

purpose of this study is to analyze patients with non-iatrogenic, non-traumatic OA who underwent scrotal exploration with or without VE anastomosis in our institution.

Methods: Retrospective chart review was done for patients treated from 2000 to 2015. Normal spermatogenesis confirmed by testis needle biopsy in all patients with OA. The operation method for VE anastomosis was two-stitch longitudinal intussusception technique. Patients who had prior vasectomy or history of vas deferens trauma/injury were excluded. The age, hormone profile, semen parameters, level of obstruction, semen quality at proximal epididymal cut-end and patency rates were analyzed.

Results: Totally 96 patients with mean age 35.4 ± 5.6 y/o were collected. The obstruction level was at: bilateral rete testis blockage (n=17), bilateral epididymis (n=49), bilateral intra-abdominal blockage (n=7), CBAVD variants (n=3). There were 68 patients received VE anastomosis. The patency rates 6 months after operation for patients with bilateral epididymal blockage were 88%. They were 90.9% for blockage at both epididymal tails, 88.9% at epididymal body, and 83.3% at epididymal head. Motile sperm at proximal cut-end had patency rates at 80.6%, while they were 50% in cases showing only immotile sperm. The mean patency rates were 79.5% for all cases 6 months after operation. Twenty-eight percent of patients failed to proceed the correction procedure, including 25% with high or low blockage, and 3.1% with CBAVD variants. Patients with CBAVD variants showed lower semen pH (6.67 ± 0.1 , $P=0.001$).

Conclusions: For patients with non-iatrogenic non-traumatic OA receiving scrotal exploration, 28.1% are not eligible for correction. The mean patency rates for all patients 6 months after micro-anastomosis were 79.5%. They were 90.9% and 88% for cases with obstruction at bilateral epididymal tails and body respectively. Motile sperm in proximal end predicts higher patency rates.

Keywords: Microsurgical vasoepididymostomy (microsurgical VE); azoospermic; non-vasectomy; non-traumatic etiologies

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AB024. Safety evaluation of low-energy extracorporeal shock wave therapy to rat testes

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Background: The purpose of the study is to determine the optimal energy frequency of LESWT to rat testes by comparing three different shock wave frequencies.

Methods: A total of 40 healthy Sprague-Dawley (SD) rats were randomly assigned to a blank group (C group) in which 5 rats received no treatment, a sham group (B group) in which 5 rats only exposed to shockwave, and LESWT groups (A group) in which 30 rats separately exposed to 0.02 mJ/mm^2 (A1 group), 0.04 mJ/mm^2 (A2 group), and 0.06 mJ/mm^2 (A3 group). The changes of rat weight among these five groups were compared. Eenzyme-linked immunosorbent assay (ELISA) was performed to measure the concentrations of testosterone (T), luteinizing hormone (LH), and follicle-stimulating hormone (FSH). Testicular mass index (TMI) of rat bilateral testes was compared. A transmission electron microscopy (TEM) was used to observe the microstructure of testes in rats among five groups.

Results: After a 2-week period of low-energy extracorporeal shock wave therapy (LESWT), the rat weight changed significantly between A1 and A3 groups but did not differ among A1, A2, B, and C groups ($P=0.09$, 0.31 , 0.47); no significant difference in T, LH, and FSH concentrations was observed when the A1 group was respectively compared with other four groups ($P=0.11$, 0.33 , 0.19 , 0.47); the TMI of rat testes in the A1 group was significantly different from that in the A3 group ($P=0.04$). Observations from the TEM showed that rats exhibited normal microstructure of testes in A1, B, and C groups. Mitochondrial swelling, dissolution of cristae, and dilated intercellular space occurred in the A2 group. Mitochondrial vacuolization, nuclear perturbations, cytoplasmic segregation, and necrotic cells were observed in the A3 group.

