THE SCIENCE OF ASSISTED REPRODUCTIVE TECHNOLOGY

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Summary

Assisted Reproductive Technology (ART) generally is divided into three fields (gametes [spermatozoa and oocytes], embryos, and third party reproduction). These topics can be subdivided into such techniques as husband insemination, in vitro fertilization and embryo transfer, gamete intrafallopian transfer, and zygote intrafallopian transfer. In addition, these aforementioned methodologies also can be used with donor gametes making it third party reproduction. There are other topics of interest that fall within the purview of ART that include human immunodeficiency virus, stem cell research, preimplantation genetic diagnosis, adoption, and rights of children. A general discussion of each of these topics is included in this narrative as well as the ethics that are involved with each of these topics. Furthermore, the pros and cons of each of these topics are offered as well as the medical indications for the use of each of these methodologies, research that is still needed in each of these areas and general references, should the reader desire further knowledge on any of these specific topics.

1.Gametes

1.1. Insemination with Semen from the Husband

1.1.1. Background

The goal of insemination with husband's semen is to have the husband's sperm reach the wife's oviducts (fallopian tubes) at the appropriate time. The technique requires that, near the time of ovulation, semen be placed near or in the wife's uterus. To insure proper timing of the procedure, the wife can use ovulation detection kits or basal body temperature charts.

One method for spermatozoa to reach the oviducts is called intracervical insemination (see Figure 1). The procedure requires that the woman lie on an examination table, while a physician places a speculum into her vagina. With a catheter attached to a syringe, the clinician dispenses the semen in and near the cervix. To keep sperm near the cervix, the clinician may place a plastic-coated sponge or a cap into the vagina. If a patient uses a sponge or cap, she usually removes it after four to six hours.

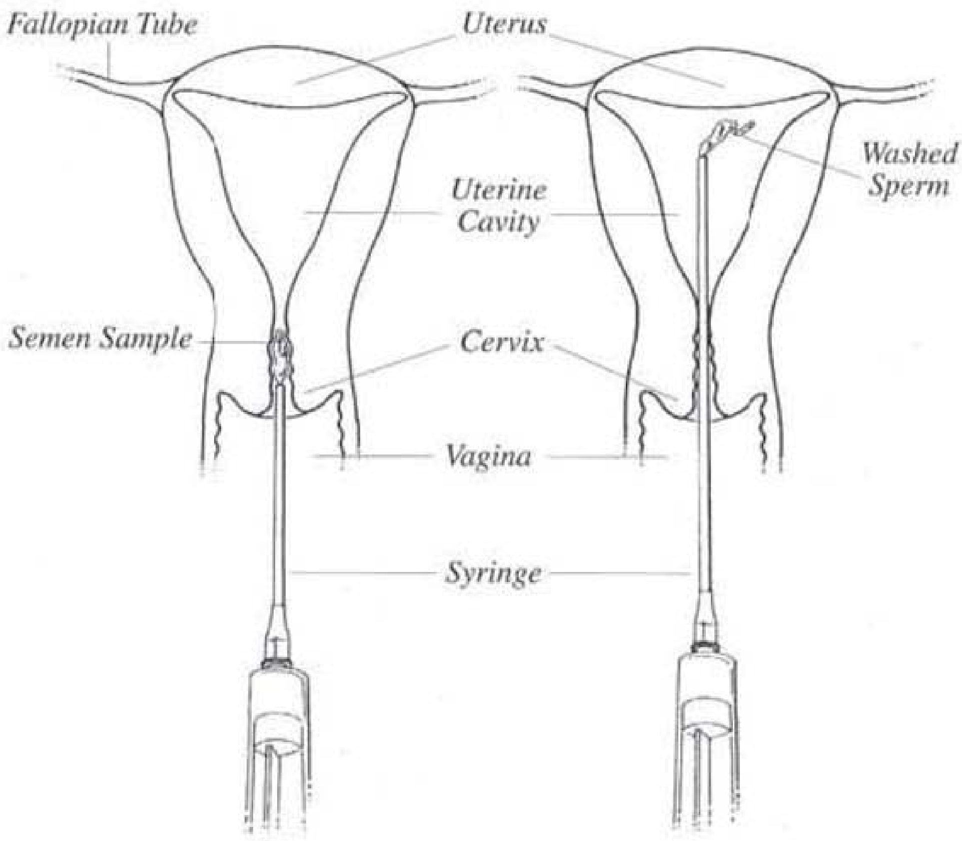

Figure 1. Two types of insemination. (Copyright© 2008 by the American Society for Reproductive Medicine. All rights reserved. No part of this presentation may be reproduced or transmitted in any form or by any means, electronic or mechanical, including photocopying, recording or by any information storage and retrieval system without permission in writing from the American Society for Reproductive Medicine, 1209 Montgomery Highway, Birmingham, AL 35216.)

