REVIEW ARTICLE

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THE ROLE OF CLINICAL PATHOLOGISTS IN THE MANAGEMENT OF MALE INFERTILITY

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Male infertility is receiving increasing attention in Africa as up to 50% of cases of infertility are ascribed to it. In the management of this condition, the clinical laboratory plays a crucial role especially in the proper identification of causes of infertility. The role of the pathologists in this respect stems from the choice of laboratory equipment, reagents, type of samples needed, proper sample collection and its preparation. Added to these functions, the pathologist should help in the proper selection of required tests so as to ensure optimum diagnosis and treatment efficiency. Semen analysis is the first test required in the laboratory assessment of the infertile male. The obtained spermogram serves as the pivot for further tests which include hormonal assays, tissue studies and karyotyping. There is a disturbing high prevalence of azoospermia among Nigerians. The expected laboratory evaluation and treatment of such patients and others with abnormal spermogram are discussed in this review.

Key words: Male infertility, review.

INTRODUCTION

Infertility is a worldwide problem (1) that is attracting the attention of various researchers (2), even in Africa (3). Before now, men who were sexually potent were also thought to be fertile. This erroneous position has been discarded as male factors have been shown to be responsible for between 30-50% of cases of infertility (4, 5). Little wonder that 1% to 30% of children cannot be genetically matched to their presumptive fathers (6).

It has been suggested that the fertility prowess of fertile men decreases with time, since semen quality has been shown to deteriorate by as much as 3% per year (2). This probably contributes to the observation that the prevalence of male infertility in Nigeria is on the increase (7).

Generally, laboratories contribute to the actual diagnosis of infertility in over 50% of couple's investigated (8). It is very important that the right diagnosis is made early, as this is a crucial factor in the management of male infertility (9). In our environment, the use of empirical treatment by doctors, in the management of infertility has yielded dismal results (10). The import of this is that the clinical pathologists' role in the management of male infertility is enormous.

PREANALYTICAL CONSIDERATION

In order to ensure quality laboratory services in male infertility management, the pathologist should be involved in patient preparation before laboratory analyses. This is in addition to his role in ensuring provision of adequate laboratory reagents and equipment, technical expertise and comfortable environment for the patients.

The pathologist should ensure that reference ranges, conditions of sample collection, sample transportation and specimen preservation, are in keeping with proper clinical applications. For example, the serum hormonal kits should be selected considering the coefficient of variation which should be 10% or less (11).

The kind of sample and frequency of sampling should be stated. Unlike in females, there is no cyclical secretion of luteinising hormone (LH), follicle stimulating hormone (FSH); prolactin (PRL) and testosterone in males, hence a single blood sample suffices for hormonal investigation of infertile males (12). In semen analysis of such men, more than one sample, collected at least one week apart after 3-5 days of sexual abstinence, is required to establish abnormal spermogram.

Sample preparation is important in the laboratory evaluation of these patients. While semen sample should be analyzed within an hour of collection, testicular biopsy specimen can be preserved. Such biopsy specimens are better kept in Zenker's solution or Bouin's fixative (13). In taking such materials, an open biopsy is more satisfactory than a punch biopsy (14). For hormonal assays, it is important that the samples are not kept for too long, and they should be devoid of gross haemolysis and lipaemia. In places where saliva is used for steroid estimation, the relevant analytical and interpretive expertise must be available (15).

ANALYTICAL/POSTANALYTICAL ROLES

When a representative sample is properly taken and well processed, the clinical pathologist will then interpret the findings of laboratory analyses. Furthermore, he/she advices the clinicians on further tests that might be of benefit to the management of such patients. The first test that should be done for suspected infertile male is semen analysis. The observed spermogram not only give a useful clue to the fertility status, it also could be a pointer to an obstructive disorder in the male.

