Sperm retrieval for obstructive azoospermia

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Advances in the treatment of male infertility now routinely allow men with obstructive azoospermia to have fertility treatment without microsurgical reconstruction. A variety of methods for retrieving sperm from men with obstructive azoospermia have been described. The goals of sperm retrieval are to obtain the best quality sperm possible, to retrieve adequate numbers of sperm for immediate use and for cryopreservation, and to minimize damage to the reproductive tract. (Fertil Steril® 2008;90:S213–8. ©2008 by American Society for Reproductive Medicine.)

Advances in the treatment of male infertility now routinely allow men with obstructive azoospermia to have fertility treatment without microsurgical reconstruction. Two developments have facilitated such treatments. First, micromanipulation techniques applied to sperm and ova in vitro allow sperm with limited intrinsic fertilizing capacity to produce embryos. Second, it is now widely recognized that viable sperm can be obtained reliably from the testis and epididymis of men with obstructive azoospermia. These developments have stimulated interest in techniques of sperm retrieval in obstructive azoospermia. Although the ideal method of sperm retrieval has not been established, the technique should be safe, efficient, and reliable in retrieving adequate numbers of sperm with optimal quality. This report describes methods for retrieving sperm from men with obstructive azoospermia for use in conjunction with assisted reproductive technology (ART).

INDICATIONS AND CONTRAINDICATIONS

Azoospermia can have obstructive or nonobstructive causes. This bulletin reviews techniques that can be used to treat individuals with obstructive azoospermia. Nonobstructive azoospermia, characterized by abnormal testicular histology such as maturation arrest or Sertoli cell only, previously was considered a contraindication to sperm retrieval. However, men with nonobstructive azoospermia now can be treated effectively with sperm retrieval from the testis. Evaluation and treatment for men with nonobstructive azoospermia are very different from those for men with obstructive azoospermia and are not discussed in this Technical Bulletin. Medical specialists who perform sperm retrieval must be familiar with the relevant scrotal anatomy, have the necessary technical expertise, and possess the experience and skill required to manage effectively any related complications.

Obstructive azoospermia may be congenital or acquired. Perhaps the best known congenital form of male reproductive tract obstruction is congenital bilateral absence of the vas deferens (CBAVD) (1). The abnormality is associated with mutations in the cystic fibrosis transmembrane-conductance regulator (CFTR) gene that have been detected in 38%–71% of individuals with CBAVD (2–4). However, failure to identify a CFTR abnormality in a man with CBAVD does not exclude completely the presence of a mutation because many mutations are not detected by routine testing methods. Since it can be assumed that a man with CBAVD harbors a genetic abnormality in the CFTR gene, it is important that the female partner be tested for CFTR mutations before treatment with IVF using sperm retrieved from a man with CBAVD. Abnormalities associated with CBAVD include partial or complete absence of seminal vesicles and epididymis and renal collecting system anomalies. A more detailed discussion of associated medical and genetic anomalies in men with CBAVD is the subject of a separate American Society for Reproductive Medicine document (5).

Additional evaluation, treatment, and counseling are indicated for men with laboratory evidence of HIV, syphyllis, or hepatitis (hepatitis B surface antigen, hepatitis B core antibody, and hepatitis C antibody) infections before considering sperm retrieval and ART. Acquired male reproductive tract obstructions may result from infection, vasectomy, or trauma (including iatrogenic injury during bladder neck, pelvic, abdominal, or inguino-scrotal surgery). In general, the optimal treatment would allow the couple to conceive naturally. For example, vasectomy reversal has been shown to be more cost effective than sperm retrieval and assisted reproduction (6, 7). When reconstruction is impossible (as in CBAVD), unlikely to succeed, or has already failed (as in previous attempts at vasovasostomy or vasoepididymostomy), sperm may be retrieved directly from the epididymis or testis and used for ART. In case series reporting the results of sperm retrieval and intracytoplasmic sperm injection (ICSI) for men with obstructive azoospermia, pregnancy rates per retrieval attempt have ranged between 24% and 64% (8, 9), comparable or better than those achieved for similar couples at the same centers using ejaculated sperm.

