Cutting-Edge Evaluation of Male Infertility

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KEYWORDS

• Male infertility • Semen analysis • Physical examination • Genetic testing • Epigenetics

KEY POINTS

- A male factor contributes to 50% of cases of infertility, yet only 7.5% of men are referred for urologic evaluation.
- The male infertility evaluation is critical to identify the cause of infertility but may also reveal other information relevant to the health of the patient and his offspring.
- The history, physical, and semen analysis remain the mainstay of the male infertility evaluation. Additional hormonal and genetic testing may be indicated.

INTRODUCTION

Infertility affects up to 15% of the world's population, with approximately half involving a male factor.[1,2] The inability to conceive can have impacts on patients' self-esteem, mental health, financial status, and even their marriages. Beyond the direct effects of infertility however, there are indirect associations of infertility with a man's health that highlight the importance of a fertility evaluation for every man. Factors contributing to infertility can range from the easily correctible (eg, changing timing of intercourse) to the currently irreversible. For example, up to 10% of the infertile couples with male factor infertility may have reversible causes such as varicoceles or obstruction.[3] Consequently, any investigation into a couple's infertility should thoroughly evaluate the male partner.

Traditionally, infertility has been defined as the inability of a man and woman to conceive after 12 months of unprotected intercourse.[4] An infertility evaluation is recommended after 6 months, however, if the woman is older than 35 years.[5] Couples are increasingly delaying attempts at conceiving until after career development, resulting in progressively later attempts at pregnancy, and apprehension surrounding their potential fertility often prompts requests for earlier workups. Although no laboratory test can guarantee that a couple is fertile, prompt evaluation can identify problems that can guide their planning.

Spermatogenesis and fertilization are complex processes involving a combination of genetic, hormonal, environmental, and other factors, and failure of any of these can result in infertility. Because of this, the goals of a thorough infertility workup are manifold. The primary goals are to identify the etiology of the infertility, and determine whether it is reversible and whether there are contributing factors that may impact the patient's overall health. Timely addressing a reversible etiology such as gonadotoxic exposure or an underlying medical condition can often result in rapid improvement of fertility.[6,7] If an irreversible etiology is uncovered, the physician should determine whether the condition is amenable to assisted reproductive technologies (ARTs) (eg, intrauterine insemination [IUI] or in vitro fertilization [IVF] with intracytoplasmic sperm injection [ICSI]). If not,

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then the patient may need to consider adoption or use of donor sperm. It is also important to identify genetic etiologies that may be passed on to the patient's offspring, warranting referral to genetic counseling.

The increasing prevalence and availability of IVF and other ARTs have greatly enhanced the ability of couples to have children. As only relatively few sperm are needed with ICSI, however, the male infertility evaluation is often skipped if there are sufficient sperm in the ejaculate for ART. Thus, fewer than 40% of subfertile men undergo evaluation.[8] This is particularly concerning, however, as male infertility may be associated with other underlying disease states. For example, infertile men have a higher risk of cancer, immune problems, cardiovascular disease, and overall mortality.[9–11] Without an infertility workup, these potentially life-threatening conditions for which infertility is a symptom may go unnoticed and the opportunity for early diagnosis missed. Thus, even if couples are planning ART due to a known female factor, the male partner also should undergo a comprehensive evaluation if semen quality is impaired.

Despite a thorough investigation, a male etiology will escape a specific diagnosis in 15% to 30% of cases.[12] Recent advances in the diagnostic workup for male factor infertility have increased our understanding and ability to diagnose contributing factors. The goal of this article is to provide an up-to-date guide to the diagnostic workup of the infertile man and highlight advances in the field that may greatly expand our ability to diagnose and treat these men in the future.

BASIC EVALUATION

A thorough medical history, physical examination, and at least 2 semen analyses are the cornerstone of any evaluation of the infertile man.[4] These provide information that can guide treatment and further evaluation.

History and Physical Examination

The workup for infertility begins with the history and physical. Emphasis should be placed on interviewing the couple. A comprehensive approach to history taking involves inquiring about reproductive, sexual, medical, surgical, infectious disease, childhood conditions, and gonadal toxin exposure history. Stress also should be discussed, as it may contribute to impaired fertility and sexual dysfunction.[13,14] Discussion should focus on present attempts at conceiving, timing of intercourse, and use of lubricants, as well as the menstrual history and previous evaluation of the female partner. Whether either partner has previously caused/been pregnant also should be noted. Essential components of the history are listed in **Table 1**.