For spermatozoa to migrate to the oviducts, spermatozoa may need to be placed in the uterus instead of the vagina or cervix. If so, the laboratory personnel must wash the semen to separate sperm from seminal fluids. The physician performs this procedure to reduce the chance of bacterial infections, contractions, or more severe reactions. These reactions are in response to the prostaglandins and other chemicals that are located within the seminal fluid portion of semen. Thus, sperm washing allows the clinician to place the sperm directly into the uterus. By placing more sperm near the cervix, the physician improves the patient's chance of becoming pregnant.

An additional aspect of a husband intrauterine insemination (hIUI) is the fact that scientists can cryopreserve sperm for later inseminations. Bunge and Sherman reported the first human pregnancy produced from frozen sperm more than 50 years ago. This technique now allows husbands to store spermatozoa for future use before undergoing a vasectomy, testicular surgery, or radiation/chemotherapy for cancer treatment. Cryopreservation even provides opportunity for inseminations without the husband's being present (see Section 1.3).

The sperm that enters the oocyte determines the gender of a child. Generally, semen carries about 50% X-bearing sperm (produces a female) and 50% Y-bearing sperm (produces a male). For gender selection to be successful, scientists must separate these sperm.

Today, few methods allow scientists to truly separate these sperm with a high degree of success. The procedure that appears to be most successful is MicroSort®. Because the X chromosome is larger and contains approximately 2.8% more DNA (chromosomes are made of DNA) than the Y chromosome, scientists can use MicroSort® fluorescence in situ hybridization (FISH) to separate these sperm. When patients used X-sorted sperm, 91% (295/325) of the babies born were females, while 76% (39/51) of the babies born were males when the patients used Y-sorted sperm. The patient can use sperm sorting, along with hIUI, to improve the odds of producing a child without an X-linked genetic defect (e.g., hemophilia, Duchenne muscular dystrophy, several X-linked diseases, fragile X syndrome) or for family balancing (see Section 4.3).

In addition to sperm sorting and hIUI to select against X-linked genetics, patients can choose Preimplantation Genetic Diagnosis (PGD). Preimplantation Genetic Diagnosis is used to identify embryos that carry X-linked genetic disorders. For a more complete description of PGD, including ethical issues involved with the technology, please see the section titled Preimplantation Genetic Diagnosis.

1.1.2. Medical Indications

There are numerous indications for hIUI. For example, if the husband is unable to successfully deposit sperm into the wife's vagina because of severe hypospadias, retrograde ejaculation, drug-induced erectile dysfunction (antihypertensive therapy), or vaginal dysfunction or cervical mucus abnormalities that cannot be corrected in the wife, then hIUI becomes an option for the wife to become pregnant. The couple potentially can alleviate other medical problems with the use of cervical or intrauterine inseminations. These medical problems include spermatogenesis problems (oligospermia [a deficiency of spermatozoa], asthenospermia [a deficiency in the percent of normal, motile spermatozoa], or teratospermia [a deficiency in the percent of normal-appearing spermatozoa]) or even antisperm antibodies. In addition, if the husband has his sperm sorted before the couple undergoes intrauterine insemination, the couple can increase their odds of avoiding X-linked genetic diseases.

While hIUI can improve the chance of a woman to conceive, the techniques may not always be appropriate, For example, if the husband has sperm that adhere to one another (antisperm antibodies), separating the sperm from the seminal fluid through washing may only enhance the agglutination process. Once adhered, the sperm are unable to migrate normally through the female reproductive tract.

1.1.3. Issues

1. **Con:** Because many medical indications are uncertain (cervical mucus abnormalities, spermatogenesis problems, and antisperm antibodies), hIUI may not be the appropriate technique to use.
Pro: With improved clinical studies, identification and resolution of these uncertainties may get better.
2. **Con:** Unless the wife undergoes hormonal synchronization to control ovulation, hIUI may be of little value. Ideally, the physician should perform serial ultrasounds to predict the time of ovulation. These ultrasounds are often expensive and consume time.
Pro: Serial urinary or serum levels of specific hormones are good predictors of ovulation; scientists have improved these tests over the years.
3. **Con:** Genetic selection of sperm for X- and Y- chromosomes is not perfected.
Pro: Granted that technology for genetically selecting sperm has not been perfected, but there are claims of 90^{+}% accuracy for X or Y selection.
4. **Con:** hIUI may separate procreation from sexual expression.
Pro: hIUI may be the only means a couple has to conceive.
5. **Con:** Physicians perform many hIUI procedures without benefit of a complete semen analysis on the specimen before they perform the insemination. Sperm concentration, motility, or morphology may be too low to be effective.
Pro: For most specimens, laboratory personnel can determine sperm concentration, motility, and gross morphological anomalies within minutes of collection. While not perfect, this relatively quick semen evaluation can eliminate some of the more severe cases of male factor before hIUI.

