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POWER DOPPLER ULTRASOUND MAPPING IN NONOBSTRUCTIVE AZOOSPERMIC PATIENTS PRIOR TO TESTICULAR SPERM EXTRACTION

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□ *This study was conducted to localize the testicular regions, which have better blood circulation by power Doppler ultrasonography in patients with nonobstructive azoospermia before testicular sperm extraction (TESE), and to investigate whether these vascularized areas have a high sperm retrieval rate or not. We evaluated 110 testes of 55 cases that were diagnosed as nonobstructive azoospermia. The mean age of the study group was 33 years (range 26 to 42). Patients with Y chromosome microdeletions, karyotype and hormonal abnormalities (except elevated FSH levels) were excluded from the study. In all cases, testes were evaluated by power Doppler ultrasonography before testicular sperm extraction. Testis was divided vertically into five equal parts and the area with maximum vascularity was determined subjectively. During testicular sperm extraction, starting from best-perfused areas, biopsies were done. If no motile or sufficient amount of sperm was found, TESE procedure was tried on the contralateral testis. TESE were performed from 82 testes and for the regions that show good and poor vascularity. The sperm finding rate was 38% and 14%, respectively ($OR = 3.55$)($p = 0.001$). Power Doppler ultrasound mapping of the testis in nonobstructive azoospermic cases is a reliable and informative method to assess spermatogenic foci. It is a noninvasive technique that minimizes the unnecessary removal of hormone producing tissue and gives chance to end the TESE earlier than currently practiced procedures.*

Keywords nonobstructive azoospermia, power Doppler ultrasound, sperm retrieval rate

INTRODUCTION

Azoospermia is defined as an entity with either obstruction of vas deferences or severe testicular failure resulting in no spermatozoa in the semen. Azoospermia is demonstrated in 20 to 30% of patients during the evaluation of infertile men [20]. Nonobstructive azoospermia is one of the main

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concerns in the treatment of infertile men. When mature spermatozoa and spermatids are present in the testis, intracytoplasmic sperm injection (ICSI) makes fertilization and pregnancy possible through the extraction of these cells from testicular tissue by testicular sperm extraction technique (TESE) [4, 8, 17]. However, TESE fails to obtain mature spermatozoa in 25–50% of men with nonobstructive azoospermia [9, 11, 13, 17, 18].

Primary testicular pathology in nonobstructive azoospermic patients is Sertoli cell only syndrome, or severe hypospermatogenesis, but rare isolated spermatogenic foci might be present [6]. Currently, there are no parameters to identify the presence of spermatogenic tissue within testis, and, consequently, multiple biopsies or microdissection tecnique are performed until spermatozoa are found [12].

There are some prognostic factors that determine sperm retrieval rate in nonobstructive patients by TESE. Testicular volume, genetic counseling, histologic pattern, serum FSH and inhibin levels have previously been investigated as a prognostic factor [2]. Nowadays, only the genetic counseling and preliminary testicular histology appear to have clinical value, but none of them give an idea in determining spermatogenic foci [16].

Regarding the geographic variation of spermatogenesis in these testes, it is important to localize these specific spermatogenic areas [5]. There is strong evidence that testicular blood flow can show the viable testicular areas as well as testicular function. Testicular blood flow depends on testicular volume and it increases with the maturation process leading to spermatogenesis [1]. Determination of testicular blood flow can be feasible in a noninvasive manner with power Doppler ultrasound that enables mapping as an informative and reliable method for the failing testis before TESE procedure [7]. Taking this information into account, our aim in this study is to investigate whether the vascularized testicular areas has a high sperm retrieval rate or not and determine the value of power Doppler ultrasound mapping in patients with nonobstructive azoospermia who are candidates for ICSI.

MATERIALS AND METHODS

Between February 2000 and December 2002, 110 testes of 55 cases who had a diagnosis of nonobstructive azoospermia and planned to undergo TESE procedure together with the in vitro fertilization (IVF) unit were evaluated. The mean age of the patients was 33 years (range 26 to 42). All patients underwent a standard diagnostic infertility evaluation that includes physical examination, hormonal analysis, genetic testing (Y chromosome microdeletion and karyotype analysis). The diagnosis of nonobstructive azoospermia was done by checking the seminal plasma fructose levels in all patients and by transrectal ultrasonography to evaluate if any dilatation

in ejaculatory ducts was present or not. Additionaly, 33 patients (60%) had already the pathological diagnosis of azoospermia in the testiculer biopsy prior to TESE. Patients with Y chromosome microdeletions, karyotype and hormonal abnormalities (except elevated FSH levels), and who had the diagnosis of normal spermatogenesis in the testiculer biopsy, were excluded from our study.