Semen analysis parameters are commonly based on the WHO guidelines (16). The parameters assessed are appearance, consistency (viscosity), volume, and pH. These are followed by microscopic examination for sperm motility, morphology, concentration, and presence of cells other than spermatozoa (16, 17). In some places, computer assisted semen analysis (CASA) is readily available. Some of the CASA systems are coupled with video technology and sophisticated microcomputers to allow for automatic image digitalization and processing. This technology is thought to be more objective in measurement of seminal parameters than the subjective measurement of the standard traditional semen analysis. It also permits the added measurement of linearity, curvilinear velocity, straight line velocity and flagellar beat frequency of spermatozoa (18). Other seminal fluid analyses include sperm sperm-cervical mucus interaction, sperm penetration assay, acrosome evaluation and hypo-osmotic swelling test (18).

WHO defines normal ejaculate as sperm concentration of $\geq 20 \times 10^6$ spermatozoa/ml, with $\geq$ 50% with forward progressive motility or $\geq$ 25% with rapid progressive motility in 60 minutes of ejaculate and with $\geq$ 30% with normal morphology (16) (Table 1). But ideally, each

laboratory should set its own normal values, reflecting the specific population analyzed. For our own environment, the values commonly applied are shown in the work of Nkposong (19, 20) (Table 2). When the result of such analyses deviates considerably from those of a large population of tested men, then sub-fertility is likely (17).

Table 1: Normal values of semen variables (WHO, 1992) (16)

Standard tests

Volume:	2.0 ml or more
pH:	7.2 – 8.0
Sperm concentration:	20 X 10^6 spermatozoa/ml or more
Total sperm count:	40 X 10^6 spermatozoa/ejaculate or more
Motility:	50% or more with forward progression or 25% or more with rapid progression, within 60 minutes of ejaculation
Morphology:	30% or more of normal forms
Vitality:	75% or more live i.e. excluding dye
White blood cells:	Fewer than 1 X 10^6/ml
Immunobead test:	Fewer than 20% spermatozoa with adherent particles
MAR test:	Fewer than 10% spermatozoa with adherent particles

Optional tests

Alpha-Glucosidase (Neutral):	20 mU or more per ejaculate
Zinc (Total):	2.4 µmol or more per ejaculate
Citric acid (Total)	52 µmol or more per ejaculate
Acid phosphatase (Total):	200 U or more per ejaculate
Fructose (Total):	13 µmol or more per ejaculate

Nomenclature of semen variables

Normozoospermia:	Normal ejaculates as defined above
Oligozoospermia:	Sperm concentration fewer than 20 X 10^6/ml
Asthenozoospermia:	Fewer than 50% spermatozoa with forward progression or fewer than 25% spermatozoa with rapid progression
Teratozoospermia:	Fewer than 30% spermatozoa with normal morphology
Oligoasthenotertozoospermia:	Disturbance of all the three variables (concentration, motility and morphology)
Azoospermia:	No spermatozoa in the ejaculate
Aspermia:	No ejaculate

Table 2: Seminal fluid analysis parameters (Nkposong, 1987) (20)

Minimal values compatible with fertility

Volume:	2-5 mls
Viscosity:	Fully liquefied within 1 hour
Sperm count:	20 million/ml
Motility:	60% (1st hour), 50% (2-3 hours)
Motility index:	3 – 4
Morphology:	60% normal forms
Vital staining:	30% dead spermatozoa
Polymorphs:	5% (No significant pyospermia)
Immature forms:	5%

In addition to semen analysis providing diagnostic/predictive values for *in vivo* fertility and conception, a number of studies (21, 22) that have examined the predictive values of the traditional semen characteristics have shown these parameters to also predict pregnancy outcome during artificial reproductive techniques (ART).

Semen analyses results usually set the pace for further investigations of the infertile male. If the spermogram is normal in all respects, no further investigation is necessary. But up to 50% of males of infertile unions have been shown to have abnormal spermogram in Nigeria (23). These include those with oligozoospermia and azoospermia. In a male infertility clinic in Ibadan, Nigeria (19), 35% of the patients

attended to, had azoospermia while 48.9% and 23.4% of patients attending a male infertility clinic in Ilorin, Nigeria (20) had oligozoospermia and azoospermia respectively.

