





# University of Sadat City Faculty of Veterinary Medicine

## Department of Animal Medicine and Infectious Diseases

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

قالوا سبحانك لا علم لنا إلا ما علمتنا

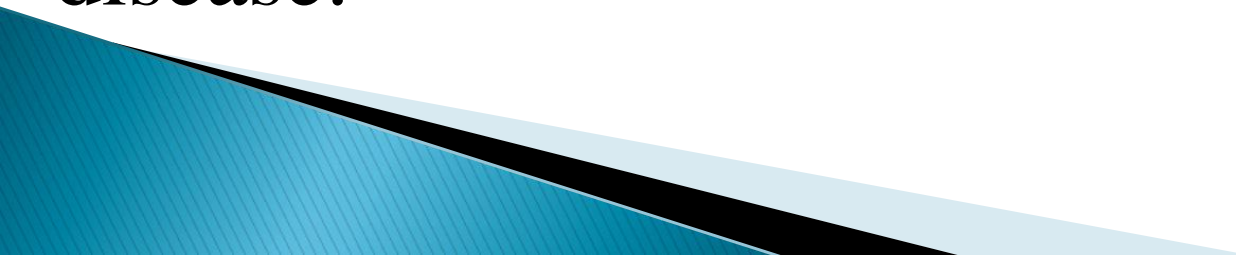
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صدق الله العظيم

سورة البقرة- الآية ٣٢

# Sample collection

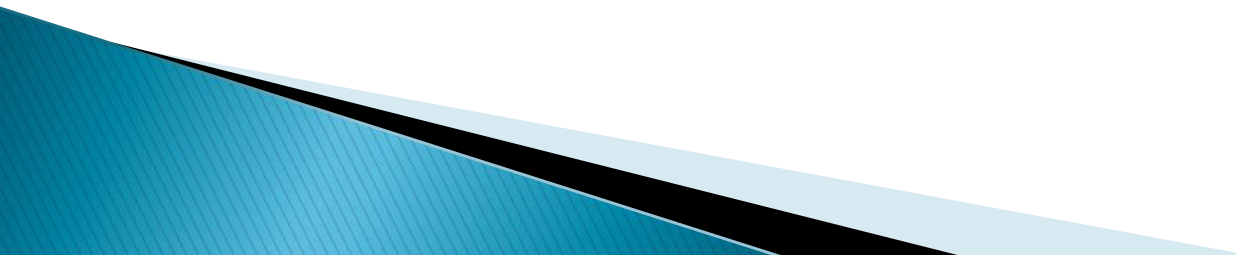
## Selection of animals

- To evaluate the prevalence of a disease or to assess the presence or absence of a disease in an animal population, a survey protocol should be prepared by the epidemiologist insuring that the sampling frame will allow the detection of the disease.
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•When an **exotic** disease, sick animals are selected in order to maximize the chance of finding the disease. **Many animals** should be **sampled instead of only one**. In practice **5 to 10** animals should be selected at **different stages** of the disease.

•If possible animals should be sacrificed to collect fresh post-mortem samples.

**To choose animals for sampling, the following should be considered:**

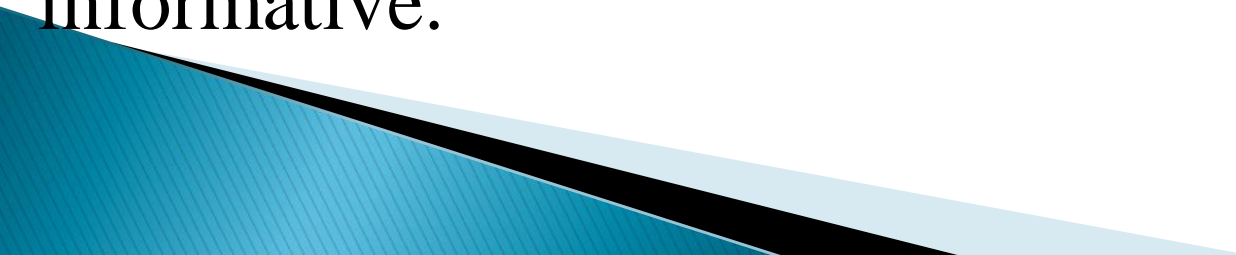
- Animals with **characteristic signs**.
  - **Early** stages of the disease are usually **associated with viremia or bacteremia peaks** and are more suitable for **pathogen identification**.
  - **Late** stages of the disease are usually **associated with higher antibody levels** and are more suitable for **serological diagnosis**.
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- Very young animals may cause a problem