In all patients, before TESE, both testes were evaluated with gray scale and power Doppler ultrasonography (PDUS) for the determination of best perfused areas. All PDUS examinations were performed by an experienced radiologist. Imaging parameters were kept constant for all patients (pulse repetition frequency: 700 Hz (low), wall filter: medium, power gain: 82%). The images were stored in the hard-disc of the ultrasonography system, and then transferred to a personal computer for evaluation. Two radiologists, who have at least five years experience on PDUS (other than the radiologist who performed the examinations), evaluated all the images for the best perfused area in the testis. Initially, each testis was divided vertically into five equal parts. Then, the radiologists decided the best perfused area by counting the vascular signals from the vessels in all parts and made the evaluations together to reach a concensus in all patients.

Ejaculate from all of the patients was taken on the day of the operation routinely, and sperm was found in none of them. For the TESE procedure, a spermatic cord and scrotal skin block were done with 10 ml of 0.25% bupivacaine and a 2 cm incision was performed in the scrotal skin horizontally. Five-power optical magnification was used during the procedure. Tunical incisions were closed with 5.0 absorbable sutures and scrotal skin was approximated in 2 layers with 3.0 absorbable sutures. During testicular sperm extraction, starting from best-perfused areas, biopsies of above described all five areas were done. If no motile sperm was found, TESE procedure was tried on the contralateral testis. For immediate intraoperative interpretation, morphologically normal and motile sperm were recovered with the use of the gradient technique. Five microliters of supernatant from the tissue excised was propelled to center of a microdroplet of spermrinse medium. Spermatozoa reaching the microdroplet edge were taken with ICSI pipette and transferred to polyvinylpyrolidine. At the end spermatozoa was injected into the oocyte.

RESULTS

By using testicular vascular mapping prior to multiple testicular sperm extraction technique, sperm retrieval was successful in 31 of 82 vascular regions of testis (38%), whereas it was successful in 12 of 82 other regions of testis (14%). Overall, sperm extraction was successful in 31 of 55 patients (56%) (OR = 3.55). (CI: 1.57–8.13) (Yates Chi-square test = 10.21)

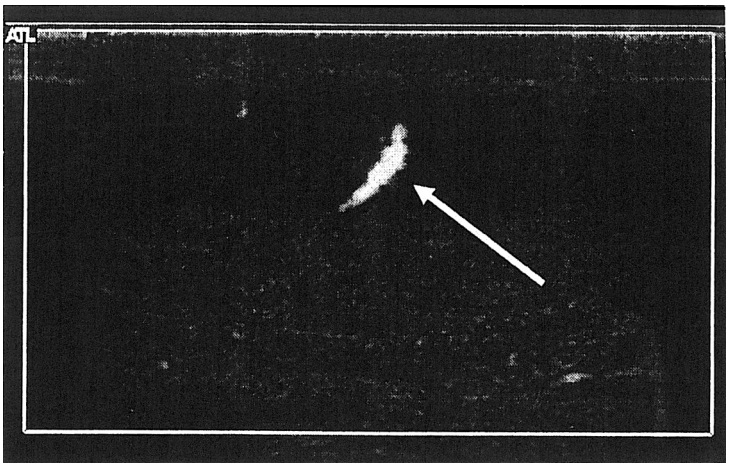


FIGURE 1 A power Doppler ultrasound imaging that shows the good perfused area (arrow).

($P = 0.001$). A power Doppler ultrasound imaging taken from a patient who has a good vascular region in his right testis, which was rich in mature spermatozoa, is shown in Figure 1.

In 27 patients, retrieval of motile sperm or the amount of qualified sperm was insufficient, so these patients underwent bilateral TESE procedure.

None of the patients needed more than 5 hours of hospitalization after TESE. Scrotal pain was inevitable and treated by oral analgesics postoperatively.

No clinical or surgical complication like hematoma, severe pain, or infection was observed in early postoperative period and during the control one month after the procedure.

