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ORIGINAL ARTICLE

Male Fertility

The impact of ipsilateral testicular atrophy on semen quality and sperm DNA fragmentation response to varicocele repair

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Varicoceles adversely impact semen quality and sperm DNA fragmentation, which typically improve with surgical repair. Some men with varicoceles have ipsilateral testicular atrophy due to damage from the varicocele. This study assessed semen quality and the sperm DNA fragmentation index (DFI) response to varicocele repair in men with ipsilateral testicular atrophy (TA) versus men with no testicular atrophy (NTA). Semen parameter values and DFI in both groups were compared preoperatively and postoperatively. The Mann–Whitney U test and the Wilcoxon signed-rank test were used where appropriate. There were 20 men in the TA group and 121 men in the NTA group with no difference in age, varicocele grade, or preoperative semen parameter values between the two groups. The NTA group had a higher preoperative DFI than the TA group. Both groups showed improvement in semen quality postoperatively, only the TA group showed a significant improvement in DFI, whereas the NTA group showed significant improvements in several parameter values and DFI. The change from preoperative to postoperative parameter values when comparing the two groups revealed a difference in total sperm motile count and DFI, with a larger mean improvement in the NTA group than in the TA group. Both TA and NTA groups showed improved semen quality and DFI after varicocele repair, but the NTA group had more improvement than the TA group. However, only total motile count (TMC) and DFI had a significantly greater mean change in preoperative to postoperative response in the NTA group than in the TA group.

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INTRODUCTION

Approximately 15% of couples in the United States are considered infertile, after being unsuccessful at achieving a pregnancy for 1 year with unprotected intercourse. Of the couples struggling to conceive, a male factor is solely responsible for 20% of these cases, while it is a contributory factor in conjunction with female infertility factors in an additional 40%, indicating that in 60% of cases where there is difficulty conceiving, there is male factor involvement.[1] Varicoceles are abnormally dilated scrotal veins, which are found in approximately 15% of men in the general population and in 40% of men presenting for infertility evaluations, making varicoceles the most common diagnosis made in infertile men.[2] Varicoceles adversely affect the function of the testicle and spermatogenesis.[3–9]

On physical examination, the testicular size or volume correlates with the level of spermatogenesis and is an indicator of a man's fertility.[10,11] A Prader orchidometer is a string of ellipsoids of increasing size with their volumes marked on each to assist in the assessment of testicular size on physical examination.[12] A low testicular volume has been traditionally defined as a volume of 20 ml or less.[13]

Varicoceles are known to be able to induce enough testicular damage in some men to result in testicular atrophy.[14–17] Varicocele repair results in significant improvement in semen parameter values and pregnancy rates.[18–20] Although studies have correlated the response of semen quality to varicocele repair with the size or grade of the varicocele,[21] the goal of this current study was to assess the response to varicocele repair in men with testicular atrophy (TA) in the ipsilateral testicle in comparison with men with no testicular atrophy (NTA). These data were sought in hopes of helping to counsel men with varicoceles and ipsilateral TA on expectations after varicocele repair.

PATIENTS AND METHODS

Men who underwent varicocele repair for infertility with palpable varicoceles after their clinical grading were categorized into two groups, those with TA in the ipsilateral testicle and those with NTA. Clinical grading of varicoceles is categorized as Grade 1: palpable only on Valsalva; Grade 2: readily palpable with dilation on Valsalva; and Grade 3: apparent on visual inspection. Testicular volumes were measured on physical examination with the assistance of a Prader orchidometer by a single reproductive urologist who was also the operating surgeon. Scrotal ultrasound was not obtained for varicocele diagnoses as this is not standard of care, nor is it advocated by the American Urological Association best practice statement on optimal evaluation of the infertile male.[22] To be included in the TA group, the

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ipsilateral testicle to the varicocele had to measure less than 18 ml and have a volume of at least 3 ml less than the contralateral testicle. The mean testicular volume in the NTA group was 20 ml. Semen parameters and sperm DNA fragmentation index (DFI) in both groups were compared preoperatively and 3 months postoperatively and the data were collected by review of the electronic health record after obtaining St. David's Healthcare Institutional Review Board exemption from full review (Austin, TX, USA) due to the de-identified nature of the data. As this was performed as a retrospective health record review with a de-identified database, it was not necessary to obtain consent from patients who had their data included. Sperm morphology was not included owing to lack of standardization of criteria in different andrology laboratories, with some laboratories using Kruger strict morphology criteria versus others using WHO 4th edition criteria.[23] Depending on the laboratory, either WHO 4th edition or WHO 5th edition criteria were applied as references for the other semen parameters.[24] Sperm DFI assessment was performed by sperm chromatin dispersion assay using Spectrum Technology's second-generation Halosperm kit (Madrid, Spain).

