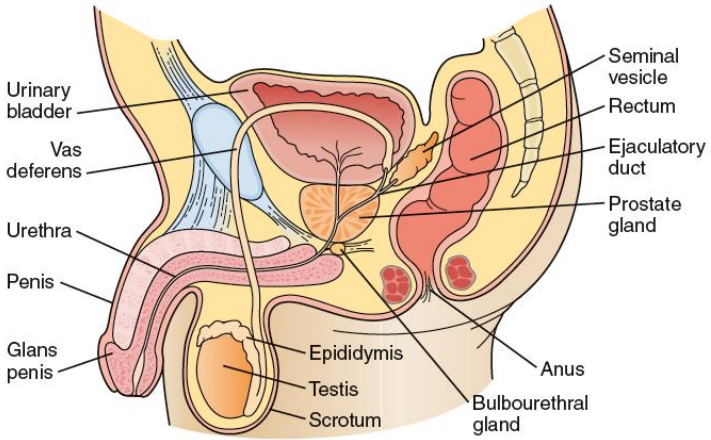
Seminal fluid analysis

Seminal fluid analysis

- What is semen?
- Semen is the fluid formed at ejaculation.
- Made of secretions of all the accessory glands of the male genital tract and testicular sperm component
- Semen quality is maintained by all the accessory glands



Seminal fluid analysis

- Formation
- Semen is formed by the four fractions of the testes
- Epididymis, seinal vesicles, prostate gland and bulbourethral glands
- Each fraction differs and mixing during ejaculation is essential for normal semen

SUMMARY 10-1 Semen Production

Structure	Function
Seminiferous tubules of testes	Spermatogenesis
Epididymis	Sperm maturation
Ductus deferens	Propel sperm to ejaculatory ducts
Seminal vesicles	Provide nutrients for sperm and fluid
Prostate gland	Provide enzymes and proteins for coagulation and liquefaction
Bulbourethral glands	Add alkaline mucus to neutralize prostatic acid and vaginal acidity

Table 10–1 Semen Composition

Spermatozoa	5%
Seminal fluid	60% to 70%
Prostate fluid	20% to 30%
Bulbourethral glands	5%

Seminal fluid analysis

- Purpose of the test
- Semen analysis mainly measures the amount of semen a man produces and determines the number and quality of sperm
- One of the routine tests done to determine male infertiliy
- To determine the effectiveness of vasoctomy
- To assess suitability of semen for artificial insemination
- Forensic and medico legal cases requires semen analysis in sex crimes

Seminal fluid analysis

- Sample collection
- Samples are collected after a period of abstinence of 2 days
  - If not false results may appear
- Complete sample collection after ejaculation is essential
  - If not false results may appear
- Sample should be collected in to pre warmed sterile, non-toxic, wide mouth container
- Should be tested within 1-2 hours
- Many methods are in practice

Seminal fluid analysis

Seminal fluid analysis

Following parameters are routinely checked

Volume, Viscocity, pH, Sperm count, Motility and morphology

Volume

Normal volume is 2-5 ml. A low volume is generally a result of problem with any of the four glands secretions and is associated with infertiligy

Color and viscocity

Normal semen has a gray-white /yellow color and has musty odor.
Viscosity refers to the consistency of the fluid and is direct relation with liquefection.
Normal semen should be drawn in to pipet and forms discrete droplets that are not clumped.
Viscosity can be reported as low (watery) to high (gel like)

pH

Normal pH range is 7.2-8.0 (increases with time)
Abnormal pH indicates abnormal secretion of any of the Four glands

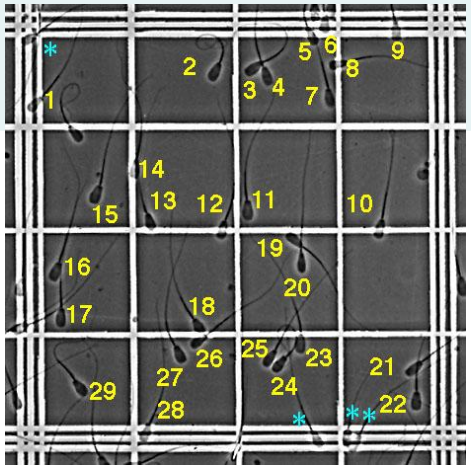
Seminal fluid analysis

Microsocpic examination

Concentration (count)

