Original Article

The relationship of testicular stiffness with Johnsen score and sperm retrieval outcome in men with non-obstructive azoospermia

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Background: The pathological Johnsen score (JS) is a quantitative histological scoring system used to assess spermatogenesis in men with nonobstructive azoospermia (NOA), while elastic modulus derived from shear wave elastography (SWE) is a diagnostic tool for evaluating spermatogenic dysfunction. In this prospective observational study, we aimed to investigate whether testicular stiffness measured by SWE could serve as a substitute for JS in predicting sperm retrieval outcomes in men with NOA.

Methods: This prospective cohort study analyzed 140 testes from 115 consecutive outpatient participants with NOA who had sought treatment at the reproductive medical center of a tertiary care hospital between January 2018 and October 2021. Testicular volume, elastic modulus, JS, and sperm retrieval outcomes were calculated. Statistical differences in parameters between the positive and negative sperm retrieval groups were determined using the Mann-Whitney test. Spearman rank correlation analysis was performed to determine the correlations between JS and either testicular volumes or elastic modulus. Receiver operating characteristic (ROC) curves were drawn to evaluate the diagnostic performance of the testicular elastic modulus and testicular volume.

Results: The JS correlated positively with testicular volume and negatively with the maximum elastic modulus (Emax) and mean elastic modulus (Emean), with correlation coefficients of 0.804, –0.686, and –0.456, respectively (P<0.01). There were significant differences in JS, testicular volume, and Emax between participants with positive and negative sperm retrieval of microdissection testicular sperm extraction (micro-TESE) (P<0.01). ROC curves were plotted for JS, testicular volume, and Emax to distinguish between participants with positive and negative sperm retrieval. The areas under the ROC curves (AUCs) were 0.783 [95% confidence interval (CI): 0.707–0.859; P<0.01], 0.737 (95% CI: 0.651–0.823; P<0.01), and 0.729 (95% CI: 0.643–0.814; P<0.01), respectively. When the cutoff value of JS was 4.5, its sensitivity and specificity were 60.3% and 89.6%, respectively. When the cutoff value of Emax was 3.75 kPa, its sensitivity and specificity were 79.1% and 64.4%, respectively. The sensitivity and specificity were 68.5% and 83.6%, respectively when the cutoff value of testicular volume was 8.17 mL. Emax combined with testicular volume improved this diagnostic value, with an AUC of 0.742 (95% CI: 0.657–0.828; P<0.01), and sensitivity and specificity were 83.6% and 68.5%, respectively.

Conclusions: Our study suggests that the combination of testicular stiffness and volume measurements may serve as a viable alternative to pathological JS in predicting the likelihood of successful sperm retrieval prior to micro-TESE procedures.

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Introduction

Azoospermia is present in approximately 5–10% of male cases of infertility, with nonobstructive azoospermia (NOA) accounting for 60% of these cases (1,2). Microdissection testicular sperm extraction (micro-TESE) is the preferred technique for testicular sperm retrieval (3), but its success rates for patients with NOA range from 39.1% to 46.8% due to severe spermatogenic dysfunction in the testes (4). As a result, patients with NOA considering micro-TESE typically want to know the likelihood of success beforehand, given the emotional and financial implications of a failed procedure (5). Histopathological examination of testicular tissue using a quantitative histological scoring system, such as the Johnsen score (JS), is commonly used to assess spermatogenesis and predict sperm retrieval rates (SRRs) before micro-TESE. The JS is based on grading multiple tubules from 10 to 1 according to the most predominant histological pattern observed in the biopsy, with a higher score indicating better spermatogenic status and a lower score indicating more severe dysfunction (6).

Although testicular biopsy has been a crucial procedure for many years in diagnosing and managing NOA, the benefits of performing a diagnostic biopsy before micro-TESE are questionable. This is because the diagnostic biopsy itself is an invasive procedure that can lead to many complications, including pain, infection, intratesticular bleeding, scrotal hematoma, intratesticular scar tissue formation, postoperative hypogonadism, and removal of focal areas of spermatogenesis (7). Patients with NOA typically have heterogeneous seminiferous tubules, meaning that the absence of sperm in a single biopsy does not necessarily indicate the absence of sperm in the entire testis (8). Moreover, a diagnostic testicular biopsy can remove focal areas of sperm production, which can ultimately affect the chances of successful sperm retrieval in the future. Considering all these factors, a noninvasive diagnostic method that can help adjust treatment programs, improve surgical success rates, and avoid unnecessary surgical interventions would be of particular value.