Family history is an increasingly important part of the patient's history given the genetic basis of infertility. The X chromosome has many genes critical to spermatogenesis. As men normally have a single copy of the X chromosome, any mutation in these genes can affect male fertility, as there is no second chromosome to compensate,[15] whereas female individuals with a mutated copy may still be fertile. Thus, men should be asked about family history of infertility, particularly in brothers and maternal uncles, as this may indicate

Table 1
History components of the male infertility evaluation

Category	Components
Reproductive history	Past attempts at conceiving Previous treatments for infertility Birth control Sexual technique and lubricants Timing of intercourse Previous pregnancies Menstrual history and female evaluation
Sexual history	Erectile dysfunction Hypogonadism Ejaculatory dysfunction Sexually transmitted disease
Medical history	Fevers or systemic illness Diabetes Spinal cord injury
Gonadotoxins	Drugs (exogenous anabolic steroids, tobacco/nicotine exposure, alcohol, narcotics, marijuana, immunosuppressants, chemotherapy) Radiation exposure Pesticides Thermal exposure Testosterone or steroids
Surgical history	Hernia repair Scrotal trauma Testicular torsion Varicocele repair Orchiopexy Transurethral resection of prostate
Family history	Infertility Genetic disorders Consanguinity

an X-linked genetic transmission. Given the elevated risk of cancers among infertile men,[10,16] history of cancers in first-degree and second-degree relatives should also be identified. Other significant illnesses in family members (eg, cystic fibrosis) also should be noted.

Physical examination should begin with gross assessment of general appearance, body habitus, and secondary sexual characteristics. Findings such as gynecomastia or gynecoid hair distribution may be indicative of underlying endocrine or genetic abnormalities. Likewise, obesity can be associated with lower testosterone and an abnormal testosterone-to-estradiol ratio.[17] Penile anatomy, such as penile curvature or plaques and location of the urethral meatus also should be inspected, as these abnormalities can impair sexual intercourse or result in an inability for ejaculate to reach the cervix, respectively. A careful examination of the scrotal contents and inguinal region should be done. Close attention should be paid to abnormal testis volume or consistency, the presence of varicoceles, and, specifically, the presence of both vasa deferentia and epididymides.

A physical examination is critically important, as findings may identify or rule out potential etiologies of infertility. Most testicular volume is composed of the seminiferous tubules, thus, abnormal testes size and consistency suggest impaired spermatogenesis, and may indicate androgen deficiency. Absence of one or both vasa should raise concern for cystic fibrosis (further discussed in "Radiological Examination" and "Cystic Fibrosis Gene Mutations"), and induration of the epididymides in the presence of normal-sized testicles is suggestive of obstruction.[18] Pertinent findings on physical examination can be found in **Table 2**. A digital rectal examination (DRE) also should be considered to examine the prostate and to check for midline cysts or enlarged seminal vesicles. Any abnormal findings on DRE should prompt a transrectal ultrasound (TRUS).

Table 2
Physical examination components of the male infertility evaluation

Category	Findings
General	Body habitus Gynecomastia Gynecoid features
Penis	Meatus location (hypospadias or epispadias) Curvature (chordee/Peyronie disease) Ulceration (venereal disease)
Testes	Size (endocrine disorder) Consistency Contours and masses (malignancy)
Epididymides	Cysts Spermatocele
Vasa deferentia	Atresia or agenesis (cystic fibrosis) Granuloma
Spermatic cords	Asymmetry Varicocele
Rectal examination	Midline cysts Dilated seminal vesicles Enlarged prostate

Semen Analysis

The semen analysis it the key laboratory test in the evaluation of the infertile man. Collection of the semen should be done after the patient has been abstinent for 2 to 5 days, as sperm concentration, volume, and motility may be affected by shorter and longer abstinence periods. Semen samples are usually collected by masturbation in the clinic; however, if masturbation is not possible for religious or other reasons, the patient may use specialized seminal collection condoms. Samples collected at home should be kept at room or body temperature and brought to the clinic within 1 hour. At least 2 analyses are recommended, as there is often variation between different analyses of the same individual.[19] When there are highly divergent analyses, a third sample is required to determine the baseline for that individual. **Table 3** contains the reference values based on data from the World Health Organization.

It should be noted that the reference values are statistically determined and do not reflect "normal" values. The values are based on the 95th percentile of parameters of men with proven fertility.[19] Thus, 5% of the fertile population would be expected to fall below the lower reference limit. Semen parameters of infertile men overlap considerably with those of fertile men. In addition, "normal" semen analyses are found in more than 40% of couples undergoing fertility evaluation,[20] suggesting although sperm are necessary for fertilization, the presence of sperm does not guarantee fertility. Likewise, low semen parameters generally do not guarantee infertility.