PATIENT EVALUATION

The evaluation for male factor infertility should begin with a thorough history of previous fertility and risk factors for

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obstruction, including scrotal, inguinal, pelvic, or abdominal surgery, and any history of genitourinary infection. The physical examination should include a careful inspection of the abdomen, inguinal region, and genitalia for surgical scars or signs of trauma. The vas deferens and epididymis should be examined for evidence of obstruction (such as epididymal induration or fullness) and partial or complete absence of the vas deferens and epididymis. A careful digital rectal examination can reveal cystic dilation of the seminal vesicles (often associated with other Wolffian duct anomalies) or a midline prostatic cyst that may cause ejaculatory duct obstruction.

Laboratory evaluation in individuals with suspected obstructive azoospermia should include at least one semen analysis and serum for baseline testosterone and FSH levels. An FSH >7.6 IU/L and testicular length <4.5 cm predicts the presence of nonobstructive azoospermia with greater than 89% probability in azoospermic men (10). Both serum FSH and a thorough physical examination are important because reproductive tract obstruction and impaired spermatogenesis are not mutually exclusive conditions. For many patients with normal FSH levels and presumptive obstructive azoospermia, a testicular biopsy may be used to evaluate sperm production. However, biopsy is not necessary in men with CBAVD having a normal serum FSH and testes volume because adequate spermatogenesis can be expected.

In men who are candidates for sperm retrieval, semen analysis should reveal azoospermia (absence of sperm after centrifugation of a semen specimen for 15 minutes at $\times 1500g$) or necrospermia (absence of live sperm in the ejaculate). However, other semen parameters also are useful. Semen volume can distinguish epididymal or vasal obstruction (normal semen volume) from ejaculatory duct obstruction (low volume). The presence of normal semen volume or fructose in semen and an alkaline pH establish that seminal vesicles are present and localize the obstruction to a site(s) proximal to the junction of the vas deferens and seminal vesicle. The presence of any mature sperm in the semen excludes the possibility of complete absence of spermatogenesis.

Men with Wolffian developmental abnormalities, including CBAVD and idiopathic epididymal obstruction, are at increased risk for harboring CFTR mutations, as discussed earlier. If both partners have CFTR mutations, the couple should receive counseling by a qualified genetic counselor to ensure that they thoroughly understand the treatment alternatives and the risks for conceiving an affected child before proceeding with sperm retrieval and assisted reproduction. Such counseling should include a discussion of methods for preimplantation genetic diagnosis that may be used to select unaffected embryos for transfer.

Ultrasound is another important diagnostic and therapeutic tool in the evaluation and treatment of infertile azoospermic men. Renal ultrasound should be offered to any individual with unilateral or bilateral vasal agenesis because these abnormalities commonly are associated with renal anomalies. Transrectal ultrasound can diagnose ejaculatory duct obstruction or an obstructing Müllerian or ejaculatory duct cyst.

SPERM-RETRIEVAL TECHNIQUES

Sperm retrieval for use in ART may be viewed as a primary treatment or as an adjunct to microsurgical reconstructive procedures. The goals of sperm retrieval are to obtain the best quality sperm possible in adequate numbers, for both immediate use and for cryopreservation, and to minimize damage to the reproductive tract. The techniques described later include sperm retrieval at the time of surgical reconstruction, microsurgical epididymal sperm aspiration, intraoperative testicular sperm retrieval, and percutaneous techniques of epididymal and testicular sperm retrieval (Table 1).

Intraoperative Testicular Sperm Retrieval During Vasovasostomy and Vasoepididymostomy

Since only 20% to 40% of couples conceive after attempted vasoepididymostomy despite patency rates of 60% to 80%, it is reasonable to consider sperm retrieval at the time of surgical reconstruction. Intraoperative sperm retrieval is particularly important when a difficult reconstruction is anticipated, as in men who have undergone a previous vasovasostomy or vasoepididymostomy or other scrotal procedure where inflammation or scarring may obscure surgical planes. Preoperative counseling should include a thorough discussion of the risks and benefits of the procedure, the potential need for subsequent ART, and cost of cryopreservation because not all couples will be willing or able to pursue ART.

TABLE 1

Sperm-retrieval techniques.

Procedure type	Epididymal	Testicular
Open	Epididymal sperm aspiration (MESA)	Open testicular biopsy
Percutaneous	PESA	Percutaneous testicular Biopsy (PercBiopsy)
		Testicular sperm aspiration (TESA), (TEFNA), (FNA)

Note: PercBiopsy = percutaneous testicular biopsy; TESA = testicular sperm aspiration; TEFNA = testicular fine-needle aspiration; FNA = fine-needle aspiration.