Shear wave elastography (SWE) is a widely used noninvasive method for assessing tissue stiffness and is an effective complement to conventional ultrasonography. Testicular elastic modulus, which is associated with sperm concentration, has been found to have a high diagnostic value for evaluating severe spermatogenic dysfunction (9,10). However, there have been few reports on the relationship between testicular stiffness and pathological findings in patients with NOA. Hu *et al.* (11) conducted a study that divided infertile males into four groups based on the results of testicular biopsies, including normal testicular spermatogenesis, hypospermatogenesis, spermatogenesis arrest, and Sertoli cell-only syndrome. Significant differences in SWE values were observed between these four groups ($P<0.01$). Similarly, Abdelaal *et al.* (12) examined patients with NOA undergoing testicular sperm extraction and found that there was a significant difference in SWE values between those with positive sperm retrieval and those with negative sperm retrieval, with a sensitivity of 75.0% and a specificity of 85.7% ($P<0.01$). However, these studies are limited by their small number of positive cases, and the results may not be generalizable to the wider population. Greater attention and additional research with refined study designs and larger sample sizes should be conducted to confirm these findings and evaluate the clinical utility of SWE in diagnosing and managing NOA.

The purpose of this study was to evaluate the correlation between testicular stiffness and JS and to determine whether SWE can serve as a substitute for testicular biopsy in predicting the efficacy of micro-TESE. We present this article in accordance with the STROBE reporting checklist (available at https://qims.amegroups.com/article/view/10.21037/qims-23-1381/rc).

Methods

Participant screening and enrollment

The prospective cohort study was conducted in accordance with the Declaration of Helsinki (as revised in 2013) and was approved by the Ethics Committee of Shengjing Hospital of China Medical University (No. 2018PS104J). Informed consent was obtained from all participants. Initially, we recruited 331 consecutive patients with

NOA who had visited the reproductive medical center of Shengjing Hospital of China Medical University from January 2018 to October 2021. To minimize selection bias, strict inclusion and exclusion criteria were applied. The inclusion criteria were the following: (I) a diagnosis of NOA according to the updated guidelines (13); (II) completion of testicular SWE examination, with the image exhibiting high clarity and the elastic modulus demonstrating consistent stability; and (III) micro-TESE performed within 7 days after the ultrasound examination, during which a part of testicular tissue was excised for pathological examination.

The exclusion criteria were as follows: (I) a history of testicular trauma; (II) testicular biopsy within the previous 3 months; (III) abnormal ultrasound findings such as testicular masses, extensive microlithiasis, hydrocele, cryptorchidism, and inguinal hernia; (IV) a biopsy sample insufficient in size to evaluate the spermatogenetic status of the testis; (V) a history of mumps orchitis; and (VI) a history of previous chemotherapy or radiotherapy.

Grayscale ultrasound and SWE examinations

Grayscale ultrasound and SWE measures were performed using an Aixplorer ultrasound diagnostic imaging system (SuperSonic Imagine, Aix en Provence, France), with a linear transducer operating at a frequency of 4–15 MHz. All examinations were conducted by an experienced radiographer with 5 years of testicular SWE experience.

Participants were examined in the supine position, and grayscale ultrasound of the scrotal contents was performed to assess the placement, contour, size, and parenchymal echoes of the testes. Testicular length, width, and height were measured, and testicular volume was calculated using the following formula: testicular volume (mL) = (length × width × height) × 0.71 (14).

Following the grayscale ultrasound assessment, SWE measurements were taken in the maximum longitudinal plane. Grayscale and SWE images were displayed simultaneously on the monitor, with the elastic modulus displayed as an SWE map in kilopascals (range, 0–180 kPa). The central part of the testis exhibits a more consistent stiffness, thereby providing a more accurate representation of the overall testicular stiffness (15), and a 10-mm region of interest (ROI) in the middle of testis was selected for three consecutive measurements of the maximum elastic modulus (Emax), mean elastic modulus (Emean), and minimum elastic modulus (Emin) (*Figure 1*). The average values were recorded as the testicular elastic modulus. If the testicular volume was small, the size of the ROI was adapted accordingly.

