**Sperm morphology**

1. Dead – live sperm.
2. Sperm abnormalities or defects.

Many different staining techniques have been devised for examining sperm morphology:

-These stains stained field or background

1. Indian ink.
2. Opal blue stain.
3. Toluidine blue stain
4. Methylene blue solution.
5. Nigrosin stain.
6. Fast green stain.

-These stains stained the dead sperms

1. Eosin
2. Erythrocin
3. Rose Bengal

An **eosin** [**- nigrosin stain**](http://www.vivo.colostate.edu/hbooks/pathphys/reprod/semeneval/semenstains.html) is commonly used because it is **effective, simple** and, in addition to **allowing sperm to be readily visualized**, it is also-called "live-dead" stain, allowing one to assess membrane integrity at the same time as morphology.

**Eosin** [**- nigrosin stain**](http://www.vivo.colostate.edu/hbooks/pathphys/reprod/semeneval/semenstains.html)

**1-**Eosin 1%

Eosin powder 1gm

Sod.citrate 2.9gm

Double distilled water 100ml

**2-**Nigrosin 5%

Nigrosin powder 5gm

Sod.citrate 2.9gm

Double distilled water 100ml (mix at 60 - 65◦c and make double filtration through quality filter paper)

- Normal live sperm exclude the eosin stain and appear white in color, whereas "dead" sperm take up eosin and appear pinkish in color.

* Eosin - mark dead cells
* Nigrosin - blackground

The technique for preparing an eosin-nigrosin-stained slide is as follows:

* Have microscope slides and nigrosin-eosin stain prewarmed to body temperature.
* Pipet a drop of stain onto the end of a slide, then pipet a small droplet of semen next to the stain.
* Place the edge of another slide into the drops of stain and semen - rock that slide back and forth a few times to mix the sperm and stain, then smear the second slide across the surface of the first.
* Dry the slide rapidly by placing in on a warming plate or waving it back and forth in the air.
* Examine using a bright field microscope (typically using a 1000X (oil)

Count 100 – 200 cells and, in a practice situation, you can just differentiate normal from abnormal cells.

Sperm abnormalities or defects

The abnormal spermatozoa classified as:-

1 – Primary abnormalities: - arise from defects in spermatogenesis inside testes; this type affected the animal fertility (double forms, knobbed sperm defects, decapitated sperm, diadem defect, pear-shaped heads, narrow at the base, small abnormal heads and free abnormal heads).

2- Secondary abnormalities:-

Occurs in epididymis related with the degree of spermatozoa maturation (protoplasmic droplets). this type not affected fertility.

3- Tertiary abnormalities:-

Occurs in any part of mature spermatozoa outside of testes and epididymis, as after ejaculation during handling of the semen such as time of collection, smear, dilution of semen. (free heads, detached acrosome, bent or coiled tail, terminally coiled tail).

Wrinkled acrosome –((القلنسوه المجعده this may reflect a nuclear problem which prevents zona attachment by the sperm cell. It is a rare condition.

Pyriform and tapered heads – (الرأس المغزلي والمدبب) the nuclear material is poorly distributed. The defect may be subtle.

Giant or small heads - This nuclear problem. If the head is twice normal size the cell is a giant cell.

Diadem defect - With this you see invaginations in the nucleus, mostly by the post nuclear cap. The pit lacks DNA. The condition may be associated with stress in bulls and may come and go as stress changes.

Dense proximal prtoplasmic droplets – (القطيره الهيوليه العليا) this arises in the epididymis and indicates maturation problem.

Dag defect - This is a sterilizing defect that occurs in the epididymis so is it is actually a secondary abnormality, but it is a major defect. The condition is inherited and the axoneme is disrupted (fibrils and helix). You see split, shattered, or fractured midpiece. The tail may coil and the motility is low.

Coiled mainpiece –(التفاف القطعه الرئيسيه) The mainpiece is coiled within the plasma membrane.

Bent tails –(الذيل الملتف) the bend in the tail may include a droplet which may be in the membrane.

Physiologic (distal )droplet - some consider this a minor defect, but in fact it may be a major defect. These cells do not freeze well because the water in droplet crystalizes and ruptures the cell membrane.

microcephalic الرأس المستدق

القطعة الوسطيه Midpiece

1. الياف وسطيه وعددها اثنان فقط central fibrils
2. الياف داخليه ناعمه وعددها تسعة ازواج 9 double inner fibrils
3. الياف خارجيه خشنه وعددها تسعه فقط 9 coarse outer fibrils
4. المتقدرات التي تحيط بالتراكيب الانفة الذكر Mitochondria
5. الغشاء الخارجي البلازمي plasma membrane

**Terminologies of SA**

* **Azospermia(absence of sperm)**
* **Oligospermia(low sperm count >15 million/ml)**
* **Asthenozoospermia(poor sperm motility)**
* **Necrozoospermia“dead” sperm**
* **Teratozoospermia(abnormal sperm morphology<4% spermatozoa)**
* **Hypospermia – low semen volume < 1.5 ml**
* **Hyperspermia – high semen volume > 6.0 ml**
* **Polyzoospermia – ++ high sperm concentration, >200M/ml**
* **Aspermia – no semen volume**
* **Pyospermia – leukocytes present in semen,**
* **Hematospermia – red blood cell present in semen**