

In the name of God

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تست های سطح اول ناباروری مردان

Semen Analysis

Introduction

Semen analysis is the first test requested when fertility potential of a man becomes questionable.

The test provides information on the functional status of the seminiferous tubules, epididymis, seminal vesicles, and the prostate.

Thus the results of the test should be interpreted in the light of a full clinical history and physical examination .

limitations is the limited **reliability** of the result of semen analysis due to variations in the method and timing of obtaining ejaculates and the lack of standardization of the semen analysis methodology .

This in turn contributes to **the relatively low prognostic power** of semen analysis as a diagnostic test. When azoospermia cases are excluded, the results of semen analyses do not always correlate with pregnancy rates or infertility .

Interrelationship between semen characteristics and reproductive organelles

	Testis	Epididymis	Seminal vesicle	Prostate
<i>Semen</i>				
Coagulation			X	X
Liquefaction			X	X
Volume	X	X	X	X
Immunoglobulins		X?	X	X
Leukocytes		X?	X	X
Erythrocytes			X	X
<i>Spermatozoa</i>				
Count	X			
Motility	X	X	X	X
Morphology	X	X		

The Reliability of Semen Analysis Result

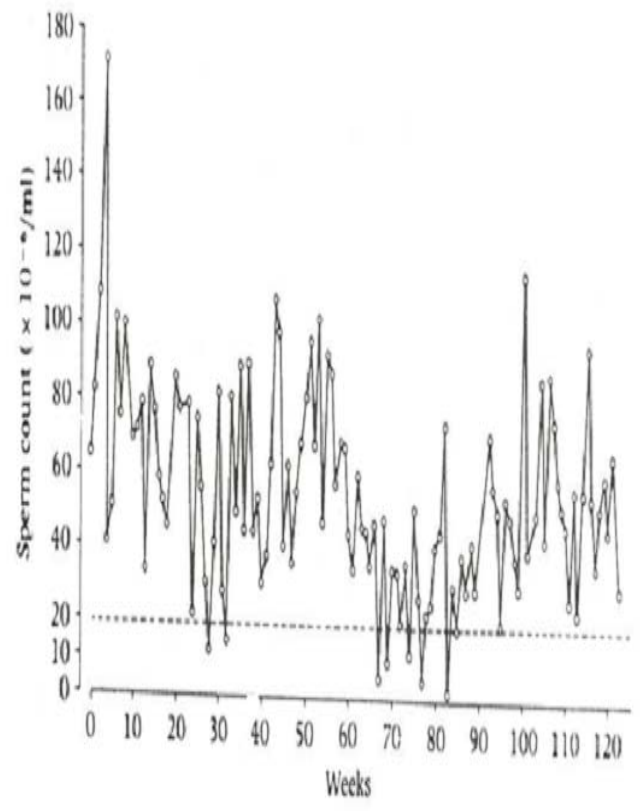
- Standardization of Semen Analysis Techniques
- Biological Variability(The completeness of sample collections-The impact of age-Testicular size-Duration of abstinence-The intensity of sexual stimulation-Delayed liquidation and viscosity-Reversible changes in sperm parameters)
- External Technical Factors that May Influence Semen Analysis Result

- Assessment of fertility
(2 samples; 7 days to 3 months - best 2 weeks)
- If results of 2-3 assessments differ greatly, additional samples must be analyzed.

- Name
- Period of abstinence - 2-7 days
- Time of collection + analysis recorded
- Entire ejaculate and not coitus interrupts in a wide mouth container
- Delivered within 1 hour of collection
- Avoid temperature extremes

Table A1.1 Lower reference limits (5th centiles and their 95% confidence intervals) for semen characteristics

Parameter	Lower reference limit
Semen volume (ml)	1.5 (1.4–1.7)
Total sperm number (10^6 per ejaculate)	39 (33–46)
Sperm concentration (10^6 per ml)	15 (12–16)
Total motility (PR + NP, %)	40 (38–42)
Progressive motility (PR, %)	32 (31–34)
Vitality (live spermatozoa, %)	58 (55–63)
Sperm morphology (normal forms, %)	4 (3.0–4.0)
<i>Other consensus threshold values</i>	
pH	≥ 7.2
Peroxidase-positive leukocytes (10^6 per ml)	< 1.0
MAR test (motile spermatozoa with bound particles, %)	< 50
Immunobead test (motile spermatozoa with bound beads, %)	< 50
Seminal zinc (μmol /ejaculate)	≥ 2.4
Seminal fructose (μmol /ejaculate)	≥ 13
Seminal neutral glucosidase (mU/ejaculate)	≥ 20