Hormonal evaluation of male infertility usually follows the observation of oligozoospermia or azoospermia. These patients will benefit from serum FSH, LH, PRL and testosterone determinations (25, 26). The commonest hormonal disorder in infertile Nigerian males is hyperprolactinaemia (12, 26) and it is associated with oligozoospermia and azoospermia (27). Up to 25% of infertile males Nigeria have primary testicular failure, probably due to infectious diseases that affect and destroy the testes (12). The common findings in such patients are severe oligozoospermia and azoospermia, elevated levels of serum FSH, LH and low serum testosterone.

Azoospermia and some cases of severe oligozoospermia can be either due to failure of spermatogenesis or obstruction disorder; intratesticular, epididymal, vasal or ejaculatory duct (19, 28). Serum FSH, LH and testosterone measurements are required in suspected cases of testicular failure or obstructive disorder (8). Determination of serum FSH can distinguish between primary and secondary gonadal failure, and can also identify those with obstructive azoospermia. Serum levels of FSH, LH, PRL and testosterone are usually normal in obstructive disorder Truly, serum FSH determination has reduced the need for testicular biopsy to confirming normal spermatogenesis in cases of genital tract obstruction.

The histological findings in obstructive azoospermia include tubules with normal or slightly reduced diameter in the presence of all stages of spermatogenesis. Also, the normal orderly arrangement of testicular architecture is lost and the central lumen is absent. Half or more of the examined tubules must be so affected to make the diagnosis of obstructive azoospermia (29). Some of such patients suffer from Young's syndrome, in which the obstructive azoospermia and chronic sino-pulmonary infections co-exist (30). Testicular biopsy in non-obstructive azoospermia shows structural immaturity of the seminiferous tubules, decreased spermatogenesis, germ cell aplasia (Sertoli cell only syndrome), germ cell maturation arrest, peritubular and tubular fibrosis (30).

Another useful test that is advised in men with azoospermia and severe oligozoospermia is karyotyping, when chromosomal disorder is suspected (32). This is particularly important in the face of elevated FSH level and markedly reduced testicular volume. The most frequent chromosomal abnormalities in infertile males are sex chromosome aneuploidies; such as 47, XYY karyotypes, autosomal Robertsonian translocations and other types of translocation (33, 34).

Klinefelter's syndrome is the best known karyotypic abnormality and it is associated with a characteristic histologic appearance of the tissue (35). Even after puberty in Klinefelters', the histological findings could be that of no spermatogenesis or some degree of spermatogenesis. The finding of some amount of spermatogenesis should not militate against the diagnosis of Klinefelter's syndrome (36). Occasionally,

the classic histologic appearance of Klinefelter's syndrome occurs in the absence of karyotypic abnormalities (36).

Other congenital problems in infertile males include congenital bilateral absence of vas deferens (CBAVD) and congenital adrenal hyperplasia (CAH) (9). Patients with CBAVD will present as cases of obstructive azoospermia (37) while those with CAH have spermatogenic defect (9). Measurement of seminal alpha-glucosidase enzyme can serve as a non-invasive method of distinguishing obstructive from non-obstructive azoospermia (37). To help in making a diagnosis of CAH, serum level of 17-ketosteroids or dehydroepiandrosterone (DHEA) should be measured (9). Additional tests in these patients include seminal total antioxidant capacity (TAC) which is low in up to 40% of infertile males, due to increased generation of reactive oxygen species (38, 39).