1.2. Sperm Aspiration

1.2.1. Background

The male produces spermatozoa in the testes, stores them in tubular pouches called epididymides, and excretes them upon ejaculation through ducts, called vas deferens. Under normal circumstances, most males will produce gametes (sperm) throughout their adult life. When men cannot produce sperm in a semen specimen (azoospermia), medical personnel classify this physiological problem as "obstructive." Thus, obstructive azoospermia indicates that ducts that carry sperm from epididymides to the exterior are blocked or missing. These ducts are ones that are cut during a vasectomy and sometimes cannot be successfully reattached, especially as the patient extends the time from vasectomy to reversal. Furthermore, a surgeon can damage these ducts during surgical repair (of a hernia) or infection can damage these ducts. Furthermore, a small percentage of men are born without vas deferens.

When a male has obstructive azoospermia, surgeons can aspirate spermatozoa from epididymides while the patient is under general or local anesthesia. Here the surgeon extrudes a testis from the scrotum that normally houses the testes, exposes an epididymis, and aspirates the spermatozoa from it. To fertilize the partner's oocytes with the aspirated sperm, the embryologist mixes the sperm with culture medium and uses the sperm to perform intracytoplasmic sperm injection (ICSI; see Intracytoplasmic Sperm Injection). Medical personnel call this technique microsurgical epididymal sperm aspiration (MESA).

Physicians also can extract sperm from the epididymis in a "nonsurgical' method they call percutaneous (through the skin) epididymal sperm aspiration (PESA). In this technique, the physician stabilizes the epididymis between thumb and forefinger, inserts a needle into the epididymis, and aspirates fluid from the epididymis. To obtain sufficient spermatozoa for the fertilization procedure, the physician may have to repeat PESA several times.

If medical personnel cannot attribute azoospermia to obstruction because sperm are not present in the epididymides, then the testes have failed to produce spermatozoa. To obtain potential spermatozoa in this case, the physician has to invade the testes with surgical or non-surgical procedures. In the surgical procedure, the patient undergoes anesthesia and the physician extrudes the testis from the scrotum. To determine if spermatozoa are present, the physician removes a small piece of testicular tissue and passes it to an andrologist for evaluation under light microscopy. However, if the andrologist does not detect sperm, the physician will need to take another biopsy. Medical personnel call this procedure testicular sperm extraction (TESE). The embryologist uses the sperm obtained from TESE to perform ICSI with the partner's oocytes (see Section 2.6).

Just as physicians can use PESA, a nonsurgical procedure, to recover spermatozoa from the epididymis, they can use percutaneous fine needle aspiration (FNA) to obtain testicular spermatozoa. A single-needle puncture into the testis followed by suction may be sufficient to obtain spermatozoa. The physician performs this procedure until the embryologist has sufficient sperm to perform ICSI. Percutaneous fine needle aspiration has, however, a much lower recovery rate for spermatozoa than does TESE.

1.2.2. Medical Indications

Men that do not ejaculate sperm (azoospermia) are candidates for this procedure.

1.2.3. Issues

1. **Con:** Testicular deterioration can be an issue after TESE.
Pro: If the partner's sperm is a genetic requirement, sperm aspiration is the only way some couples can bear children. To deny couples the opportunity to have children is to deny them of their procreative rights.

2. **Con:** MESA requires microsurgical expertise, general anesthesia, and postoperative discomfort.
Pro: When the patient has azoospermia, MESA and TESA are the best methods to obtain spermatozoa.
3. **Con:** The same issues that apply to ICSI apply to sperm aspiration because Assisted Reproductive Technology Specialists use ICSI to initiate fertilization.
Pro: See ICSI for this side of the issue.

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Biographical Sketch

William R. "Bill" Boone earned a Ph.D. degree from Clemson University. He established the Assisted Reproductive Technology Laboratory for the Department of Obstetrics and Gynecology, at the Greenville Hospital System, in Greenville, South Carolina. He is certified as a High-Complexity Laboratory Director and holds three full professorships (Clemson University, The Medical University of South Carolina, and the University of South Carolina School of Medicine). He has written two books and has over 50 peer-reviewed journal articles to his credit as well as more than 100 scientific abstracts.