The Significance of Semen Volume

- The seminal vesicles 70% volume.
- The lower reference limit for semen volume is 1.5 ml.
- **Incomplete collection ,acquired obstruction** of the ejaculatory duct.
- **CBAVD**
- **Retrograde ejaculation** The sperm is found mostly dead due to the combined effects of osmotic stress, low pH, and urea toxicity The recovery of high-quality sperm after the induced modification of the urine composition and pH to facilitate its use in the intracytoplasmic sperm injection technique (ICSI) has been described .
- Occasionally, the orgasm is associated with a miniscule amount of ejaculate or no ejaculate at all (dry ejaculation, aspermia). This happens in a diversity of **neurological diseases and subsequent to surgical procedures on the lower urinary tract**

Volume

– Accessory sexual gland secretions

Aspermia (no semen)

Surgery

Retrograde ejaculation

Retrieve sperm from bladder and process for artificial insemination

Hyperspermia (More than 6 ml)

Long period of sexual abstinence

Overproduction of glands

Concentrate the sperm for artificial insemination

Hypospermia (less than 0.5 ml)

- Procedural causes
- Clinical factors
 - without sperm, pH < 7.4
 - With sperm, pH < 7.4
 - Seminal Vesicle (↓ fructose)
 - With sperm, pH > 7.4
 - Accessory sex gland impairment, addiction, drug
 - Without sperm, pH > 7.4
 - Hypoandrogenism

The Significance of Semen pH

The balance between the alkaline secretion of the seminal vesicles and the acidic prostatic secretion determines the semen pH. The importance of assessing the semen pH and its physiological reference range has been a matter of intense debate. However, WHO 2010 sets the lower reference value of the pH of liquefied semen at 7.2.

In CBAVD, the semen pH is characteristically lower (pH 6.8) because of the absence of the seminal vesicles' alkaline secretion. In these cases the scanty seminal plasma is formed mainly from the relative acidic prostatic secretion.

Macroscopic Analysis

- Appearance
 - Turbid (Normal)
 - Opaque (debris or WBC)
 - Translucent (poor sperm quantity)
- Coagulation & liquefaction
 - Lack of coagulation (gel-like clots)
 - Seminal vesicles
 - Lack of liquefaction
 - Prostate gland

- Color & Odor
 - Whitish- grey, opalescent fluid
 - Reddish (RBC) Hematospermia
 - Drug may change color
- Viscosity
 - More than 60 mm is abnormal
 - ↑viscosity → ↓sperm motility

Microscopic Analysis

Leukocytes:

- Leukocytospermia (>5 wbc/hpf or >1 mil./ml)

- Produce ROS, cytotoxic cytokines, motility, agglutination

- Pyospermia (high amount of wbc)

- Antibiotic treatment

Erythrocytes:

- Hematospermia

 - Inflammation, neoplasms, vascular abnormality,..

 - Urological consultation

Epithelial cell:

- Collection way by coitus interrupts,..

Microorganism:

- Infection or contamination

Cells of spermatogenic origin

- Acute distress, fever, radiation , cytotoxic drugs

Sperm concentration

- Azoospermia (no sperm)
 - Obstruction
 - Hormonal insufficiency
 - Congenital
 - Drugs
 - Immunological
 - Idiopathic
- Oligozoospermia (<10 mil/ml)
 - Procedural Causes
 - Varicocele
 - Thermal stress
 - Behavioral
- Polyzoospermai (>250 mil/ml)
 - Long abstinence
- **Recommendation:** Genetic screening for Y-deletions in azo and oligozoospermia prior to TESE for ICSI

Sperm motility

– Asthenozoospermia

- Less than 32% forward progression within 60 min. of ejaculation
- Procedural causes
 - Psychological conditions (sexual stimulation)
 - Physiological conditions (loss of initial semen fraction, long periods of abstinence)
 - Methods of semen collection
 - Collection container
 - Etc.
- Astheno. With ↑ wbc and seminal debris (infection)

– Necrozoospermia

- All sperm are immotile

Sperm morphology

– Teratozoospermia

- Fever
- Varicocele
- Allergic reactions
- Therapeutic factors
- Congenital
 - Microdeletion in the AZF region of Y
- Stress
 - Physical
 - Psychological

Sperm agglutination

- Sperm clumping into aggregates
- Non-specific
 - Sperm adhere to other cells
 - infection
- Site-specific
 - Sperm adhere to each other
 - Immunological factor

The possible transmission of **HBV, HCV , RPR** and **HIV** viruses from mother to fetus supports a recommendation for the generalized screening of all infertile couples.