Between October 2010 and January 2019, 359 varicocele repairs were performed by a single microsurgeon after evaluation in a private fertility center (Austin Fertility and Reproductive Medicine/Westlake IVF, Austin, TX, USA), and 141 varicocele repairs met the inclusion criteria. Excluded were men who underwent bilateral varicocele repair, with bilateral testicular atrophy, who had testicular atrophy and a varicocele with a history of cryptorchidism, who had undergone varicocele repair for hypogonadism for orchialgia and not for fertility, and who did not obtain a 3-month postoperative semen analysis because they achieved a pregnancy before then or who did not follow up. All varicoceles were repaired by a subinguinal microsurgical technique. The Mann–Whitney U test and the Wilcoxon signed-rank test were used where appropriate, with a $P < 0.05$ considered statistically significant. Results were expressed as mean ± standard deviation.

RESULTS

Of the 141 men in the study, 20 were in the TA group and 121 in the NTA group. There was no significant difference in mean age between the two groups (**Table 1**). The grades of varicocele were similar in both groups: TA group had 10.0% Grade 1, 55.0% Grade 2, and 35.0% Grade 3, while the NTA group had 6.7% Grade 1, 58.7% Grade 2, and 34.7% Grade 3. There was no difference in preoperative semen parameter values between the two groups, including semen volume, sperm concentration, motility, forward progressive motility (FP), and total motile count (TMC). The NTA group had a higher preoperative DFI than the TA group, but the difference did not reach statistical significance (**Table 1**). Although both groups revealed an improvement in semen quality postoperatively, the TA group only showed a statistically significant improvement in DFI, whereas the NTA group showed significant improvements in sperm concentration, motility, FP, TMC, and DFI (**Table 2**). The mean amount of change in preoperative to postoperative parameters when comparing groups only revealed a significant difference in TMC and DFI, with a larger mean improvement in the NTA group than in the TA group (**Table 3**).

DISCUSSION

Varicoceles have an adverse impact on testicular function and spermatogenesis through mechanisms of cellular damage that have been proposed to include sperm DNA fragmentation, apoptosis, increased reactive oxygen species through oxidative stress, intracellular ionic and metabolic changes, and predisposition to sperm aneuploidy.[3–9] There are a number of hypotheses of the mechanisms by which varicoceles adversely affect spermatogenesis and testicular function. The majority of the data indicate that varicoceles impair testicular function by increasing intratesticular temperatures owing to interruption of the counter-current heat exchange in the pampiniform plexus with opposing blood flows in a central arterial and venous system.[25–27] A rat model supports the hypothesis of retrograde flow of adrenal and renal metabolites worsening varicocele induced testicular damage.[28] In some men, the testicular damage is sufficient to result in testicular atrophy. It has been well established that men with varicoceles have significantly higher levels of sperm DNA fragmentation owing to increased oxidative stress in semen.[3,4,6,8,29] It has also been shown that repair of varicoceles reduces the amount of sperm DNA fragmentation.[30–32]

Varicocele repair results in significant improvement in semen quality in 60%–80% of men, with reported pregnancy rates after varicocele repair ranging from 20% to 60%.[18] A randomized controlled trial revealed a pregnancy rate of 44% 1 year following varicocele repair compared with a pregnancy rate of 10% at the same time interval when varicoceles were left intact.[19] Another randomized controlled trial revealed that men who underwent varicocele repair had significant improvement in semen quality and the spontaneous pregnancy rate in couples within 1 year following varicocele repair was 32.9% in comparison with a spontaneous pregnancy rate of 13.9% when varicoceles were left intact.[33] Another study assessing 1500 varicocele repairs revealed that 43% of couples achieved pregnancy 1 year postrepair, and 69% of couples achieved a pregnancy 2 years following varicocele repair when couples with female factor for infertility were excluded.[20]

The goal of our current study was to assess whether men with unilateral varicoceles with ipsilateral testicular atrophy responded to varicocele repair as well as men with varicoceles without ipsilateral testicular atrophy, by means of assessing semen quality and sperm

Table 1: The preoperative versus postoperative characteristics, semen analysis, and sperm DNA fragmentation index changes in the testicular atrophy and no testicular atrophy groups, expressed in mean±standard deviation

	Preoperative TA	*Preoperative NTA*	*P*	*Postoperative TA*	*Postoperative NTA*	*P*
Age (year)	34.3±6.5	34.1±5.8	0.90			
Varicocele grade	2±0.6	2±0.6	0.89			
Semen volume (ml)	2.9±0.9	3.3±1.5	0.41	3.0±1.3	3.5±0.4	0.16
Sperm concentration ($\times 10^6$ ml^{-1})	25.5±32.4	25.7±29.7	0.95	36.0±37.0	37.5±13.7	0.70
Total motility (%)	47.5±20.3	46.9±20.6	0.97	53.4±14.5	55.4±11.6	0.55
Forward progression (%)	25.8±3.8	16.7±20.5	0.12	30.6±14.3	22.1±3.6	0.85
Total motile count (10^6)	41.2±63.8	39.1±54.1	0.89	54.8±80.0	77.8±32.3	0.24
Sperm DFI (%)	29.7±5.0	35.3±11.6	0.16	22±0	19.6±5.3	0.38

