

SEMEN ANALYSIS AND ASSESSMENT

فحص وتقييم السائل المنوي

Semen: a white thick creamy fluid produced by the male reproductive organs, composed of spermatozoa and semi-viscous fluid (seminal plasma).

- A. **Cellular part:** sperms 95% as well as few epithelial cells and leucocytes.
- B. **Liquid part (seminal plasma):** 98% accessory sex glands secretion as well as epididymal and vas deference secretion.

Semen analysis and assessment mean an evaluation of the ejaculate for breeding soundness or diagnosis and treatment of the infertility problems, it includes macroscopic and microscopic examination.

Macroscopic examination:

Consist of directly observing and examining the ejaculate in the tube to rule out any defect due to volume, color, odor, and viscosity.

1. **Volume:** This is a parameter that depends on the function of the semen vesicle and other accessory sex glands, plus other factors such as age, species, training... etc., and varies from 1-8 ml. and most bulls provide about 6 ml ejaculate.
2. **Color and appearance (opacity):** Depends on the number of spermatozoa:

color evaluation of semen

	Semen color	semen quality
1	Milky or creamy	good
2	Watery	poor
3	Yellow	contaminated with urine
4	Pink-red color	presence of blood

3. **Odor:** in good condition, semen has a smell similar to fresh milk. The smell of urine indicates that the semen is contaminated with it. If the smell is unpleasant, some kind of disease is suspected in the testicles or elsewhere in the reproductive tract.
4. **pH:** determined using a paper strip between the range (6.4-6.9). Above (6.9) is indicated of low semen quality.

Microscopic examination:

Completed under a bright field microscope to analyze the mass motility, individual motility, morphology, and concentration (numbers) of the sperms.

1. Masal motility:

It is determined by:

* placing a drop of semen on a slide.

* Observing it under a bright field microscope with little magnification.

* The presence of waves and eddies throughout the whole drop is evaluated and is given a classification from (0 to +++), as below:

Masal motility evaluation

	Motility description	Quality	Mark
1	Many dark waves moving rapidly	Very good	+++
2	Less dark waves with moderate movement	Good	++
3	Clear waves with very slight movement	Normal	+
4	No waves and the spermatozoa are immobile	Poor	0

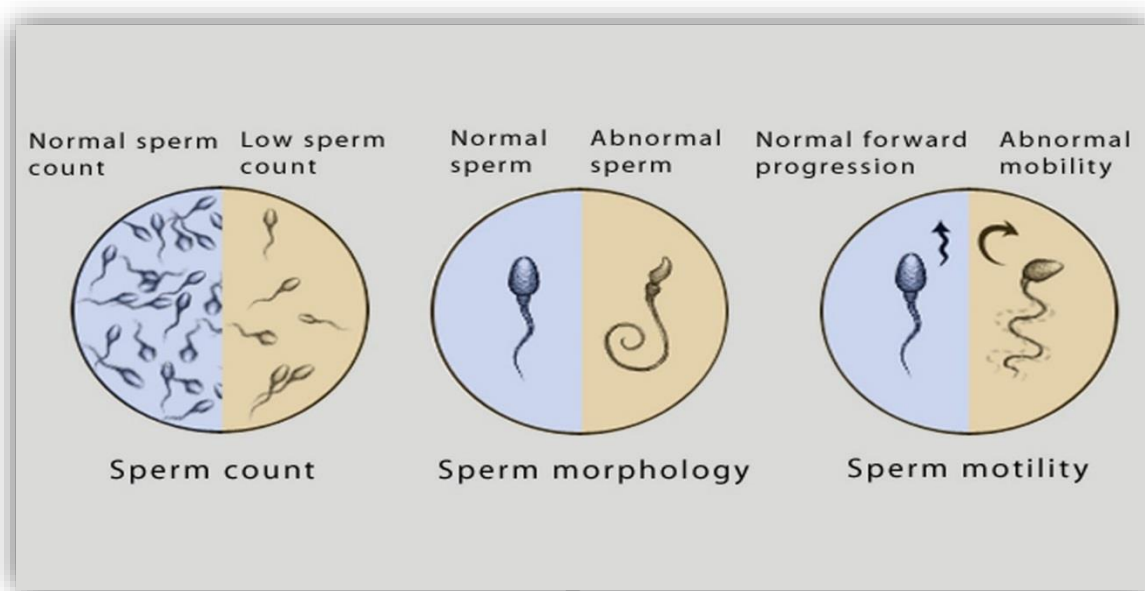
2. Individual motility:

This parameter can be analyzed more objectively with automated systems such as (CASA) system. However, it is also determined by a bright-field microscope examination by placing a drop of semen on the slide may be diluted with saline or sodium citrate (0.9 %).