because of maternal antibodies persistence.

- It is preferable to ask the laboratory that will perform the analysis advice about animal selection.

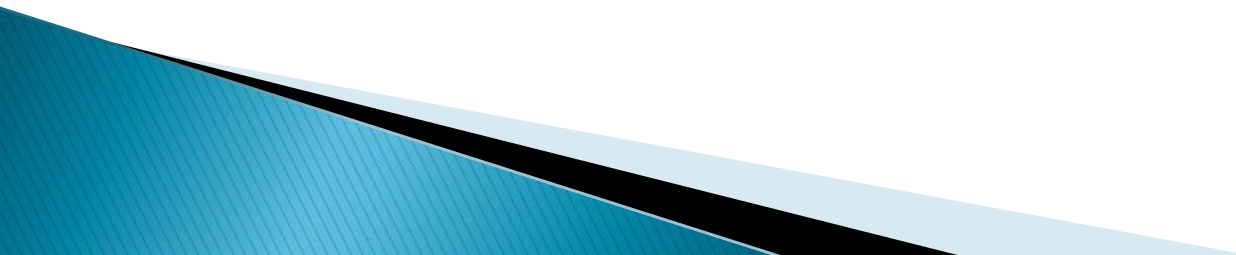
# Post mortem examination

- Post-mortem examination (PM) should be performed on fresh carcasses as decomposition is fast in warm climates.
  - PM performed on partially decomposed animals can lead to false conclusions as many organs take an abnormal aspects a few hours after death, after 24 hours outside PM is no longer informative.
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- All organs and tissues, including external body, should be examined in a systematic way (position color, size, weight, shape, consistency, content, smell, extension of lesions, and aspect of section). The findings should be recorded so as to give the reader a clear mental picture of the lesions seen.

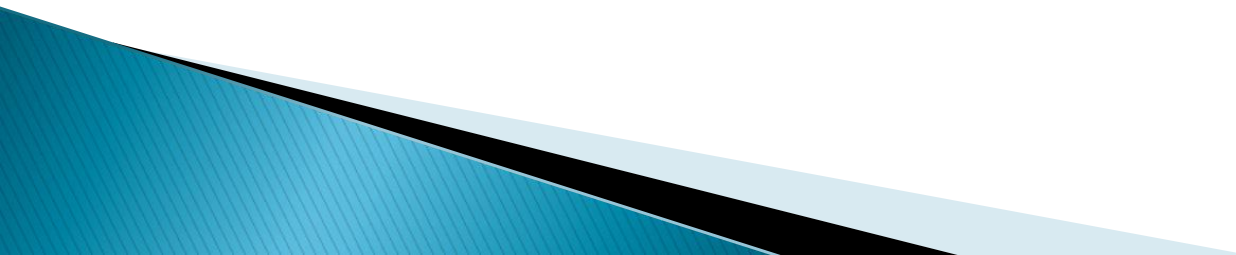
- When performing a post-mortem, **protective clothing** should be **discarded** according to biohazard level of suspected disease and when finished attention paid to **disposal** of the **carcasses**, appropriate **disinfection** of **self** and **equipment**, to avoid further spread of the disease.



