Abstract. – OBJECTIVE: The present study introduces the application of percutaneous epididymal sperm aspiration (PESA) for diagnosis of obstructive azoospermia and non-obstructive azoospermia.

PATIENTS AND METHODS: 96 cases diagnosed with azoospermia were selected, standard methods were used to measure testicular volume, chemiluminescence was used to test serum sexual hormone levels, and No. 7 butterfly needles were applied to puncture the head of the epididymis and aspirate epididymal luminal fluid.

RESULTS: Among 96 cases of azoospermia, sperm was found in the epididymal luminal fluid of 49 cases, among which there were 41 cases with normal testicular volume and 8 cases with low volume. 39 cases had normal serum FSH levels, and 10 cases had increased serum FSH levels. There were 47 cases with no sperm, among which there were 26 cases with normal testicular volume and 21 cases with low volume. 29 cases had normal serum FSH levels, and 18 cases had increased levels. The success rate of puncture for patients with normal testicular volume was higher than that of patients with low volume, and the difference was statistically significant ($p < 0.05$). The success rate of puncture for patients with normal serum FSH levels was higher than that of patients with increased levels, and the difference was statistically significant ($p < 0.05$).

CONCLUSIONS: PESA is simple and efficient, and is a feasible method for diagnosis of azoospermia.

Key Words: Azoospermia, Percutaneous epididymal sperm aspiration, Follicle stimulating hormone.

Introduction

5-15% of male infertility is caused by obstructive azoospermia. Previously, patients lacked effective treatment methods. Due to the emergence and development of intracytoplasmic sperm injection (ICSI) technology, patients suffering from obstructive azoospermia have a chance to reproduce. For infertile patients, the chances of having babies through ICSI is higher than with other treatment methods, apart from artificial insemination by a donor. Percutaneous epididymal sperm aspiration (PESA) or testicular sperm aspiration (TESA) with ICSI is a new technique to assist reproduction, which has been applied and generalized by more and more reproductive centers because it is easy to operate and is less painful for patients. Also, it brings higher fertility rates and pregnancy rates to infertile couples where the male suffers from aspermia. Diagnostic PESA technology can replace testicular biopsy which is more damaging, and results can be important references to identify obstructive azoospermia (OA) and non-obstructive azoospermia (NOA). Obstructive azoospermia is caused by seminal duct obstruction but not due to testicles not being seminiferous, while the latter is caused by spermatogenic dysfunction. Differential diagnosis is highly important because these two diseases are different in etiology, pathology, diagnosis, treatment and prognosis. From January 2010 to June 2014, PESA was used to treat 96 patients with azoospermia, among which there was mature sperm in the epididymal luminal fluid of 49 cases.

Patients and Methods

Patients

96 cases of azoospermia were selected in the Urinary Surgery Department of our hospital from...
January 2010 to June 2014. Their average age was 34.5 years old (range: 23-42 years old) and average infertility period was 4.5 years (range: 1-14 years) with normal sexual life. During a routine examination of semen, no sperm was found after at least 3 centrifugations. Serum hormone levels (FSH, LH, TE2 and PRL) were tested for all patients.

**Inclusion Criteria**
(1) At least one of the testicles had a volume of more than 12 ml; (2) Serum FSH level between 2.5-40 IU/L; (3) Elasticity of testicle is above average and epididymis is full; (4) Coagulation function is normal.

**Exclusion Criteria**
(1) Serum FSH level is more than 40 IU/L; (2) Both testicles had a volume less than 12 ml; (3) Patients had acute epididymitis, orchitis, chordeitis, vesciculitis, prostatitis, scrotal skin infection or eczema; (4) Patients with history of tuberculosis or epididymis in a beaded shape;

**Surgical Method (PESA)**
Lidocaine spermatic was used for enclosed local anesthesia. A 10 ml sample injector, with 3 ml sperm culture solution was connected with a No. 7 butterfly needle. The assistant fixed the testicle with the three-fingers technique, and the surgeon placed the epididymal caput below the scrotal skin and fixed it between the index finger and thumb; the other hand was used to hold the No. 7 butterfly needle to puncture the epididymal caput (blood vessels in scrotal skin were avoided during puncture), and aspirated the yellow or milk white epididymal luminal fluid until there was no outflow (if no epididymal luminal fluid came out, the location of the puncture needle was carefully adjusted). The epididymal luminal fluid was fixed on slides and sent for examination. If sperm was found under the microscope, the patient was diagnosed as having obstructive azoospermia. If not, the puncture was performed again on the same side, and if there was still no sperm, then the puncture was performed on the other side. The surgery was not performed if no sperm was found.

**Statistical Analysis**
Differences of success rates of puncture for different testicular volumes and different serum FSH level were analyzed by $\chi^2$-tests using SPSS14.0 software (SPSS Inc., Chicago, IL, USA). $p < 0.05$ was taken as statistically significant.