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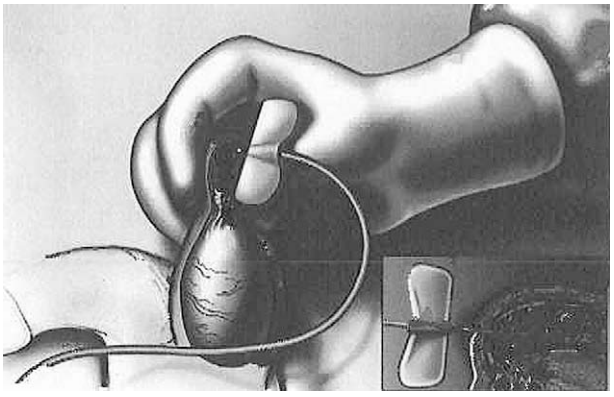
FIGURE 1

Schematic representation of percutaneous epididymal sperm retrieval. With the testis stabilized between the surgeon's thumb and forefinger, a butterfly needle attached to a syringe is inserted into the caput epididymis and withdrawn until fluid is seen entering the tubing.



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If motile sperm are found at the site of reconstruction, they may be aspirated and cryopreserved. Whereas the most motile and best quality sperm usually are obtained from the most proximal epididymis, reconstruction is most successful when the repair is more distal. The choice of the optimal site for microsurgical reconstruction/repair should not be compromised to obtain better sperm for cryopreservation because retrieving sperm from a proximal tubule may cause obstruction at that site. Alternatively, sperm may be retrieved via testicular biopsy. The tunica albuginea is incised and a portion of seminiferous tubules is extruded by gentle compression. Sperm then may be extracted from the excised testicular tissue by mincing, vortexing, or sequential passage of the tissue suspension through a narrow gauge needle. Care must be taken to avoid injuring testicular blood vessels in the tunica albuginea during biopsy retrievals. Sperm retrieved directly from the testis are generally limited in numbers and often exhibit no motility or nonprogressive motility but nevertheless typically still are viable and almost always functional for use in ART.

EPIDIDYMAL SPERM-RETRIEVAL TECHNIQUES

Microsurgical Epididymal Sperm Aspiration

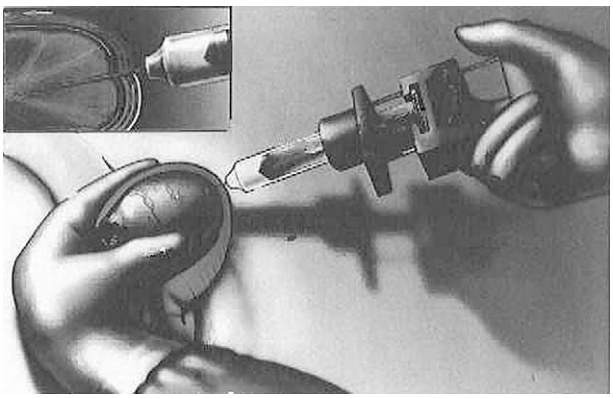
Microsurgical epididymal sperm aspiration (MESA) is a term used to describe microsurgical techniques for sperm aspiration from the epididymis. Several techniques have been described, including incision of individual epididymal tubules with aspiration of the effluent, and micropuncture of specific individual epididymal tubules (11, 12). Briefly, single epididymal tubules can be identified under the operating microscope and individually aspirated with an atraumatic technique. Sequential aspirations can be performed until sperm of optimal quality have been obtained. The best quality sperm are typically found in the proximal epididymis close to the testis. Puncture sites may be closed or cauterized. Only trace volumes of aspirate need to be retrieved since sperm are highly concentrated in the epididymal fluid (approximately 1 million sperm/μL). MESA typically yields sufficient numbers of sperm for both immediate use and for cryopreservation.

