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Testicular microdissection following failed sperm aspiration: a single-center experience

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Abstract. – OBJECTIVE: Azoospermia is a cause of infertility in a subgroup of infertile men. Sperm retrieval techniques including testicular sperm aspiration (TESA) and microscopic testicular sperm extraction (mTESE) are widely used. In this study, we have reviewed our findings regarding mTESE performed following a negative TESA outcome.

PATIENTS AND METHODS: This is a retrospective chart review study that included 41 infertile patients who underwent mTESE after a negative TESA outcome. Charts were reviewed for demographic data, type of infertility, and type of azoospermia. Hormone level analysis was done for follicle-stimulating hormone, luteinizing hormone, and testosterone. Testicular volume was estimated by ultrasound.

RESULTS: The study included 41 patients who underwent mTESE following a negative TESA outcome. Most patients had primary infertility (n = 32; 78%). Of the 41 patients, four had a previous history of either TESE or orchidopexy, and two had a history of varicocelectomy before the recent percutaneous TESA procedure. There was no significant association between sperm retrieval and the different surgical procedures that had been performed. Of the 41 patients, 27 had positive sperm retrieval by mTESE with a success rate of 65.9%.

CONCLUSIONS: The positive sperm retrieval rate of mTESE performed following a negative TESA outcome was reasonable (65.9%). No significant correlations were identified with all variables studied.

Key Words:
Microscopic testicular sperm extraction, Testicular sperm aspiration, Male infertility, Azoospermia.

Abbreviations

TESA: Testicular sperm aspiration; mTESE: Microscopic testicular sperm extraction; cTESE: Conventional Testicular sperm extraction; SRR: Sperm retrieval rate; LH: Luteinizing hormone. FSH: Follicle-stimulating hormone. IVF: *In vitro* fertilization.

Introduction

Azoospermia is defined as the absence of sperm in the ejaculate. It is a major cause of infertility and it is found in approximately 1 to 2% of all population and 10 to 20% of infertile couples. Azoospermia was classified into two types: obstructive, which is caused by an obstruction in the ductal system or congenital absence of vas deferens and/or seminal vesicles which will prevent the transport of sperms from the testis to the ejaculate; non-obstructive, which means impaired spermatogenesis and failure of the testis to produce sperms in the ejaculate[1].

In obstructive azoospermia (OA), fertility can be achieved either by reconstructive surgery or by sperm retrieval techniques. Percutaneous Epididymal Sperm Aspiration (PESA) and Testicular Sperm Extraction (TESE) were usually performed for sperm retrieval in patients with OA or ejaculatory dysfunction[2,3]. In a recent meta-analysis of eight studies including more than 2,000 men diagnosed with OA, Shih et al[2] reported comparable pregnancy and miscarriage rates between PESA and TESE. Nevertheless, the implantation rate was significantly higher (odds ratio of 1.58) following TESA *vs.* PESA.

In non-obstructive azoospermia (NOA), sperm retrieval surgeries, such as testicular sperm aspiration (TESA) and microscopic testicular sperm extraction (mTESE), are the mainstay of treatment[4]. The decision to use TESA or mTESE

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for sperm retrieval is based on three important factors: clinical presentation, patient preference, and treating urologist's preference. However, TESA was associated with a lower sperm retrieval rate (SRR, 11-60%)[5-7], compared with mTESE (45-63%)[8,9]. Furthermore, a recent level 1 evidence study by Jensen et al[10] showed higher SRR following mTESE compared with the multiple needle-pass testicular sperm aspiration (TESA) in patients with NOA. Therefore, most of the patients who fail TESA will eventually opt for mTESE. This study aimed at assessing the outcome of mTESE after a negative TESA and identifying predictors of a positive outcome.

Patients and Methods

This is a single-center retrospective study that was conducted in a tertiary center (King Abdulaziz Medical City, Riyadh, Saudi Arabia) with a dedicated *in vitro* fertilization unit (IVF) between January 2014 and January 2019. All male patients who were diagnosed with azoospermia and underwent TESA with negative outcomes followed by mTESE were included. Azoospermia patients with positive TESA and those with negative TESA who did not undergo mTESE were excluded.

Patient charts were reviewed for age, body mass index (BMI), type of infertility, previous urological surgeries, type of azoospermia, hormone analysis, testicular volume, and the outcome of mTESE. Testicular volume was estimated by testicular ultrasound. Hormone levels included were follicle-stimulating hormone (FSH), luteinizing hormone (LH), and testosterone level. Diagnosis of azoospermia was based on two semen analyses performed after 3-4 days of abstinence. Hormonal treatment was offered after TESA failure in some cases.

Testicular Sperm Aspiration

After examining both testes, the urologist proceeded to perform a spermatic cord block on the side of the relatively larger testis using 5 cc of marcaine and 5 cc of bupivacaine. The scrotal skin was also infiltrated. A 16-gauge angiocath needle was used to aspirate from the testis. The aspirated fluid was sent to the *in vitro* fertilization (IVF) laboratory, to confirm the presence of spermatocytes by microscopy. The testis was aspirated three times. In case of failure, the contralateral testis was aspirated in the same manner. Failure was defined as the inability to aspirate any spermatocytes or the aspiration of severely malformed spermatozoa that could not be used in intracytoplasmic sperm injection.