Semen volume abnormalities, such as aspermia (total absence of semen) or seminal hypovolemia (<1.0 mL), may point to specific anatomic factors as a cause of infertility. These findings may be the result of functional issues, such as retrograde ejaculation, or anatomic variations, such as

Table 3
World Health Organization semen analysis reference ranges (5th edition)

Parameter	Lower Reference Limit (95% Confidence Interval)
Semen volume, mL	1.5 (1.4–1.7)
Total sperm number, 10^6/ejaculate	39 (33–46)
Sperm concentration, 10^6/mL	15 (12–16)
Total motility, %	40 (38–42)
Progressive motility, %	32 (31–34)
Sperm morphology, normal forms, %	4 (3.0–4.0)

Data from World Health Organization. WHO laboratory manual for the examination and processing of human semen. 5th ed. Geneva: World Health Organization; 2010.

ejaculatory ductal obstruction or hypoplasia of the prostate or seminal vesicles due to congenital bilateral absence of the vasa deferentia (CBAVD) or androgen deficiency, respectively. If the vasa are palpable bilaterally, a postejaculatory urinalysis should be obtained to determine whether retrograde ejaculation is present. TRUS can be used to visualize the seminal vesicles and prostate to determine whether ejaculatory duct obstruction (dilated seminal vesicles), hypoplastic seminal vesicles (seen in CBAVD), or other structural abnormalities may be causing obstruction (eg, prostatic cysts).

Sperm concentrations less than 15 million/mL define oligozoospermia.[21] Absence of sperm from the ejaculate is azoospermia; however, this can be diagnosed only if the semen sample has been centrifuged and the pellet found to lack sperm.[19] Sperm concentrations less than 10 million/mL should prompt endocrine testing, whereas concentrations less than 5 million/mL should prompt genetic testing.[4] An increased sensitivity in genetic testing is found when one limits testing to less than 1 million, but this may increase missed positive findings.

Asthenozoospermia, or impaired sperm motility, is another potential hindrance to fertility, as sperm progression is requisite for natural fertilization. The 3 categories of motility are progressive (comprising all sperm moving in a linear or circular pattern), nonprogressive, and immotile; the latter 2 consisting of all sperm that do not progress. In some conditions (eg, Kartagener syndrome), the sperm may be uniformly immotile. Vitality testing can be used to differentiate alive but immotile sperm from necrozoospermia (ie, all dead sperm).[19] The total motile sperm count (concentration * volume * percent motility) is often used clinically; this calculated result is most useful to determine what degree of assisted reproduction may be needed.

Sperm morphology refers to the shape of the spermatozoa, with the lower reference limit in fertile men being 4% normal forms.[19] Thus, even in fertile men, the vast majority of sperm have abnormal morphology. The definition of normal morphology has become progressively stricter in each of the 5 editions of the World Health Organization guidelines. Meta-analyses have demonstrated, however, that abnormal sperm morphology using the current guidelines does not predict IUI, IVF, or ICSI success,[22,23] and thus the true significance of this number has been called into question. Indeed, Kovac and colleagues[24] demonstrated that among 24 men with severe teratozoospermia (ie, 0% normal morphology), 25% were subsequently able to conceive naturally.

Zero percent normal forms should not be confused, however, with 100% of sperm showing the same abnormal morphology. These diseases, such as globozoospermia, are usually associated with genetic abnormalities. Most of these patients have extremely low success rates of natural and assisted conception, although some may be amenable to modifications of the ICSI procedures (eg, ICSI success rates are improved with oocyte activation in globozoospermia due to *DPY19L2* mutations[25]). Others, such as macrocephalic sperm, have high rates of aneuploidy.[26] As genetic testing for these conditions is not routine, men with these types of abnormal morphology should be referred for genetic counseling.

Testosterone and Follicle-Stimulating Hormone

Approximately 3% of cases of male infertility are attributable to endocrine problems.[27] It is recommended that testosterone and follicle-stimulating hormone levels be measured as part of an endocrine evaluation in men with sperm counts of less than 10 million/mL or if there are features on physical examination suggestive of endocrine dysfunction.[4] The endocrine evaluation of infertile men is covered in more detail in Sarah C. McGriff and colleagues' article, "Optimal Endocrine Evaluation and Treatment of Male Infertility," elsewhere in this issue. Based on the physical examination, semen analyses, and endocrine testing, further testing may be indicated.