Micro-TESE and testicular pathological analysis

All participants underwent micro-TESE after ultrasound examination, from which partial testicular tissue was removed for biopsy (*Figure 1*). The testicular samples were then immersed in Bouin's solution and sent to the pathology laboratory for analysis. The biopsy samples were stained with hematoxylin and eosin and examined under light microscopy. Histopathological analysis of the testicular samples was performed by a single pathologist who was an expert in andrology. The pathologist was blinded to the clinical data and SWE results. Based on the level of spermatogenesis, a JS from 1 to 10 was assigned to each testicular biopsy specimen. Sperm retrieval outcomes were also recorded.

Sample size

The sample size was determined using PASS 15.0 software (Number Cruncher Statistical Software, LLC, Kaysville, UT, USA). Based on previous literature (12) and our preliminary experiment, the area under the receiver operating characteristic (ROC) curves for SWE and JS in distinguishing successful from negative sperm retrieval were found to be 0.869 and 0.750, respectively. We employed tests for two ROC curves to estimate the required sample size. With a power of 90% and an alpha level of 5% (two-sided), the minimum sample size per group was calculated as 65. With a dropout rate of 40%, the total minimum sample size needed was determined to be 182 participants. During our study, we unexpectedly encountered a higher number of patients who did not undergo micro-TESE and testicular biopsy than anticipated; therefore, we included additional patients.

Statistical analysis

Statistical analysis was performed using SPSS 25.0 (IBM Corp., Armonk, NY, USA). The normality of the data and the homogeneity of the variables were examined according to the Kolmogorov-Smirnov test. The data did not satisfy the normal distribution and are presented as medians with 25^{th} and 75^{th} percentiles. Statistical differences in parameters between the positive and negative sperm retrieval groups were determined using the Mann-Whitney

