21	32.8
[9]	Prospective	17	17	c-TESE	6	7	8	35.0
[10]	Prospective	26	26	m-TESE	13	4	2	36.0
[11]	Retrospective	27	27	Mixed (m-TESE and c-TESE)	8	4	5	32.3
[12]	Retrospective	68	91	m-TESE	45	33	28	33
[13]	Retrospective	33	39	m-TESE	22	7	5	32
[14]	Retrospective	106	106	m-TESE	50	26	29	34.3
[15]	Prospective	38	38	Mixed (m-TESE and c-TESE)	15	15	16	35.3
[16]	Retrospective	18	18	m-TESE	3	1	1	30.3
[17]	Retrospective	65	65	c-TESE	25	16	17	33.8
[18]	Retrospective	134	134	m-TESE	38	4	5	32.6
[19]	Retrospective	16	16	c-TESE	6	4	3	32.1
[20]	Retrospective	83	88	Mixed (m-TESE and c-TESE)	35	23	25	33.7

TESE: Testicular sperm extraction, m-TESE: Micro-TESE, SRR: Sperm retrieval rate, C-TESE: Conventional TESE

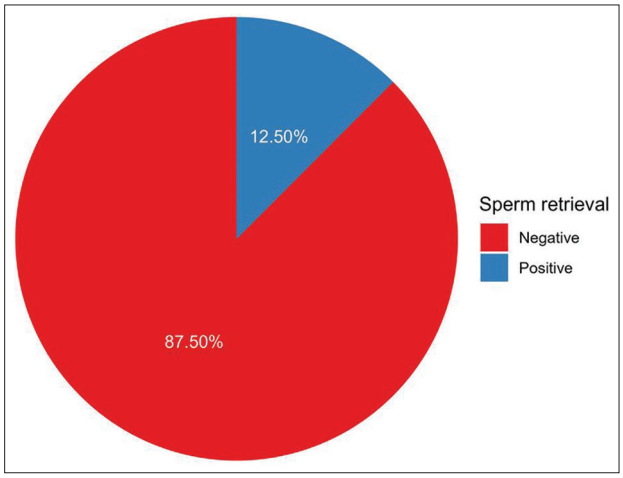

Figure 2: The proportions of sperm retrieval statuses among patients under study

From the studies included in this review, we observed success rates of sperm extraction by two different techniques and a mixture of both. More success rate for extraction of sperm was observed in case studies that adopted the mTESE approach (47.13%) as compared to cTESE (37.75%) and mixed approach (42.21%). Increased SRR was also reported similarly in both adults and adolescents with KF in other studies.[25,27]

The results of this review also suggest that KF patients undergoing the TESE approach have 20%–28% chance to have a pregnancy and live birth of a child. These findings are higher those of a meta-analysis study where live childbirth was reported in 16% of the patients who adopted the TESE approach for extraction of sperms.[21]

The use of different therapy options, such as testosterone, in KF patients has been controversial as it is reported to have a negative impact on fertility in the future.[8] Contrary to this, various other studies reported that the use of testosterone supplementation with other drugs was beneficial before undergoing the TESE procedure. These studies reported an improved retrieval rate of sperm in KF patients.[28,29] Nevertheless, enough data to support the use of testosterone or any other treatment option was not found; thus, more research is required to ascertain the role and effectiveness of such therapies.

CONCLUSION

Our observed SRR was marginally lower than the reported literature. Younger-age patients were significantly more likely to benefit from the procedure. We hypothesize that the lower rate is mainly due to our patient population composing of nonmosaic patients. From the analysis of the current literature, we concluded that sperm extraction in KF patients was quite successful and significantly higher than what's found in our cohort. Using both approaches of sperm retrieval, the overall rate is more than 40% in this special population. Timely use of mTESE and cTESE approaches in such patients leads to successful pregnancy and, thus, live birth of a child in almost one-fourth of patients. Further studies to minimize bias and give more reliable data in this regard are required.

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Conflicts of interest
There are no conflicts of interest.

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