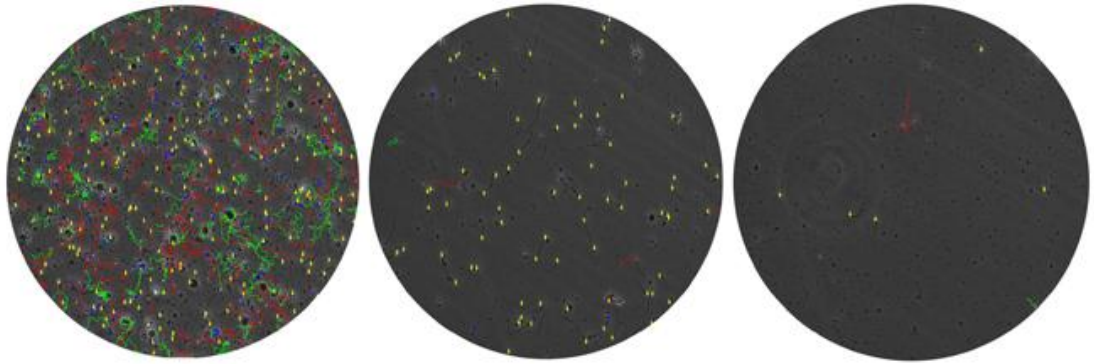
This is the measurement of how many million sperms are present in 1 ml of the sample
Various techniques are in practice
Average sperm concentration is more than 60 million / ml (60-150 mil/ml)
Counts lessthan 20 million are considered sub-fertile

Azoospermia
Total absence of sperms in the sample



Oligozoospermia
Reduced number of sperms (<20 mil/ml) of 5-6 sperms / HPF
Severe Oligospermia
1-2 sperms / HPF

Polyzoospermia
Increased number of spermatozoa i. e in excess of 350 mil/ml



Seminal fluid analysis

Microsocpic examination

Motility – or Mobility

Describes the percentage of sperm which are movint.
Generally 50% of the sperm should be moving
For normal fertilization sperm not only moving but must be capable to move in forward progression (progressive activity)

Rapid progressive	**Slow or sluggish**	**Immobility**
The sperm are moving swiftly across field in a straight line	Straight line mobility but slow **Non-progressive** Sperm not moving in straight line (twitching or shaking)	No movement at all

Live and dead sperms can be differentiated by eosin stain (dead – stained)

Table 10–4 Alternative Sperm Motility Grading Criteria[1]

Progressive motility (PM)	Sperm moving linearly or in a large circle
Nonprogressive motility (NP)	Sperm moving with an absence of progression
Immotility (IM)	No movement

Seminal fluid analysis

Microsocpic examination

Other cells in semen

Leukocytes normally 1-4 / HPF
High number (leukocytospermia) indicates infection

Epithelial cells normally 1-2 /HPF

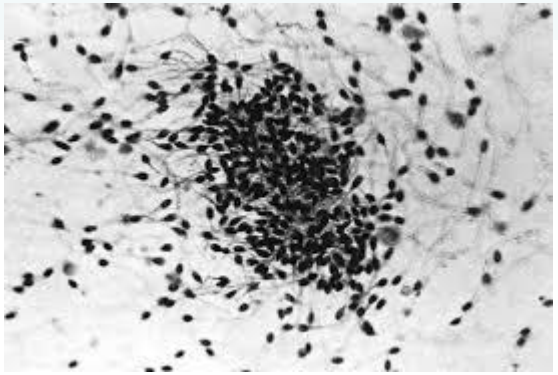
Spermatocytes (immature germ cells) 1-2 / HPF

Erythrocytes 1-2 / HPF. Increased number indicates trauma or infection

Bacteria or protozoan such as trichomonas vaginalis are rare but presence indicates infection

Agglutination or clumping

Presence of agglutination indicates immunological infertility
Presence of anti-sperm antibody