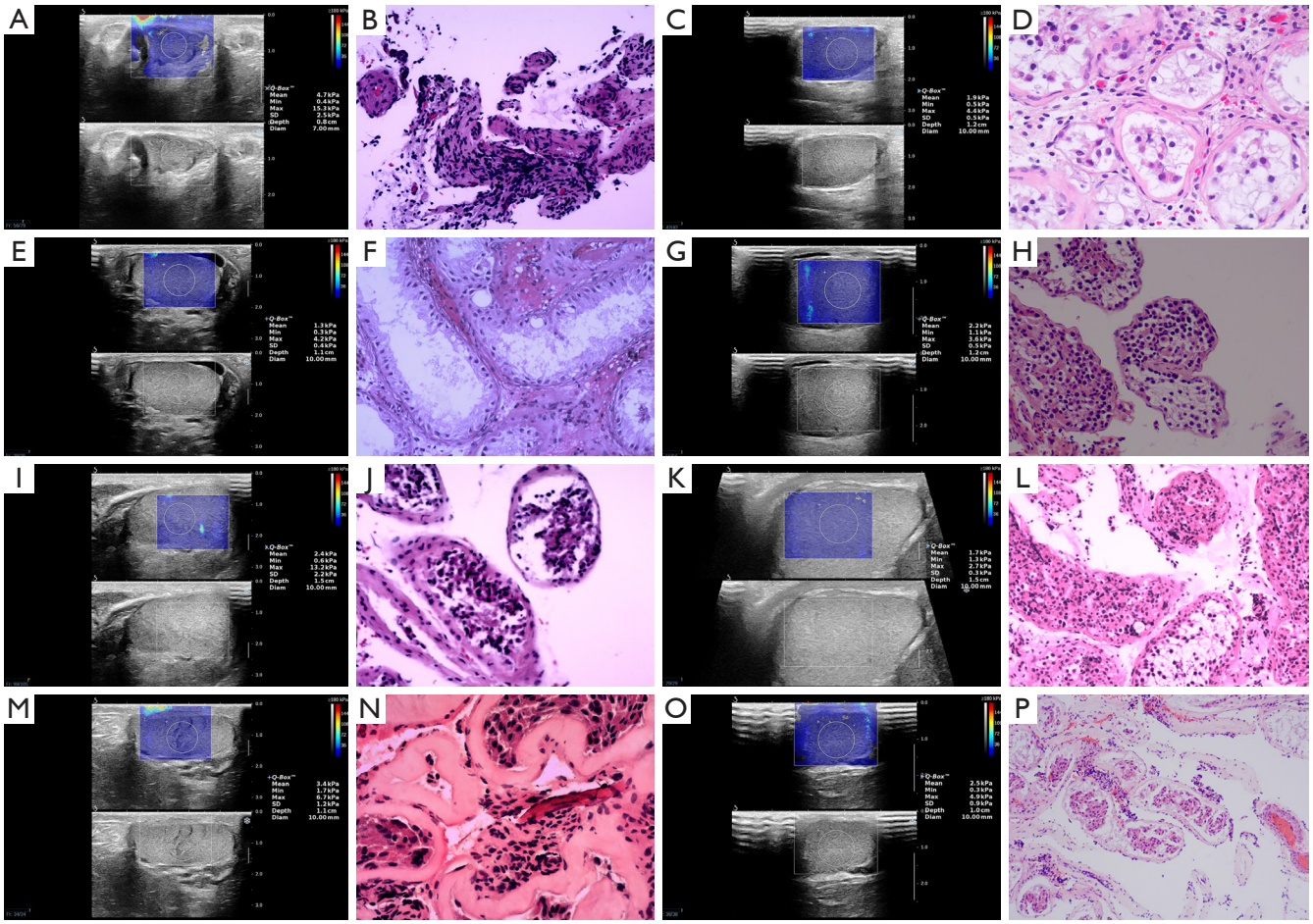

Figure 1 The testicular SWE maps and pathological sections of patients with NOA. (A,B) A 38-year-old man: 47,XXY; testicular volume, 0.94 mL; Emean, 4.7 kPa; Emin, 0.4 kPa; Emax, 15.3 kPa; and JS, 1. (C,D) A 33-year-old man: 45,XY,der(13;14)(q10;q10); testicular volume, 5 mL; Emean, 1.9 kPa; Emin, 0.5 kPa; Emax, 4.4 kPa; and JS, 3. (E,F) A 52-year-old man: Y chromosome AZF regions sY152, sY254, sY255 microdeletions; testicular volume, 10.24 mL; Emean, 1.3 kPa; Emin, 0.3 kPa; Emax, 4.2 kPa; and JS, 2. (G,H) A 27-year-old man: 46,XY,t(20;21)(p11.2;q11.2); testicular volume, 8.59 mL; Emean, 2.2 kPa; Emin, 1.1 kPa; Emax, 3.6 kPa; and JS, 5. (I,J) A 29-year-old man: 46,XY(9qh+); testicular volume, 11.87 mL; Emean, 2.4 kPa; Emin, 0.6 kPa; Emax, 13.2 kPa; and JS, 6. (K,L) A 33-year-old man: 46,XY(1qh+); testicular volume, 15.39 mL; Emean, 1.7 kPa; Emin, 1.3 kPa; Emax, 2.7 kPa; and JS, 8. (M,N) A 30-year-old man: 46,XY; testicular volume, 7.93 mL; Emean, 3.4 kPa; Emin, 1.7 kPa; Emax, 6.7 kPa; and JS, 2. (O,P) A 30-year-old man: testicular volume, 4.45 mL; Emean, 2.5 kPa; Emin, 0.3 kPa; Emax, 4.9 kPa; and JS, 2. (B,D,F,H,J,L,N,P) Samples stained with hematoxylin and eosin and observed by under microscopy (magnification ×100). SWE, shear wave elastography; NOA, nonobstructive azoospermia; Emean, mean elastic modulus; Emin, minimum elastic modulus; Emax, maximum elastic modulus; JS, Johnsen score; AZF, azoospermia factor.

test. Since JS is a rank variable, Spearman rank correlation analysis was performed to examine the correlations between JS and either testicular volumes or elastic modulus. ROC curves were drawn to evaluate the diagnostic performance of the testicular elastic modulus and testicular volume. The areas under the ROC curves (AUCs), sensitivities, and specificities were calculated, and the cutoff values were determined using the Youden indexes. A P value of <0.05 was considered statistically significant.