ROLE IN TREATMENT

Effective treatment of male infertility begins with a careful history and physical examination. Specific childhood diseases such as cryptochordism, post-pubertal mump orchitis and testicular trauma/torsion should be sought. History of exposure to occupational and environmental toxins, excessive heat and radiation should be elicited. Drug history is equally important and such drugs as anabolic steroids, cimetidine and spironolactone, are known to affect reproductive cycle (18). Excessive alcohol consumption is associated with a decrease in sperm count and hormonal abnormalities (18). Past history of improperly treated sexually transmitted infections should be excluded in our environment.

Treatment could be medical or surgical or both. The role of clinical pathologists in the medical management of infertile males is prominent in those due to infections, either latent or ongoing. It is important that in such cases, the organisms associated with genital infections are accurately identified and treated with appropriate antimicrobial agents as well as anti-inflammatory drugs. Success of treatment can be monitored by periodic seminal fluid analysis.

However, more often than not, patients are seen at late stage when only commensal flora rather than genital pathogens are isolated (39). Nevertheless, attempt at identifying and treating any recent or latent infections should be done. In some cases, androgen may be administered to improve sperm count in those with mild to moderate oligozoospermia. Other medical treatment modalities include endocrine therapy for men with hypogonadotropic hypogonadism, immunosuppressive drugs (corticosteroids) for immunologic infertility and α-adrenergic stimulation using sympathomimetic drugs for those with retrograde ejaculation (18).

The commonly treatable surgical conditions associated with male infertility are undertaken by urologists. The commonest surgical procedure carried out on infertile male is varicocelectomy, because scrotal varicocoele is found in about 40% of infertile male (20). Surgical procedure can also be performed for cases of obstruction of the reproductive tracts when the spermatogenic potential of the testes is preserved, and pituitary adenoma can be removed by surgical ablation (18). The role of the clinical pathologists and the

laboratory in this regard include post-operative serial monitoring of patients for response to surgery. The concerned laboratory should ensure appropriate quality control measures so that subtle changes in seminal, endocrine and other laboratory indices are noted.

It should be emphasized that infertile males can benefit from assisted reproductive techniques (ART), to fertilize the ovum of their spouse. This is possible in those, in whom spermatozoa can be obtained by microsurgical epididymal sperm aspiration (MESA) following stimulation of spermatogenesis, or retrieval of spermatozoa from the urinary bladder in those with retrograde ejaculation (18). The clinical pathologist has a role to play in determining the semen parameters that are important in predicting pregnancy outcome or success rate of ART procedures in infertile males and their spouse.

THE NIGERIAN SITUATION

The high prevalence of azoospermia in Nigeria is worrisome. These are related possibly to infections (12, 20, 28, 40-44). With the emergence of new and increase in the number of antibiotic resistant venereal pathogens (43, 44, 45), complications from simple genital infection have increased with a consequent increase in the number of males with primary or secondary infertility (46, 47).

Hence, the quality of care offered at sexually transmitted diseases (STD) and infertility clinics need to be improved. Highly sensitive and specific newer diagnostic methods, such as the DNA probes and polymerase chain reaction (PCR) as well as serologic tests that can promptly detect acute and subclinical infections are now available (48) though not yet widely applicable in developing countries.

Prompt diagnosis and treatment of cases of STD is the backbone of prevention of male infertility in this environment. Contact tracing and treatment, community awareness campaign about the attendant sequelae of improperly treated STDs, health seeking behaviour and proper administration of the syndromic approach to STD management are valuable aspects of the control of STD related male infertility in developing countries.

Males who are infertile should also readily have hormonal evaluation especially serum PRL, FSH and LH, when they have abnormal spermogram. Clinical pathologists in our environment should as part of the assessment for laboratory investigation of infertile males, suspect common cause such as varicocoele which is associated with abnormal spermogram. This will require prompt referral to the urologist for appropriate treatment. Karyotypic studies are not readily available in most centres. This should be a matter for attention by pathologists in Nigeria.

CONCLUSION

In the management of male infertility, a team of clinicians and pathologists working closely together will enhance the achievement of the desired success in the diagnosis, treatment and prevention of this condition.

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