VDRL

Serological screening for *T. pallidum* is still recommended considering the extremely high morbidity associated with congenital infection .Serological tests should be obtained initially and need not be repeated unless clinically indicated .

Screening for HBV Before ART

- Approximately 1.5 million people in Iran are living with hepatitis B virus (HBV) infection (mild to moderate prevalence according to WHO classification).
- The HBV infection prevalence in Iran is estimated to be 2.14% .
- The HBV infection rate in Iranian men and women is estimated to be 2.55% and 2.03% .

Screening for HBV before ART is necessary, as **transplacental transmission** due to leakage can occur, for example during a threatened abortion.

Transmission at birth is more likely if the mother is hepatitis B or HBsAg positive. Knowledge of the couple's serostatus allows immunoprophylactic measures to be taken; first to reduce the risk of virus transmission to the partner or fetus, and second precautions need to be taken against cross-contamination during sample handling and embryo cryo storage (Cryostorage of embryo). It also enables couples to make an informed decision regarding whether to pursue treatment.

Vertical transmission may occur from a HBV seropositive mother as a consequence of **intrauterine exposure, transplacental transmission, or breastfeeding.**

Interestingly, HBV infection has also been detected in the newborns of HBV seronegative mothers, because HBV DNA has been detected in semen and spermatozoa ,and it is inferred that **father neonate transmission** of HBV might be possible .

In male HBV patients, sperm washing is not necessary to prevent sexual transmission risk unless the female partner has not been effectively vaccinated. However, in ART, sperm washing is routinely used Theoretically, this could prevent introduction of the HBV into the oocyte in the case of ICSI.

Infections in the male partner Chronic hepatitis B To prevent transmission of HBV to the female partner, and thus to prevent perinatal transmission to the child, the woman should be vaccinated . Once the anti-HBsAg titre has risen above 10 mIU/ml she is protected and IVF can be carried out.

HCV

- Continuous screening of couples attending IVF clinics is necessary.
- Low frequency of hepatitis B and C infections in infertile couples, but
- The low awareness of patients and staff
- Less support from facilities such as health insurance
- Greater likelihood of transferring these infections from patients to other patients and personnel and even from mother to fetus.

HCV

- It would appear that the prevalence of HCV infection in the Iranian population is 0.16.
- Chronic infection with HCV is a major cause of cirrhosis and hepatocellular carcinoma.
- The infants of HCV-positive mothers were more likely to be of low birth weight, small for gestational age, need assisted ventilation, or require neonatal intensive care. HCV-positive mothers with excess weight gain had an increased risk of gestational diabetes.
- Vertical transmission of the hepatitis C virus does occur, but it appears to be much less efficient than for hepatitis B.
- Early diagnosis of infection in newborns requires HCV-RNA testing.

Chronic hepatitis C

- A vaccine for HCV is not available but as the sexual transmission rate of HCV is small IVF is not contraindicated .
- The couple should, however, be informed and should sign a document with the information given. When a chronic active hepatitis is concerned, it is recommended that the male partner be treated before starting an IVF procedure .
- Since HCV is a RNA virus lacking reverse transcriptase activity, it is impossible that the viral RNA can integrate into the genome of the host .

HIV

- Every couple in which the future parent is HIV-positive should be carefully counselled.
- If assisted fertilization is started, it will be of great importance to reduce the transmission risk of HIV to the female partner and the future child.
- It has been shown that anti-retroviral treatment results in a significant drop of the viral load in semen. However, replication-competent provirus DNA has been demonstrated in the seminal cells of HIV-1 infected men after highly active anti-retroviral therapy.
- It is strongly recommended that the spermatozoa be isolated and that contamination with white blood cells be avoided as seminal white blood cells are host cells for HIV in the semen of HIV-infected men .

HIV

- Some data have been published on the removal of cell-associated HIV from the semen of HIV-seropositive men by gradient centrifugation and repeated washing, followed by a swim-up procedure .
- Controversy surrounds the question of whether spermatozoa and sperm cells can be infected with HIV. A number of groups have presented data suggesting that HIV attaches to and infects spermatozoa .
- The first group described the transfer of HIV-1 by the spermatozoon into the oocyte. These findings raised new questions concerning not only the problem of HIV transmission to children, but also transmission to women.