Microscopic Testicular Sperm Extraction

The mTESE procedure was performed three months after the initial TESA to allow the tunica albuginea to heal. It was performed under general anesthesia using the operating microscope. A midline scrotal incision was made, and the layers of the scrotum were incised over the larger testis. Through an incision in the tunica albuginea, large opaque tubules were dissected and sent to the IVF laboratory.

Statistical Analysis

Data were analyzed using SPSS version 20.0 (IBM Corp., Armonk, NY, USA). Categorical variables were presented as frequencies and percentages, and numerical variables were described as mean and standard deviation. The Chi-square test was used to perform a subgroup analysis for continuous variables. A p-value of < 0.05 indicate statistical significance.

Results

A total of 226 patients had undergone TESA during the study period; 168 patients were excluded as they had a positive TESA outcome or did not undergo mTESE after the negative TESA. Only 41 patients had a negative TESA outcome and underwent mTESE later on. Therefore, a total of 41 patients were enrolled in the study Among enrolled patients, the mean age was 40.29±8.69 years. The mean BMI was 30.9±5.69. Overall, testicular units were 82 and no patient had a solitary (unilateral) testis in our study. Most patients presented with primary infertility (32, 78%). The majority of patients were diagnosed with NOA (37, 90.2%). Prior urological interventions before the failed TESA included: four patients who had cTESE, four who had orchidopexy, and two who had varicocelectomy. Of the 41 patients (82 testicular units) patients who had previously obtained a negative TESA outcome, 27 patients (54 testicular units) had positive sperm retrieval by mTESE, with a success SRR of 65.9%. All patients who had a prior history of orchidopexy had positive sperm retrieval by mTESE, whereas one of the two patients who had undergone varicocelectomy

had positive sperm retrieval. Of the four patients who had previously undergone cTESE, three had positive sperm retrieval. The patient characteristics are shown in Table I.

The median levels of different hormones investigated (LH, FSH, and testosterone) were as follow: the median LH level was 6.30 IU/L, the median FSH level was 19.07 IU/L, and the median testosterone level was 11.30 nmol/L. The right and left testicular volumes were measured by testicular ultrasound. Overall, bilateral testicular units were 82, and the bilateral testicular volume ≥ 10 mL was found in more than two-thirds of patients. The investigational characteristics are shown in Table II.

Overall, in both testicular units, patients with testicular volume ≥ 10 showed higher positive SRR compared to negative mTESE. However, this was not statistically significant (Table III).

Patients with FSH levels >18 IU/L had a higher positive sperm retrieval rate (SRR) (78.3%) in mTESE. However, this was statistically insignificant compared to negative mTESE (p-value = 0.058). Similarly, patients with testosterone levels ≥ 10 nmol/L had a higher positive SRR (59.3%) in mTESE. However, this was statistically insignificant (p-value = 0.305) (Table IV).

Table I. Patients' characteristics.

Variables (n = 41)	Mean ± SD or number (%)
Age (year)	40.29 ± 8.69
BMI	30.90 ± 5.69
Testicular Unit	
Bilateral	82 (100)
Unilateral	0
Type of Male Infertility	
Primary	32 (78.0)
Secondary	9 (22.0)
Type Azoospermia	
Obstructive	4 (9.8)
Non-Obstructive	37 (90.2)
Previous interventions	
cTESE	4 (9.8)
Varicocelectomy	2 (4.9)
Orchidopexy	4 (9.8)
Patient number with positive SR after (mTESE)	27 (65.9)
Overall testicular units with positive SR after (mTESE)	54 (56.9)

SR= sperm reterival; mTESE = microscopic testicular sperm extraction.

Discussion

Approximately 1% of the general population suffers from azoospermia, and 10% of men assessed for infertility have NOA[11]. In 1999, in the era of advancing male infertility management strategies, a unique surgical technique called microscopic testicular dissection was introduced by Prof. Schlegel causing a breakthrough in sperm retrieval[9,12]. A lack of homogeneity in spermatogenesis within the testis was the basis of mTESE. Large tubules with a whitish hue were associated

Table II. Investigational characteristics.

Hormonal profile	Median	IQR
Luteinizing Hormone (LH)	6.30 *(0.57-12.07 IU/L)	4.15-7.59
Follicle Stimulating Hormone (FSH)	19.07 *(0.95-11.95 IU/L)	9.35-26.05
Testosterone	11.30 *(4.94-32.01 nmol/L)	9.49-12.64
Testicular units (n = 82)	**Median**	**IQR**
Right Testicular Volume	10.00	7.42-12.0
Left Testicular Volume	11.00	9.0-12.0
Right testicular volume (cc)	**N (%)**	
< 10	15 (36.6%)	
≥ 10	26 (63.4%)	
Left testicular volume (cc)	**N (%)**	
< 10	13 (31.7%)	
≥ 10	28 (68.3%)	

*Reference values; IQR = Inter-Quartile Range.