Percutaneous Epididymal Sperm Aspiration

Epididymal aspiration also can be performed without surgical scrotal exploration, repeatedly, easily, and at low cost, without an operating microscope or expertise in microsurgery. Percutaneous epididymal sperm aspiration (PESA) can be performed under local anesthesia. After induction of anesthesia (typically with a spermatic cord block), the epididymis is stabilized between the surgeon's thumb and forefinger. A butterfly needle attached to a 20-mL syringe is inserted into the caput epididymis and withdrawn gently until fluid can be seen entering the attached tubing. The procedure is repeated until adequate amounts of epididymal fluid are retrieved (8) (Fig. 1). If no sperm are retrieved, MESA, testis biopsy, or testicular aspiration can be performed. Concerns have been raised that sperm obtained via PESA may include older, degenerated sperm. In one study, sperm retrieved by PESA exhibited more DNA damage than sperm retrieved

FIGURE 2

Schematic representation of percutaneous testicular sperm retrieval. The testis is stabilized between the surgeon's thumb and forefinger and the needle is inserted along the long axis of the testis. The needle is withdrawn slightly and redirected in order to disrupt the testicular architecture. The procedure is repeated until adequate testicular material has been aspirated. A Franzen needle holder can be used to provide negative pressure for needle aspiration.



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directly from the testis (13). However, the same risk does not appear to be associated with selective direct sperm retrieval from selected sections of the epididymis via MESA.

TESTICULAR SPERM-RETRIEVAL TECHNIQUES

Open Testicular Biopsy

Open testicular biopsies can be used to obtain sperm, although the epididymis is a richer source of sperm than the testis. There is no clear difference in pregnancy rates achieved with ART using testicular or epididymal sperm from men with obstructive azoospermia. However, open testicular biopsy is necessary to find sperm for men with non-obstructive azoospermia. Multiple testicular incisions are best avoided because the testicular arteries are end arteries and injury during biopsy may cause partial testicular infarction (14). The risk of injury to the testicular blood supply may be minimized by using an operating microscope to visualize testicular vessels directly (15).

Percutaneous Testicular Sperm Aspiration

The technique of percutaneous testicular fine-needle aspiration was initially described as a diagnostic procedure performed under local anesthesia in azoospermic men (16). The approach sometimes also is called testicular sperm aspiration. After stabilizing the testis between the surgeon's thumb and forefinger, a needle is inserted along the long axis of the testis, withdrawn slightly, and redirected in attempts to disrupt the testicular architecture. The procedure is repeated until adequate testicular material has been aspirated (Fig. 2). The goals of the procedure are to disrupt tubules and aspirate their contents and to remove individual tubules.

Percutaneous Testicular Biopsy (PercBiopsy, Needle Biopsy)

In the technique of percutaneous testis biopsy, a 14-gauge biopsy gun with a short (1 cm) excursion is used to retrieve testicular tissue. The gun removes a small cylinder of testicular tissue atraumatically. Anesthesia is achieved with a spermatic cord block, and multiple biopsies can be obtained through a single entry site. The core needle provides better sperm yield than fine-needle aspiration and is relatively simple to use (17). The advantages and disadvantages of each of the techniques for sperm acquisition are listed in Table 2.

ROLE OF ASSISTED REPRODUCTION

Once viable spermatozoa have been retrieved, the likelihood for achieving pregnancy via ART is greatest with ICSI. Although there are anecdotal reports of pregnancies after

TABLE 2

Advantages and disadvantages of sperm retrieval techniques.

	Advantages	Disadvantages
MESA	Best clinical pregnancy rates Large number of sperm retrieved Excellent results with cryopreservation Reduced risk of hematoma	Requires microsurgical expertise Increased cost General or local anesthesia Incision required Postoperative discomfort
TESE	No microsurgical expertise required Local or general anesthesia Few instruments Fast and repeatable	Relatively few sperm retrieved Limited risk of testicular atrophy (with multiple biopsies)
PESA	No microsurgical expertise required Local anesthesia Few instruments Fast and repeatable Minimal postoperative discomfort	Few sperm retrieved Risk of hematoma Damage to adjacent tissue
PercBiopsy, TESA, TEFNA	No microsurgical expertise required Local anesthesia Few instruments Fast and repeatable Minimal postoperative discomfort	Few sperm retrieved Risk of testicular atrophy Risk of hematoma

Note: TESE = testicular sperm extraction; PercBiopsy = percutaneous testicular biopsy; TESA = testicular sperm aspiration; TEFNA = testicular fine-needle aspiration.