**Results**
Out of 96 cases of azoospermia, sperm was found in the epididymal luminal fluid of 49 patients, among which there were 41 cases with normal testicular volume and 8 cases with low volume. There were 39 cases with normal serum FSH levels and 10 cases with increased levels. There were 47 cases with no sperm, among which there were 26 cases with normal testicular volume and 21 cases with low volume. There were 29 cases with normal serum FSH levels and 18 cases with increased serum FSH levels. Our results show that the success rate of puncture for patients with normal testicular volume is higher than that of patients with lower volume, and the difference was of statistical significance ($p < 0.05$). Furthermore, the success rate of puncture for patients with normal serum FSH levels is higher than that of patients with increased levels, and the difference was statistically significant ($p < 0.05$) (Table I).

**Discussion**
Azoospermia refers to a sperm density of zero. That is, no sperm is found after centrifugation of sperm. The rate of azoospermia among male infertility patients is 15-20%\(^2\) and there are

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**Table I.** Comparison of puncture results for patients with different testicular volumes and FSH levels.

<table>
<thead>
<tr>
<th>Puncture results</th>
<th>Testicular volume (ml)</th>
<th>Serum FSH level (IU/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>≥ 20</td>
<td>12-20</td>
</tr>
<tr>
<td>With sperm</td>
<td>41</td>
<td>8</td>
</tr>
<tr>
<td>Without sperm</td>
<td>26</td>
<td>21</td>
</tr>
</tbody>
</table>

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many causes for this disease, which are divided into two categories: spermatogenic dysfunction, which is referred to as primary or non-obstructive azoospermia. The other cause is when there is a normal spermatogenic function, but the seminiferous duct is obstructed, preventing excretion of sperm, which is called obstructive azoospermia. These conditions lacked effective treatment methods until ICSI technology appeared and developed over the last ten years, allowing those with obstructive azoospermia to have children. Due to the development of ICSI technology, many sperm extraction methods appeared. Traditional methods, such as epididymis microsurgery and testicular biopsy, have high rates of sperm extraction but also disadvantages such as large operative wounds, high cost, and occurrence of complications\textsuperscript{5,6}. PESA has been applied and generalized by more and more reproductive centers because it is easy to perform and causes less pain to patients. It also offers higher fertility rates and pregnancy rates to couples with aspermic males\textsuperscript{7,9}.

The clinical indications of PESA include: (1) At least one of the testicles has a higher volume than 12 ml; (2) Elasticity of testicle is above average; (3) Serum FSH levels between 2.5-40 IU/L; (4) No history of tuberculosis or epididymis in a beaded shape; (5) Exclude acute epididymitis, orchitis, chordeitis, vesiculitis, prostatitis, scrotal skin infection or eczema; (6) Coagulation function is normal.

The advantages of PESA include: (1) It can easily, quickly and accurately identify obstructive azoospermia versus non-obstructive azoospermia; (2) When diagnosed as obstructive azoospermia, the specificity is high. That is, it is confirmed once sperm is found; (3) There is no need to cut the scrotal skin, resulting in little damage and less postoperative complications; (4) It is highly reproducible; (5) Sperm extracted through puncture can be cold stored for future use.

Among 96 cases of azoospermia in this study, sperm was found in the epididymal luminal fluid of 49 cases, among which there were 41 cases with normal testicular volume and 8 cases with low volume. There were 39 cases with normal serum FSH levels and 10 cases with increased serum FSH levels. There were 47 cases with no sperm, among which there were 26 cases with normal testicular volume and 21 cases with low volume. There were 29 cases with normal serum FSH levels and 18 cases with increased serum FSH levels. We show that

the success rate of puncture for patients with normal testicular volume is higher than that of patients with low volume, and the difference was of statistical significance ($p < 0.05$). The success rate of puncture for patients with normal serum FSH levels was higher than that of patients with increased levels, and the difference was of statistical significance ($p < 0.05$). We believe that in order to improve the success rate of puncture: (1) Based on detailed medical history, physical examination and routine tests, results such as sex hormone levels and B ultrasound, azoospermia should first be divided into obstructive azoospermia (OA), non-obstructive azoospermia (NOA) and combined azoospermia. It should not be diagnosed as azoospermia prematurely based on invasive tests such as testicular biopsy. (2) FSH, LH, and T (testosterone) indicators should be analyzed along with testicular volume, which is of predictive value for the testicular spermatogenic function.

Conclusions

Besides differential diagnosis of obstructive azoospermia and non-obstructive azoospermia, PESA can also be used in the detailed diagnosis and etiological analysis of azoospermia, aiming to predict prognosis and select a correct treatment plan.

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Conflict of Interest

The Authors declare that they have no conflict of interests.

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