DISCUSSION

Testicular sperm extraction and intrastoplasmic sperm injection were introduced into urologic armamentarium in 1993, and these techniques led us to new possibilities for the treatment of men with nonobstructive azoospermia [14]. Success rates of the TESE procedure are predictible by the evaluation of the prognostic factors affecting the sperm retriaval rate. Testicular volume, serum FSH level and the histologic pattern of the testis have been investigated, but only the presence of sperm in preliminary biopsy appear to have some predictive power [18].

Most patients with azoospermia have the nonobstructive type who present with poor spermatogenesis, small testis, normal ejaculate volume and elevated serum FSH level [10, 13]. These are the reliable indicators

of germinal epithelial damage associated with a Sertoli cell only pattern, maturation arrest, or hypospermatogenesis [13].

Mullhall et al. reported the use of simultanous diagnostic and theurapeutic TESE with cryopreservation and detected 70% sperm recovery rate [11]. Again, Tournaye performed TESE with the multibiopsy approach and reported 85% sperm recovery rates in 54 nonobstructive patients. However, their overall results by expanding their series with 222 cases were only 52% [18]. Another method to increase these sperm retrieval rates is the multibiopsy approach performed with optical magnification. In 1997, Schlegel et al. found a 62% sperm recovery rate using this approach [13].

Regarding the emotional and financial costs associated with recurrent IVF cycles, need of prior evaluation of the testis in order to localize spermatogenic foci is obvious. For this purpose, Silber and Tournaye made extensive multiple biopsies from every area of the testis to find spermatogenic foci before the TESE procedure [15, 18], but this resulted in testicular damage and even limited successful TESE procedure in patients whom they could not find sufficient spermatozoa by needle biopsy.

On the other hand, TESE itself is not entirely a benign procedure and, unfortunately, testicular sperm aspiration and TESE involves considerable amount of trauma related with pain, hematoma formation and infection. Also postoperative adhesions and fibrosis limit the application of additional surgey to testis in the future [3]. Turek et al. found sperm in 27% of cases when previous testicular biopsies were negative by fine needle aspiration mapping [19].

Foresta revealed that testicular structure in nonobstructive azoospermia is severely altered and testicular blood flow is strongly decreased or absent in intratesticular arteries [7]. Bader et al. reported that in young boys, testicular blood flow depends on testicular volume, and it increases with the maturation process leading to spermatogenesis [1]. This suggests a relationship between blood flow and the testicular function. Therefore, certain regions in testis where the blood vessels survived are the places that residual spermatogenic activity persist [7]. From this point of view, in this study, we tried to determine the relatively hypervascularized places of the testis by power doppler ultrasound imaging to characterize the blood supply of each testis before the TESE procedure. These regions were assumed to be the residual spermatogenic areas in nonobstructive azoospermic patients. This assumption seems to be true as sperm retrieval was significantly more successful in the relatively hypervascularized areas of testis than in other areas.

Variation in spermatogenic foci in testicular tissue explains why removal of a small amount of tissue blindly with needle or open procedure may or may not yield spermatozoa [15]. So the solution seems to be either removal of larger amounts of tissue or microsurgical selection of small

spermatogenic foci during TESE procedure or detecting the focus, which is rich in spermatogenic activity. Our results show that power Doppler ultrasound mapping can find these areas with localizing the high vascular foci, and this method can be used to increase the TESE rates.

Doppler ultrasound is a noninvasive technique and can evaluate the intratesticular blood flow [7]. It is also cheap and easy to perform, however, it needs an experienced radiologist to localize the vascular foci. There are two different modes of Doppler ultrasound (color and power Doppler) for detecting the vascular structure. Foresta et al. reported that both color and power Doppler ultrasound could be effectively used for the presence of blood vessels, indicating the possible presence of residual spermatogenic foci in nonobstructive patients [7]. The reason why we preferred power Doppler ultrasound rather than color Doppler ultrasound in the assessment of intratesticular vascularization depends on the study done by Bader et al., who reported that power Doppler ultrasound is more sensitive than color Doppler ultrasound [1].

As a conclusion, power Doppler ultrasound mapping of the testis in nonobstructive azoospermic cases seems to be a reliable and informative method to assess spermatogenic foci. Taking into account the unpredictable variation of these foci in testis, guiding TESE with power Doppler imaging is undoubtedly more logical and less invasive then prior mapping with fine needle aspiration or multiple TESE. Furthermore, it minimizes the unnecessary removal of hormone producing tissue as it gives a chance to end the TESE earlier then currently practiced procedures. This is especially important in nonobstructive men with small testicular volumes.

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