Conclusions: Taken together, we conclude that 0.02 mJ/mm^2 LESWT exerts no significant influence on rat weight, T, LH, FSH, TMI of testes, and microstructure, which may

be regarded as the safe frequency for rat testes undergoing LESWT. Furthermore, our study demonstrates that TEM is one of the most reliable methods for evaluating the effect of LESWT on microstructures of rat testes.

Keywords: Low-energy extracorporeal shock wave therapy (LESWT); testes; safety evaluation

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AB025. Semen liquefaction molecular pathways

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Background: Human semen is the jelly-like substance mainly containing Semenogelin 1 (Sg1) and fibronectin (Fn) with the characteristics of coagulation and liquefaction in a short time. In our previous study, we have identified that Eppin could interact with Sg. Eppin75–133 C-terminal fragment bind the Sg164–283 fragment containing the only cysteine in human Sg1 (Cys-239). Besides that, during semen liquefaction, physiologically prostate specific antigen (PSA) hydrolyzes some region of Sg1 which inhibits sperm motility. Therefore, complex interaction among Eppin, Sg and PSA plays a major role in regulating semen liquefaction process. The aim of this study is to investigate the molecular pathways during semen liquefaction.

Methods: Molecular cloning was used to recombination *in vitro* 6-His-Eppin protein with N- and C-terminal fragments. Protein in seminal vesicle fluid was transferred to Immobilon-P Polyvinylidene Difluoride membrane by western blot analysis, followed by incubation with 6-His-Eppin protein, Eppin283–423 C-terminal fragment and Eppin73–288 N-terminal fragment at 4 °C overnight respectively in order to find the protein which can be bind

to 6-HisEppin protein. 2-D electrophoresis was used to identification of Eppin binding partners. After that, anti-His was used to visualize using enhanced chemiluminescence and mass spectrometry to identify the sequence of protein.

Results: We found that the protein specifically binding to Eppin through Far-western immunoblot analysis demonstrated only the N-terminal of recombinant epididymal protease inhibitor (N-rEppin) and rEppin can binds to reduce seminal plasma protein, while MS identified that Fn can specifically bind to Eppin. Our study was the first evidence that some protein existed in seminal vesicle fluid does bind to Eppin, regulating the semen coagulation and liquefaction.

Conclusions: Two molecular pathways occurred in semen liquefaction. Eppin C- and N-terminal fragment interacted with Sg and Fn. Eppin N-terminal has a binding site of fibronectin, which is an important protein for semen coagulation. EpiPen regulated the process of semen coagulation and liquefaction through its N- and C-terminal bound to fibronectin and Sg respectively, influencing sperm capacitation. What's more, the C- and N-terminal fragment of Eppin self-formed double ring type molecular structure respectively and closely bound to the core structure named β -sheet by four-disulfide, which make the combination of Wap and Kunitze type inhibitor external surface just in the opposite two terminals of the transection of molecular structure. This structure contributes to interact with other proteins in increasing its roles. During human ejaculation, spermatozoa pass through the ampulla of the vas deferens and then move into the proximal extension of the seminal vesicle and finally enter into the ejaculatory duct. At this juncture spermatozoa are first mixed with copious secretion from the seminal vesicles. Thereafter the spermatozoa and seminal fluid is mixed with prostatic secretions when they enter into the prostatic urethra. It can be imagined that after spermatozoa enter into the ejaculatory ducts their surface Eppin would be saturated by binding with Sg and Fn. This process inhibits human sperm capacitation, making the initial ejaculated spermatozoa be in an immotile state. Purified plasma Fn, added at various concentrations to a preparation of live spermatozoa, was found to inhibit sperm motility in a dose dependent manner. During semen liquefaction, physiologically PSA hydrolyzes Sg and Fn to increase sperm motility. Therefore, seminal liquefaction is a process increasing the capacitation of sperm progressive motility and fertilization. Fibronectin could affect the process of sperm coagulation and liquefaction through specificity combined with Eppin, involving in sperm capacitation and fertilization. Our findings revealed novel