Statistical analysis performed by Mann–Whitney U test. DFI: DNA fragmentation index; TA: testicular atrophy; NTA: no testicular atrophy

Table 2: The preoperative versus postoperative semen analysis and sperm DNA fragmentation index changes in the testicular atrophy and no testicular atrophy groups, expressed in mean±standard deviation. Statistical analysis was performed by the Wilcoxon signed-rank test

	Preoperative TA	*Postoperative TA*	*P*	*Preoperative NTA*	*Postoperative NTA*	*P*
Semen volume (ml)	2.9±0.9	3.0±1.3	0.84	3.3±1.5	3.5±0.4	0.07
Semen concentration ($\times 10^6$ ml^{-1})	25.5±32.4	36.0±37.0	0.25	25.7±29.7	37.5±13.7	0.0008
Total motility (%)	47.5±20.3	53.4±14.5	0.29	46.9±20.6	55.4±11.6	0.0007
Forward progression (%)	25.8±3.8	30.6±14.3	0.38	16.7±20.5	22.1±3.6	0.0041
Total motile count (10^6)	41.2±63.8	54.8±80.0	0.13	39.1±54.1	77.8±32.3	0.00001
Sperm DFI (%)	29.7±5.0	22±0	0.0001	35.3±11.6	19.6±5.3	0.0001

DFI: DNA fragmentation index; TA: testicular atrophy; NTA: no testicular atrophy

Table 3: The amount of change in semen parameters and sperm DNA fragmentation index expressed in mean±standard deviation from prior to and after varicocele repair in men with testicular atrophy and no testicular atrophy

	Atrophy (n=20)	*No atrophy (n=121)*	*P*
Δ Sperm volume (ml)	0.1±0.4	0.2±1.1	0.71
Δ Sperm concentration ($\times 10^6$ ml^{-1})	10.5±4.6	11.8±16.0	0.71
Δ Motility (%)	5.9±5.8	8.5±9.0	0.23
Δ Forward progression (%)	4.8±10.5	5.4±16.9	0.88
Δ Total motile count (10^6)	13.6±16.2	38.7±21.8	0.0001
Δ Sperm DFI (%)	7.7±5.0	15.7±6.3	0.0001

Statistical analysis was performed by the Mann–Whitney U test. DFI: DNA fragmentation index. Δ represents change

DNA fragmentation. Although we found an improvement in semen quality overall and reduced DFI in men who had ipsilateral TA, they were not as robust improvements as in the NTA men. This may be important in counseling patients with a varicocele with ipsilateral TA as far as realistic expectations on responses to varicocele repair and helping to predict what levels of further fertility treatments may be indicated for the couples on the basis of the expected responses and baseline semen parameters and DFI. Our hypothesis on the difference in responses between the two groups is that men with a varicocele and testicular atrophy in the ipsilateral testicle have had enough testicular damage to induce atrophy which may make the responsiveness of the testicular cells less robust with the severity of testicular damage that induces atrophy.

Limitations of this study include the potential for inaccuracy for testicular volume measurement with Prader orchidometer, although this measurement technique has been used in multiple publications assessing testicular volumes and was used in this study by a single reproductive urologist with over a decade of experience with testicular volume measurement with this tool. However, it may be argued that the Rochester orchidometer or scrotal ultrasound will offer more accurate measurement tools. Another limitation is that not all men had semen analyzed at one andrology laboratory, which was the reason morphology was not included, as different criteria in assessing morphology were used in different laboratories. As some patients traveled from further distances and others selected laboratories on the basis of insurance coverage, this is not easily standardized. The 3-month postoperative semen analysis represents one cycle of spermatogenesis following varicocele repair, but it may be argued that a 6-month postoperative semen analysis would be useful to assess as well. That data would be more difficult to capture due to patients coming from distant locations and patients being lost to follow up. Although the optimal endpoint of such a study would be live birth, owing to confounding factors such as female factor and patients being lost to follow up beyond 3 months postoperatively, semen analysis results and sperm DNA fragmentation were taken as biochemical responses to varicocele repair. To our knowledge, this is the first study assessing the difference in responses between these two groups, which aids in counseling patients on expectations in terms of expected improvements in these metrics with varicocele repair.

AUTHOR CONTRIBUTIONS

PKK carried out research design, acquisition, analysis, and interpretation of data. NA, MSG, CH, GLM, SHC, KMK, AE, JDW, and SKK carried out acquisition, analysis, and interpretation of data. All authors read and approved the final manuscript.

COMPETING INTERESTS

PKK is on speaker bureau for AYTU Biosciences, Metuchen Pharmaceuticals and Antartes Pharmaceuticals. None of the above is relevant to this work. SKK is on speaker bureau for Abbvie, which is not relevant to this work. No other authors have disclosures.

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