EXTENDED EVALUATION

Radiological Examination

Scrotal, transrectal, and renal ultrasonography are generally not part of the initial evaluation of male infertility, but may be useful adjuncts to better delineate anatomy and identify potential etiologies.

Although varicocele is typically a clinical diagnosis, scrotal ultrasound can be used to objectively measure varicocele vein diameters and document reversal of blood flow with Valsalva.[28] Ultrasonography can be particularly useful in individuals whose body habitus makes physical examination difficult or in individuals with a history of varicocele repair; however, ultrasound is not necessary in most situations. Scrotal ultrasound should be performed if there is a testicular mass or if hydroceles, scarring, or other factors making direct palpation of the testicles difficult on physical examination. A recent publication, however, calls into question the suitability of surgery for subclinical varicoceles, showing the same increase in total motile count when comparing results of clinical and subclinical varicocele repair results.[29]

Transrectal ultrasound (TRUS) is used to evaluate abnormal DRE findings or to assess in the diagnosis of ejaculatory duct obstruction in patients with low semen volume.[30] TRUS enables visualization of enlarged seminal vesicles or cysts at the ejaculatory ducts, which may be the source of anejaculation, hematospermia, or painful ejaculation. Seminal vesicle aspiration is often used concurrently. In cases of CBAVD, TRUS also may be used to assess hypoplasia or agenesis of the seminal vesicles.[3] The vasa deferentia can be identified on TRUS, thus this can also be used if their presence is unclear on physical examination.

Renal and urinary tract ultrasonography are used less frequently, but they are indicated in cases of CBAVD and unilateral absence of the vas deferens to rule out unilateral renal agenesis, which is found in 10% and 25% of these patients, respectively.[31]

MRI of the pituitary fossa is indicated in men found to have elevated prolactin levels or unexplained hypogonadotropic hypogonadism during endocrine evaluation to rule out pituitary adenoma (see Sarah C. McGriff and colleagues' article, "Optimal Endocrine Evaluation and Treatment of Male Infertility," elsewhere in this issue). Less commonly, pelvic MRI can be used to identify the internal accessory organs; however, TRUS is usually sufficient and less expensive.

Laboratory Assessment

Depending on the sperm concentration or suspected etiology, genetic testing or sperm integrity testing (eg, seminal oxidant levels, DNA fragmentation, fluorescence in situ hybridization (FISH)) may be indicated.

Karyotype testing

A karyotype is recommended in men who have a sperm concentration less than 5 million/mL and should be considered if there is suspicion for numerical chromosome abnormalities (eg, Klinefelter syndrome) or large structural abnormalities (eg, translocations, deletions).[4] Chromosomal abnormalities are identified more frequently as sperm concentrations decrease: karyotypic abnormalities are found in fewer than 1% of men with normal sperm parameters, ~5% of men with severe oligospermia, and 10% to 15% of men with azoospermia.[32,33] Kleinfelter syndrome (XXY) is the most common chromosomal abnormality associated with male infertility.

Balanced translocations can result in phenotypically normal men; however, failure of meiotic pairing of the chromosomes can result in decreased sperm concentrations and sperm with imbalanced translocations. Thus, the male partner in couples with recurrent pregnancy loss should also have a karyotype performed.[4] As chromosomal abnormalities can potentially be passed on to offspring, men with chromosomal abnormalities should be referred for genetic counseling before consideration of ART.

Y chromosome microdeletion testing

Y chromosome microdeletion (YCMD) testing is indicated in men who have sperm concentrations less than 5 million/mL. Seven percent of men with impaired spermatogenesis have microdeletions of regions of their Y chromosome compared with 2% of normozoospermic men.[34] YCMDs are classified by regions called azoospermia factor regions (AZFa, AZFb, and AZFc). Depending on the region of the microdeletion, the outcome can range from moderate impairment of spermatogenesis to complete azoospermia. These regions are too small to be detected with karyotype testing and require polymerase chain reaction amplification of sites within each region. Complete AZFa and/or AZFb deletions are incompatible with spermatogenesis, and men with these deletions should not undergo attempts at testicular sperm extraction.[35] Spermatogenesis can occur in men with AZFc deletions, however, and azoospermic men with these deletions have approximately 50% likelihood of having sperm on microsurgical testicular sperm extraction.[36] YCMDs affect the Y chromosome and will be passed on to 100% of their male offspring, so men with YCMDs should meet with a genetic counselor before pursuing ART.