Results

Participant screening and enrollment

The process of participant screening and enrollment

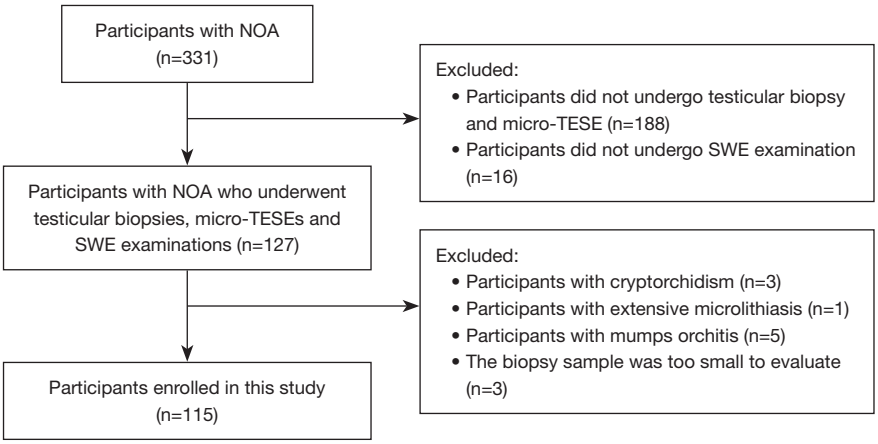

Figure 2 Participant screening and enrollment. NOA, nonobstructive azoospermia; micro-TESE, microdissection testicular sperm extraction; SWE, shear wave elastography.

is shown in *Figure 2*. As it has been reported that the histological features of the testes can vary between the left and right sides (16), we included all pathology results from testicular biopsies in our study. In total, 140 testes from 115 participants were enrolled.

Descriptive statistics

Descriptive statistics of age, testicular volume, elastic modulus, number participants with successful sperm retrieval, autosomal abnormalities, and Y-chromosome azoospermic factor region microdeletions for each participant at different JS levels are presented in *Table 1*. We observed an increase in testicular volume with increasing JS, while Emax and Emean showed a decrease (*Figure 3*). No significant changes in Emin or age were observed in our study (*Figure 3*).

The association of JS with testicular elastic modulus and testicular volume

The JS was positively correlated with testicular volume but negatively correlated with Emax and Emean, with correlation coefficients of 0.804, −0.686, and −0.456, respectively (P<0.01). The JS was positively correlated with Emin, with the correlation coefficient of 0.182 (P<0.05).

ROC curves of JS, Emax, and testicular volume (Figure 3)

Out of the 140 testes analyzed, for participants with positive sperm retrieval (73 testes, 52.1%), the median (25^{th}, 75^{th} percentile) values of JS, testicular volume, Emax, and Emean were 8 [2, 8], 11.35 (5.53, 14.16) mL, 3.10 (2.55, 5.40) kPa, and 1.90 (1.60, 2.45) kPa, respectively. For participants with negative sperm retrieval (67 testes, 47.9%), the median values of JS, testicular volume, Emax, and Emean were 2 [2, 3], 5.75 (4.32, 7.87) mL, 5.60 (4.00, 7.90) kPa, and 2.20 (1.70, 3.30) kPa, respectively. Significant differences in JS, testicular volume, and Emax were observed between participants with positive and negative sperm retrieval (P<0.01).

We constructed ROC curves to evaluate the diagnostic value of JS, testicular volume, and Emax in distinguishing between participants with positive and negative sperm retrieval. The AUCs were 0.783 [95% confidence interval (CI): 0.707–0.859; P<0.01], 0.737 (95% CI: 0.651–0.823; P<0.01), and 0.729 (95% CI: 0.643–0.814; P<0.01), respectively. The optimal cutoff values were determined using the maximum Youden index. The optimal cutoff value for JS was 4.5, with a sensitivity of 60.3% and a specificity of 89.6%; for Emax, the optimal cutoff value was 3.75 kPa, with a sensitivity of 79.1% and specificity of 64.4%; and for testicular volume, the optimal cutoff value was 8.17 mL, with a sensitivity of 68.5% and specificity of 83.6%. Combining Emax and testicular volume improved the diagnostic value, with an AUC of 0.742 (95% CI: 0.657–0.828; P<0.01) and a sensitivity and specificity of 83.6% and of 68.5%, respectively.