Table III. Association between testicular volume and sperm retrieval rate in mTESE.

Testicular volume (n = 82)	Sperm retrieval rate (mTESE		p-value
	Positive n (%)	Negative n (%)	
Right Testicular Volume			0.443
< 10	11 (73.3)	4 (26.7)	
≥ 10	16 (61.5)	10 (38.5)	
Left Testicular volume			0.269
< 10	7 (53.8)	6 (46.2)	
≥ 10	20 (71.4)	8 (28.6)	

*Chi-square test.

with complete spermatogenesis as opposed to thin semi-collapsed tubules. In our study, we compared the SRR among patients who underwent mTESE following a negative TESA outcome. The overall SRR following mTESE was 65.9% and patients with higher FSH levels had a higher positive SRR (78.3%). However, this effect was not statistically significant.

Among studies that examined the SRR after salvage mTESE, only two papers[13,14] were identified and both were focused on salvage mTESE performed after different sperm retrieval techniques. Kalsi et al[13] reviewed the findings in 58 patients who underwent salvage mTESE after TESA/TESE and positive SRR was noted in 46.5%. Another retrospective study, performed by researchers from China, included 52 patients who underwent salvage mTESE after failed TESE and reported an SRR of 38.4%[14]. Our study found a relatively higher SRR (65.9%) than that reported by Kalsi et al[13] (46.5%) and Xu et al[14] (38.4%). Some investigators suggested that in patients with favorable histology such as hypospermatogenesis, proceeding with TESA is practical to avoid the added costs of directly starting with mTESE.

In one study by Yücel et al[15] reported factors that may influence the success of salvage mTESE were investigated. They retrospectively reviewed 49 patients who underwent salvage mTESE after a failed mTESE. Age, body mass index (BMI), hormonal profile, history of cryptorchidism or varicocele, and histopathology findings were reviewed. The success rate of salvage mTESE was 49.1%, and pre-operative FSH level was the only statistically significant variable[15]. Tsujimura et al[16] reviewed the findings of 46 salvage mTESE procedures after a failed cTESE. They concluded that histology and the outcome of previously used sperm retrieval techniques did not affect salvage mTESE. In contrast, findings by Ramasamy and Schlegel[17] were similar to our findings. They included 176 patients with previous negative conventional testicular sperm extraction (cTESE), of which 17 patients had undergone cTESE 3-4 times before mTESE and the remaining patients had undergone cTESE once or twice before. Both groups were compared with 135 patients who had not undergone cTESE before their mTESE. The SRR was lower in the group of patients with a history of 3-4 cTESE procedures. However, the SRR was not significantly different between cTESE-naïve patients and patients with a history of 1-2 cTESE

Table IV. Association between sperm retrieval rate in mTESE with FSH level (IU/L) and testosterone level (nmol/L).

Variable	Sperm retrieval rate (mTESE)		OR	95% CI		p-value
	Positive number (%)	Negative number (%)		Lower	Upper	
FSH level			0.278	0.072	1.077	0.058
≤ 18	9 (50.0)	9 (50.0)				
> 18.1	18 (78.3)	5 (21.7)				
Testosterone level			2.521	0.568	11.181	0.305
< 10	11 (78.6)	3 (21.4)				
≥ 10	16 (59.3)	11 (40.7)				

OR=odd ratio; CI=confidence interval, *Chi-square test.

procedures. Ramasamy and Schlegel[17] reported that FSH levels before mTESE were not significantly associated with SRR, which is similar to our findings.

A systematic review and meta-analysis by Bernie et al[12] compared sperm retrieval techniques and found that mTESE had a 17% higher SRR than cTESE, and cTESE had a 28% higher SRR than TESA. They speculated that the SRR reported for mTESE in the current literature was lower than expected because patients undergoing mTESE typically had a history of failed interventions and were not treatment naïve[12]. A head-to-head comparison between TESA and mTESE was done by El-Haggar et al[18]. One hundred patients underwent TESA on one testis and mTESE on the other one and the overall SRR was 54% by mTESE and 10% by TESA[18].

Limitations

Our study has certain limitations, such as small sample size and lack of case homogeneity. Our study showed higher SRR than that previously reported in patients who had undergone mTESE following a negative TESA outcome. However, there was no significant association between SRR and testicular volume, FSH, or testosterone level.

Conclusions

The positive SRR of mTESE performed after a negative TESA outcome was 65.9%. Testicular size and hormonal profile had no significant impact on SSR following mTESE after failed TESA. Our findings are consistent with the fact that a negative sperm aspiration result cannot be considered a negative predictor of a future mTESE outcome.

Conflict of Interest

The Authors declare that they have no conflict of interests.

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Ethics Approval

This study received IRB approval from King Abdullah International Medical Research Center (KAIMRC-Riyadh) under Project Number (RC19/195/R).

Informed Consent

Patients' confidentiality was secured and all patients signed informed consent.

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