Cystic fibrosis gene mutations

Cystic fibrosis is due to mutations in the *CFTR* gene on chromosome 7. CBAVD is present in all men with clinical symptoms of cystic fibrosis; however, approximately 80% of men with CBAVD have mutations in the *CFTR* gene even in the absence of respiratory manifestations of the disease.[37] Thus, patients with CBAVD should undergo *CFTR* gene testing regardless of whether they have pulmonary manifestations of cystic fibrosis. Complete *CFTR* gene sequencing should be considered in all ethnic minorities with CBAVD given higher rates of less common variants. Congenital unilateral absence of the vas deferens (CUAVD) is variably associated with mutations in the *CFTR* gene.[37] Individuals with CUAVD should undergo renal ultrasound due to a high percentage having renal agenesis on the ipsilateral side, yet not necessarily associated with *CFTR* mutations.[31] Patients with these mutations should receive genetic counseling, and their partner should be tested given the relatively high prevalence of *CFTR* mutation carriers in the general population.

Seminal oxidants

Reactive oxygen species (ROS) are naturally produced by oxidative reactions. ROS play a critical role in the sperm acrosomal reaction; however, polyunsaturated fatty acids in the sperm membrane are particularly susceptible to oxidation by ROS, resulting in impaired sperm motility and DNA damage.[38–40] Elevated ROS levels are found in men with a variety of impaired semen parameters and may be a contributing factor in 25% to 40% of men.[41–43] ROS testing can be challenging, however, as not all laboratories have the necessary equipment. In addition, specimens must be tested shortly after ejaculation, as antioxidants in the seminal plasma may quench the ROS.

DNA fragmentation

Sperm lack mechanisms for DNA repair and, therefore, accumulate DNA damage as they pass through the reproductive tract. Although there is robust DNA repair on fertilization, excess DNA damage can prevent embryo development. Direct assays of DNA fragmentation measure the number of breaks in DNA, whereas indirect assays measure the sensitivity of DNA to acid-induced denaturation.[4] Higher levels of DNA fragmentation are seen as sperm counts decrease and are associated with IVF failure.[44,45] Thus, DNA fragmentation testing should be considered in individuals planning ART and those who have had unexplained recurrent pregnancy loss. Some studies have shown superior DNA quality in testicular sperm compared with ejaculated sperm.[46]

Fluorescence in situ hybridization

Sperm are normally haploid, containing a single copy of the 22 autosomes and an X or Y chromosome. Errors in meiotic segregation, however, can result in aneuploid sperm. As many as 6% of infertile men have elevated levels of aneuploid sperm.[47] Depending on the chromosome, sperm aneuploidy can result in viable embryos (eg, Klinefelter syndrome, Down syndrome, Turner syndrome); however, gain or loss of most chromosomes are incompatible with life. Thus, men with a high number of aneuploid sperm are at risk for recurrent pregnancy loss or fetal abnormalities. Sperm FISH can be used to identify the percentage of aneuploid sperm; however, there are currently few centers offering this testing.

Advanced Testing/Future Directions

Our understanding of male fertility continues to evolve at a rapid rate, and now includes important roles for genetics, epigenetics, metabolomics, and extracellular vesicle function. Each new discovery, however, highlights how much we have yet to understand and discover. Although not currently part of the recommended testing, diagnostic and therapeutic advances in these fields may bring them to the forefront of the male infertility evaluation in the upcoming years.

Additional Genetic Testing

Testing for *CFTR* mutations in men with CBAVD is currently the only specific gene recommended by guidelines. This traditionally has been due to the cost of sequencing and the relative infrequency of specific mutations in the general infertile population. Nonetheless, men with specific phenotypes could benefit from additional genetic testing and referral to a genetic counselor should be considered. These potentially significant genes include *ADGRG2* testing in men with *CFTR* mutation-negative CBAVD[48]; *DPY19L2*, *PICK1*, or *SPATA16* testing in men with globozoospermia[49–51]; or *AURKC* in men with macrocephalic sperm with multiple flagella.[52] Easily identifiable characteristics such as these may increase the yield of genetic testing in specific populations.