Seminiferous tubule hyalinization

Among the participants with JS 1–2, 20 (20/71) had

Table 1 Descriptive statistics of participants with a Johnsen score of 1–9

Johnsen score	Patients, N	Testicular volume, mL[†]	Emax, kPa[†]	Emean, kPa[†]	Emin, kPa[†]	Age, years[†]	Sperm retrieval success (%, n/N)	Autosomal abnormality and chromosome polymorphic variants	Y chromosome AZF region microdeletions
1	18	3.92 (1.76, 5.30)	8.45 (6.43, 14.65)	3.00 (2.48, 3.90)	0.75 (0.58, 1.23)	30.00 (29.25, 31.00)	22.22 (4/18)	47,XXY (n=4)	–
2	53	5.67 (3.37, 7.27)	5.20 (3.90, 7.00)	2.20 (1.65, 2.75)	1.00 (0.80, 1.40)	30.00 (27.00, 35.50)	33.96 (18/53)	47,XXY (n=6)	Microdeletions of sY152, sY254, sY255
3	9	5.00 (4.90, 9.29)	5.00 (3.90, 5.70)	2.00 (1.80, 2.60)	0.90 (0.60, 1.45)	30.00 (28.50, 32.00)	44.44 (4/9)	45,XY,der(13;14)(q10;q10)	–
4	9	9.46 (5.85, 11.35)	6.50 (4.85, 8.25)	2.50 (1.50, 4.10)	0.80 (0.35, 1.65)	28.00 (26.00, 33.00)	33.33 (3/9)	46,XY,(21ps+) (n=2)	–
5	4	9.74 (6.46, 11.76)	3.65 (3.23, 4.53)	1.95 (1.63, 2.28)	1.00 (0.75, 1.40)	27.50 (23.25, 28.75)	75 (3/4)	46,XY,t(13;14)(p12;q21); 46,XY,t(20;21)(p11.2;q11.2)	–
6	4	13.09 (7.35, 22.37)	4.90 (2.33, 9.88)	2.05 (1.60, 3.33)	1.20 (0.73, 1.45)	29.00 (26.00, 32.00)	50 (2/4)	46,XY(9qh+)	–
7	3	11.48 (N/A, N/A)[‡]	4.50	2.10	1.50	27.00	66.67 (2/3)	–	Microdeletions of sY152, sY157, sY239, sY242, sY254, sY255 (n=2)
8	39	13.96 (12.09, 17.35)	2.60 (2.50, 3.10)	1.70 (1.50, 1.90)	1.10 (0.90, 1.50)	30.00 (27.00, 34.00)	92.31 (36/39)	46,XY(1qh+)	–
9	1	9.89 (N/A, N/A)[‡]	2.40	1.4	0.8	28	100 (1/1)	–	–
Total	140	7.29 (4.77, 12.09)	4.55 (2.83, 6.88)	2.05 (1.60, 2.70)	1.05 (0.80, 1.40)	30.00 (27.00, 33.00)	52.14 (73/140)	–	–

[†], values are presented as the median (25th, 75th percentile); [‡], for groups with fewer participants, only the median is provided due to insufficient data for interquartile range calculation. Emax, maximum elastic modulus; Emean, mean elastic modulus; Emin, minimum elastic modulus; AZF, azoospermic factor; N/A, not applicable.