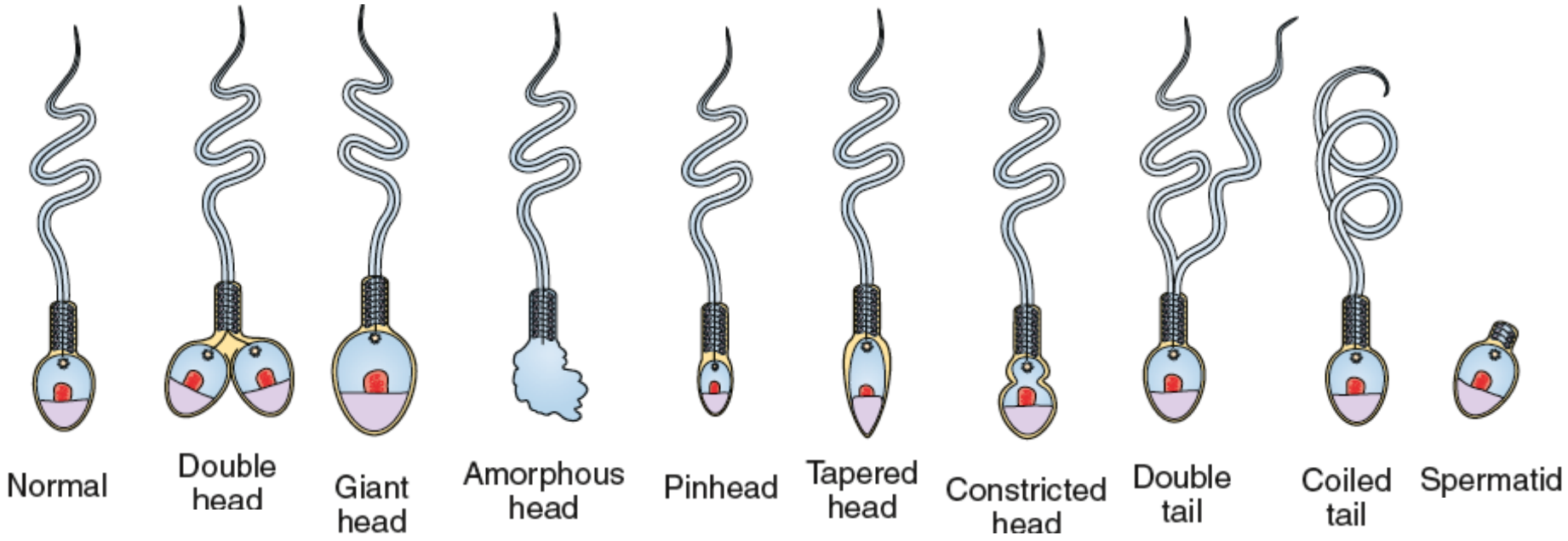
Seminal fluid analysis

Microsocpic examination		
Morphology – describes the shape of the sperm		
At least 30% of the sperm should be normal for fertility Must meet specific sets of criteria to be classified as normal		
Normal spermatozoa should have oval shaped head (4-5.5 µm X 2.5-3 µm)		
The middle piece should be cylindrical (45-50 µm long and 0.5 µm wide) The tail should also be cylindrical (45-50 µm long and 0.5 µm wide)		
Head shape/size Large, small, tapering, pinhead form, amorphours, vacuolated, multiple heads	**Neck & middle piece** Irregular, bent middle piece, thin middle piece (no mitochondria),	**Tail defects** Short, multiple, hairpin, broken, irregular width, coiled tails **Cytoplasmic droplets**

Seminal fluid analysis

Microsocpic examination

Morphology – describes the shape of the sperm



Seminal fluid analysis

Biochemical examination

Fructose

Main energy source for sperm is fructose

If spermatozoa are separated by centrifugation and not provided with fructose cells will die
Tested spectrophotometrically

Other chemicals like zinc, citric acid and acid phosphatase enzyme can also be tested

Seminal Fructose Screening Test[6]

1. Prepare reagent (50 mg resorcinol in 33 mL concentrated HCl diluted to 100 mL with water).
2. Mix 1 mL of semen with 9 mL of reagent.
3. Boil.
4. Observe for orange-red color.

Table 10–6 Reference Semen Chemical Values[1]

Neutral α-glucosidase	≥20 mU/ejaculate
Zinc	≥2.4 µmol/ejaculate
Citric acid	≥52 µmol/ejaculate
Acid phosphatase	≥200 Units/ejaculate

Next class….

- 2nd mid exam