Alternatively, the rapidly decreasing cost and increasing throughput of next generation sequencing technologies has allowed for sequencing of entire panels of genes, and even whole exome and whole genome sequencing, well below the traditional cost of sequencing a single gene. For example, a targeted panel sequencing 87 genes previously associated with male and female infertility cost only $599

and had nearly 100% accuracy for detecting mutations and sex chromosome aneuploidies and 94% accuracy for YCMD.[53] These targeted sequencing technologies are also amenable to benchtop sequencers, facilitating integration into the andrology laboratory of the future. Thus, targeted or more extensive sequencing may ultimately become part of standard infertility testing.

Epigenetics

DNA modifications such as methylation (the most common type of epigenetic modification) can silence gene expression without altering the fundamental genetic sequence. Indeed, epigenetic modifications allow cell type–specific gene expression despite all cells sharing the same genetic code. Thus, abnormal methylation could silence genes in a spermatogonial stem cell or other germ cell, resulting in infertility. Abnormal epigenetic modifications, or epimutations, are harder to detect than genetic mutations, as they may be cell type specific. To test for epigenetics, DNA from the target tissue (eg, testes) is needed, limiting the usefulness of current testing. Sperm DNA methylation has been studied, and global methylation has been shown to increase with age and is altered in male infertility.[54,55] As there is high heterogeneity of sperm DNA methylation within a single sample, however, the clinical utility of sperm methylation testing remains unclear.[56] Nonetheless, as epimutations are potentially reversible, further research may allow for identification of epimutation-driven male infertility and targeted treatment to reverse it.

Metabolomics, Proteomics, Lipidomics, and Other "-omics"

Genetic mutations and epimutations can affect gene expression and protein function, ultimately altering the production of metabolites and other factors necessary for spermatogenesis and fertility. Metabolomics, on the other hand, focuses on the concentrations of the metabolites within a sample to identify factors that may be associated with disease, and secondarily assesses the pathways that may be contributing to the abnormal concentration. Similarly, proteomics can identify protein concentrations, lipidomics, lipid concentrations, and so forth. This can be particularly advantageous, as there may be many pathways converging and diverging from a specific metabolite or other factor, and the ultimate concentration of one or more substances may contribute more to infertility than a specific pathway. Thus, these studies often identify molecular signatures of disease that can subsequently be developed into biomarkers for diagnostic purposes.

As metabolomics looks at metabolite concentrations, it is naturally inclined to look at the production and consumption of metabolites, the most common of which are often involved in energy production. Thus, metabolomic analysis in male infertility has often focused on men with asthenozoospermia, as altered energy production can contribute to decreased motility.[57] One challenge with this approach, however, is that although molecular signatures can be identified for asthenozoospermia, there is limited clinical utility for that information. For example, a molecular signature was identified in the seminal plasma of men with asthenozoospermia compared with controls, and when converted to an algorithm, the signature accurately predicted the motility of 5 of 6 subjects.[58] Standard semen analysis, however, is sufficient to classify asthenozoospermia from normozoospermia.

Other similar types of studies have highlighted the challenges in trying to use metabolomic and other molecular concentrations as a basis to treat disease. Altered concentrations of a substrate may be the cause of infertility; however, it may also just be a by-product of some other reaction. Thus, correcting the abnormal signature may not improve fertility. For example, lipidomics have identified that seminal plasma and sperm docosahexaenoic acid (DHA) levels decrease with worsening semen parameters.[59] A double-blind, placebo-controlled trial of DHA supplementation in men with infertility failed to improve motility or count in asthenozoospermic men.[60] Thus, whether the low DHA is impairing motility, or whether it is simply a by-product of another process remains unclear. Nonetheless, as molecular signatures for infertility become better defined, these may be amenable to targeted interventions in the future.

Extracellular Vesicles

Exosomes and other extracellular vesicles are secreted by cells and can transmit RNAs, proteins, metabolites, and other substances. Extracellular vesicles produced by the epididymis (also known as, epididymosomes) have been shown to play an important role in sperm maturation by delivering protein cargos to the sperm as they transit the epididymis.[61] Exosomes in the seminal plasma have been shown to affect sperm motility.[62] Exosomes and other extracellular vesicles are found throughout the male and female reproductive tract. We are just beginning to understand the critical and complex role they play in male fertility.[63]

SUMMARY

The proper evaluation of the male is a critical component of the evaluation of the infertile couple. Despite advances in ART that permit paternity with a limited number of sperm, the purpose of the examination of the infertile man goes beyond identifying the cause of infertility and may identify factors that could affect the health of the patient and/or his offspring. Although the fundamental components of the evaluation have remained constant, advances in our understanding of the pathophysiology, combined with advances in technology, have enhanced our ability to diagnose and treat male infertility.

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