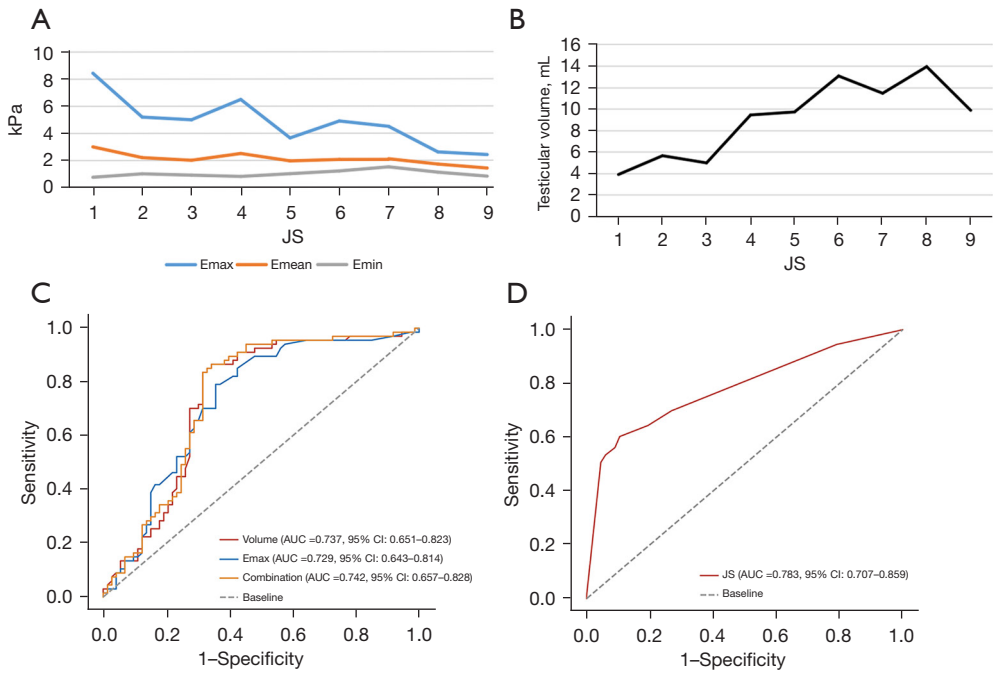

Figure 3 Results of elastic modulus and testicular volume and ROC curves. (A) Emax, Emean and Emin values with increasing JS. (B) Testicular volume with increasing JS. (C) The ROC curves for Emax, testicular volume, and the combined value in differentiating between participants with positive and negative sperm retrieval. (D) ROC curve for JS in differentiating between participants with positive and negative sperm retrieval. JS, Johnsen score; Emax, maximum elastic modulus; Emean, mean elastic modulus; Emin, minimum elastic modulus; AUC, area under the curve; CI, confidence interval; ROC, receiver operating characteristic.

seminiferous tubule hyalinization, with 10 cases of Klinefelter syndrome (KS) and 10 cases without KS. For those with KS, the median (25^{th}, 75^{th} percentile) values of Emax, Emean, and testicular volume were 9.15 (7.13, 13.43) kPa, 4.60 (3.50, 5.28) kPa, and 1.41 (1.03, 1.49) mL, respectively. For those without KS but with seminiferous tubule hyalinization, the median (25^{th}, 75^{th} percentile) values of Emax, Emean, and testicular volume were 8.75 (6.18, 17.25) kPa, 2.90 (2.10, 3.83) kPa, and 5.38 (4.09, 6.41) mL, respectively.

Mumps orchitis

We excluded five participants with a confirmed history of mumps orchitis due to the extremely high testicular stiffness. The median (25^{th}, 75^{th} percentile) values for these participants were 10.70 (9.15, 21.95) kPa for Emax, 3.60 (2.05, 4.20) kPa for Emean, 7.16 (6.17, 14.51) mL for testicular volume, and 8 [5, 8] for JS. All of these participants had successful sperm retrieval.

Varicocele

In our study, 15 participants were diagnosed with varicocele according to scrotal color Doppler ultrasound. The median (25^{th}, 75^{th} percentile) values for Emax, Emean, testicular volume, and JS were 6.90 (2.40, 13.00) kPa, 2.60 (1.30, 6.70) kPa, 6.48 (1.48, 15.37) mL, and 2 [2, 8], respectively.

Discussion

In recent years, there has been significant interest in developing an effective means to evaluating patients with NOA prior to micro-TESE. Our study discovered a strong correlation between testicular stiffness and pathological findings in patients with NOA. Moreover, we found that combining testicular stiffness with testicular volume led to a highly effective method for identifying patients with NOA who are likely to have positive sperm retrieval during micro-TESE.

Numerous studies have explored the connection

between histopathology and SRR in patients with NOA. Histopathology is a potent predictor of the success of micro-TESE in these patients (17). One quantitative histopathological method is the JS, which independently predicts sperm retrieval. Studies have shown that the presence of mature spermatozoa in histopathology specimens, regardless of overall spermatogenesis, strongly predicts successful SRR, with a JS score of ≥8 being the most predictive (5,16). For example, Rohan *et al.* (18) reported that the mean JS was 7.8 in men with successful micro-TESE compared to 2.84 in those with unsuccessful sperm retrieval. When the optimal JS cutoff value was set to 5, the SRR was 6% for scores <5 and 88% for scores ≥5, which is in line with our results. Although histopathological findings offer a relatively higher accurate prediction of SRR, performing a diagnostic biopsy before micro-TESE is akin to conducting a surgical procedure twice, which patients are reluctant to accept.

Fibrosis, characterized by thickening of the seminiferous tubule wall, is a hallmark of male infertility and a common feature of impaired spermatogenic function (19). The resulting increased stiffness can be detected by SWE, which evaluates the degree of testicular fibrosis and parenchymal damage (20). Many studies have shown that severe oligozoospermia and NOA are associated with increased testicular SWE-assessed stiffness (9,10). In our study, we more deeply examined the relationship between SWE and NOA, discovering that testicular stiffness increased as JS decreased, suggesting a strong correlation between stiffness and the parenchymal damage associated with spermatogenic function. These results suggest that SWE has the potential to replace JS as a predictor of SRR, given the high level of agreement between the two measures.