A coverslip is then placed over it and observation is performed under a bright field microscope with maximum magnification. The semen is classified according to the type of movement of the individual sperm, as follows:

Individual motility examination

	Individual motility of sperms	Evaluation
1	≤ 70 progressive individual motilities	Very good
2	50 – 69 individual motilities	Good
3	30 – 49 individual motilities	Normal
4	≥ 29 individual motilities	Poor



Microscopic examinations of semen

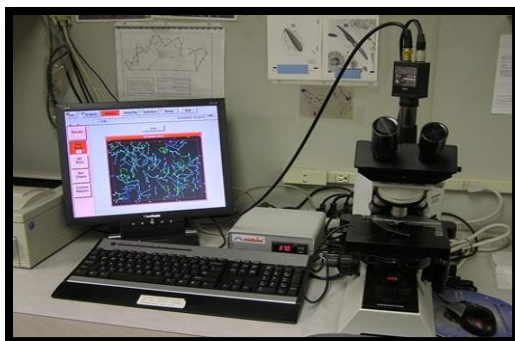
3. Sperm viability, acrosome, and morphological abnormality:

Determined by microscopic observation of a smear of semen subjected to special staining fluids, usually (**Eosin-Nigrosine**). The method involves:

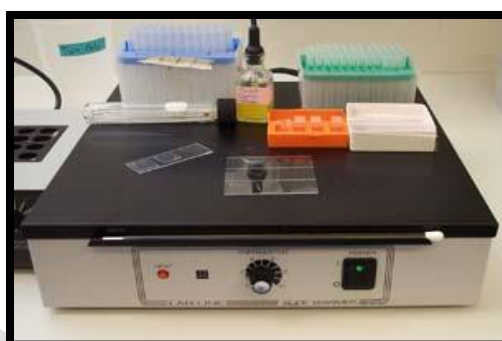
1. Placing a drop of approximately 10 microliters of pure semen on a prepared slide (cleaned and degreased at a temperature of 36 – 37 c° on the heated plate).
2. The drop of semen mixed gently with a drop of eosin-nigrosine, which must be at the same temperature as the semen (using the pipette tip).
3. Another slide, prepared as above, is supported on the edge of the drop until the liquid begins to spread over the slide by capillary action. It is then spread evenly, firmly, and softly.
4. The assessment of sperm viability, acrosome, and morphological abnormality is performed under a bright field microscope at 100x magnification.
5. A total of 200 sperms are counted and the percentage of non-viable cells, abnormal acrosomes, and malformations of the sperm(colored) are calculated
6. The minimum value is 70% normal acrosomes.
7. The abnormalities observed are classified as primary or secondary:

- Primary malformations are by definition, those that originate within the testes during spermatogenesis.
- Secondary malformations are originating within the epididymis.

In general, the maximum number of head abnormalities allowed in the ejaculate is between 15-20%. Acrosome and tail abnormalities are acceptable up to 25%.



Computer assay semen analyzer



Heated plate

4. Concentration:

Number of spermatozoa/ml and is calculated by counting the sperm to the dilution and volume, in a counting chamber (Burcker or similar) under a bright field microscope. Another way to determine this semen parameter is an examination with an automatic system such as the CASA system or using a spectrophotometer.



Normal and abnormal sperm morphology

Characteristics values of bovine semen for normal fertility

	Parameter	Percentage
1	Progressive motility	> 50 %
2	Concentration	> 500 million \ ml
3	Sperm vitality	> 50 %
4	Abnormal head	< 20 % (normally 8-12 %)
5	Proximal cytoplasmic droplets	< 4 %
6	Distal cytoplasmic droplets	< 4 %
7	Abnormal middle pieces	< 15 %
8	Double tails	< 4 %.
9	Crooked tails	< 3 %