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intrauterine insemination with surgically retrieved sperm, reports from several centers comparing fertilization and pregnancy rates achieved with conventional methods of IVF and with ICSI have demonstrated the importance of ART. In the Sperm Microaspiration Retrieval and Assisted Reproductive Technologies study, which reviewed the overall United States experience (219 retrieval procedures), the observed clinical pregnancy rate was only 10% using microsurgically retrieved sperm and conventional IVF (18). The results achieved with retrieved sperm and ICSI are far superior. Contemporary pregnancy rates of 24% to 64% (comparable to that achieved with ejaculated sperm at individual centers) have been achieved using sperm retrieved from azoospermic men (8, 9). Maternal factors (maternal age, oocyte number, and oocyte quality) alone now are considered the principal determinants of outcomes achieved with ART and ICSI for couples with infertility related to obstructive azoospermia (19, 20).

IMPORTANCE OF CRYOPRESERVATION

Cryopreservation of sperm is an essential technique in the treatment of infertile couples wherein the man has obstructive azoospermia. Cryopreservation avoids the logistical difficulties associated with attempts to coordinate sperm retrieval and ART on the same day and permits the detection of infectious disease prior to insemination.

Whenever available, excess retrieved spermatozoa should be cryopreserved to avoid unnecessary subsequent sperm retrieval procedures. Although cryopreservation has detrimental effects on sperm motility (21) and sperm acrosomes (22), outcomes achieved with ICSI using frozen-thawed or fresh spermatozoa retrieved from men with obstructive azoospermia are comparable (23).

RISKS OF INTERVENTION

More invasive techniques of sperm retrieval such as MESA or testicular sperm extraction are associated with a longer recovery interval than percutaneous testicular or epididymal sperm retrieval procedures. The incidence of hematomas and other complications after MESA, PESA, or percutaneous testicular aspiration appears to be low. Since more sperm of higher quality typically are retrieved via MESA, only one intervention is usually needed to provide adequate sperm for multiple attempts at ICSI. Repeat percutaneous testicular aspirations may become necessary if the first ART procedure does not result in a live birth.

Sperm-retrieval techniques appear to be safe, especially when performed using local anesthesia. Proximal sperm retrieval may result in permanent obstruction of the proximal epididymis and preclude subsequent microsurgical reconstruction. The risk should be considered carefully before choosing sperm retrieval over reconstruction. Permanent testicular devascularization has been reported after attempted sperm retrieval from multiple testicular sites using an open biopsy approach (14). However, operative morbidity and permanent injury to the testis appear to be rare, especially for men with obstructive azoospermia who typically require only a single testicular incision. The risk of birth defects after sperm retrieval with ICSI is comparable to that associated with ICSI using ejeculated sperm.

SUMMARY AND CONCLUSIONS

- Sperm retrieval with ART is an alternative to microsurgical repair for men with correctable reproductive tract obstruction and represents the only treatment that can offer men with irreparable obstruction the opportunity to have their own genetic children. Almost all men with obstructive azoospermia have abundant sperm in the testes that can be retrieved successfully using a variety of different techniques.
- Genetic (CFTR) testing of at least the female partner is essential for couples in which the male partner has bilateral vasal agenesis, and is recommended for all couples wherein the male partner has congenital reproductive tract obstruction.
- Microsurgical reconstruction should be offered to men having a reparable reproductive tract obstruction.
- The best technique for sperm aspiration for men with obstructive azoospermia has not been determined. The choice of technique should consider the need for microsurgical expertise and the longer recovery accociated with open surgical procedures compared with percutaneous procedures.
- The goals of sperm retrieval are to obtain the best quality sperm possible, to retrieve adequate numbers of sperm for immediate use and for cryopreservation, and to minimize damage to the reproductive tract.
- Regardless of which approach is used to retrieve sperm from men with obstructive azoospermia, ICSI should be used to maximize fertilization efficiency and overall results.
- When treatment requires a complex reconstruction of the male reproductive tract, cryopreservation of retrieved sperm should always be considered because surgery may not be successful.

Acknowledgments: This report was developed under the direction of the Practice Committee of the American Society for Reproductive Medicine as a service to its members and other practicing clinicians. While this document reflects appropriate management of a problem encountered in the practice of reproductive medicine, it is not intended to be the only approved standard of practice or to dictate an exclusive course of treatment. Other plans of management may be appropriate, taking into account the needs of the individual patient, available resources, and institutional or clinical practice limitations. The Practice Committee of the American Society for Reproductive Medicine and the Board of Directors of the American Society for Reproductive Medicine have approved this report.

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