Testicular volume is a valuable clinical tool for evaluating the severity of male infertility (9), as it has a high predictive value for spermatogenesis, especially in patients with NOA (21). However, testicular volumes in these patients are typically smaller except in cases of early or late maturation arrest, which aligns with our study's findings (22). Our research revealed a significant positive correlation between testicular volume and JS, with smaller testes being associated with JS scores of 1–3. Although testicular volume has been linked to the success rate of micro-TESE in some studies (23), others have reached conflicting conclusions. For instance, one study found that SRRs did not differ significantly among men with testicular volumes of ≤2, >2–10, and >10 mL (24). Moreover, a recent meta-analysis of 1,764 cases found no threshold of testicular volume associated with SRR (25). Additionally, testicular volume varies widely due to numerous factors, including race (26). Although testicular volume is a useful diagnostic tool for evaluating spermatogenic function in patients with NOA, its role as an independent predictor of successful sperm retrieval remains controversial.

Our study found that combining Emax and testicular volume was similarly effective to using JS in predicting successful sperm retrieval in participants with NOA. Our results suggest that testicular elastic modulus combined with volume could serve as a noninvasive alternative to testicular biopsy for predicting SRR before micro-TESE.

The etiology of NOA is a crucial factor in predicting successful sperm retrieval. Our study found that quantitative SWE features demonstrated good diagnostic performance in NOA patients with mumps orchitis and KS. Additionally, we found that patients with NOA caused by mumps orchitis exhibited characteristic SWE features of ultrahigh testicular stiffness, with median Emax and Emean values of 10.70 and 3.60 kPa, respectively. These values were considerably higher than those observed in the NOA participants enrolled in our study, who had median Emax and Emean values of 4.55 and 2.05 kPa, respectively. These findings are consistent with previous reports indicating that patients with orchitis have the highest and most effective SRR of 100%, while that of other patients is an average 46.0% (27). Our study also showed that the median JS of patients with NOA and mumps orchitis was 8, and the SRR was 100%. However, due to the limited number of cases in this study, further research with a larger sample size is necessary to validate these findings.

KS (karyotype 47, XXY) remains the most common chromosomal disorder that causes NOA. The testes of individuals with KS are characterized by extensive fibrosis and hyalinization of the seminiferous tubules, as well as hyperplasia of the interstitium (28). The increased stiffness of KS testes is due to seminiferous tubule hyalinization. However, in our study, we observed obvious seminiferous tubule hyalinization in the testes of 10 participants without KS who had a JS of 1–2, which is consistent with previous research (29). Seminiferous tubule hyalinization has been reported not only in the testes of those with KS, but also in the undescended testes and testes of patients with severe oligozoospermia or NOA (30). Moreover, in our study, participants without KS but with significant seminiferous tubule hyalinization had testicular stiffness similar to that of KS participants but larger testicular volumes. Therefore, a combination of testicular stiffness and volume showed high

diagnostic efficacy in distinguishing KS from other causes of NOA.

We did not exclude participants with varicocele from the analysis for several reasons. First, among participants with the same JS, we observed no differences in testicular volume or elastic modulus between those with varicocele and those without. This suggests that varicocele did not significantly affect testicular pathology in our study population. Second, to minimize the potential impact of varicocele on testicular biopsy results, we only performed biopsies on the contralateral testes. Therefore, we believe that varicocele did not have a significant effect on our study findings.

Our study had several limitations that need to be considered. First, even though participants with NOA had significant impairments in their sperm production, it is important to note that there could be regions within the testicular tissue that still exhibit active spermatogenesis. Therefore, a single testicular biopsy may not accurately reflect the overall spermatogenic function of the entire testis. Second, since all the ultrasound and SWE examinations were performed by a single operator, we could not assess interobserver variability.

Conclusions

Our study suggests that the combination of testicular stiffness and volume measurements may serve as a viable alternative to the pathological JS in predicting the likelihood of successful sperm retrieval prior to micro-TESE procedures.

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Footnote

Reporting Checklist: The authors have completed the STROBE reporting checklist. Available at https://qims.amegroups.com/article/view/10.21037/qims-23-1381/rc

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at https://qims.amegroups.com/article/view/10.21037/qims-23-1381/coif). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. This study was conducted in accordance with the Declaration of Helsinki (as revised in 2013) and was approved by the Ethics Committee of Shengjing Hospital of China Medical University (No. 2018PS104J). Written informed consent was provided by all participants.

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