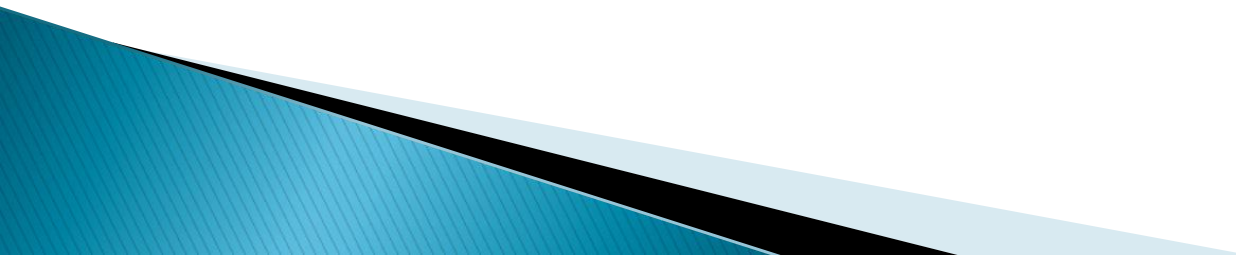
- A full range of diagnostic specimens should be collected. In addition a range of samples for other diseases (both endemic and exotic) that could be considered in the differential diagnosis should be collected.

# Samples collections

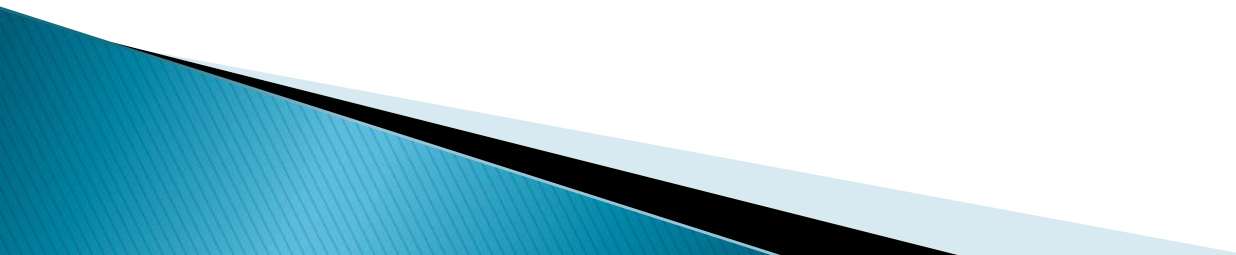
- Collection of **blood** samples
  - Two different kinds of **blood specimens** can be collected for infectious disease .
  - The **whole blood** for pathogen identification.
  - The **serum** for serological diagnosis.
  - Blood samples are collected by **venal puncture** on live animal, as cleanly as possible.
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- Use new needle and syringe for each animal.
  - Dispose needles properly
  - Clean the puncture area with 70% alcohol
  - Handle animals calmly and use appropriate restraint
  - Collect at least 2.5ml of blood , but ideally 10ml for large animals and 4–6ml for small animals.
  - Identify the sample with permanent method and fill the sampling form at the time of sampling.
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# Sites include:

- Brachial.
  - Brachiocephalic.
  - Femoral.
  - Cranial Vena cava in pigs.
  - Mammary veins.
  - Jugular vein
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## Whole blood samples

- For many infectious diseases, the agent can be identified from blood, by detection of antigens or culture and isolation of the agent, or demonstration of DNA.
  - In order to do such examinations the blood needs to be stored properly, with an **anticoagulant**. Different anticoagulants are available in commercial tubes such as: EDTA (**purple tubes**) or Heparin (**green tubes**) which are commonly used for bacteriology or virology.
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EDTA Tube  
K3EDTA

Name:

Pat No.:

LOT 20180219

Exp. Date: 20180219

4ml



EDTA Tube  
K3EDTA

Name:

Pat No.:

LOT 20180423

Exp. Date: 20180423

2ml



EDTA Tube  
K3EDTA

Name:

LOT 20180423

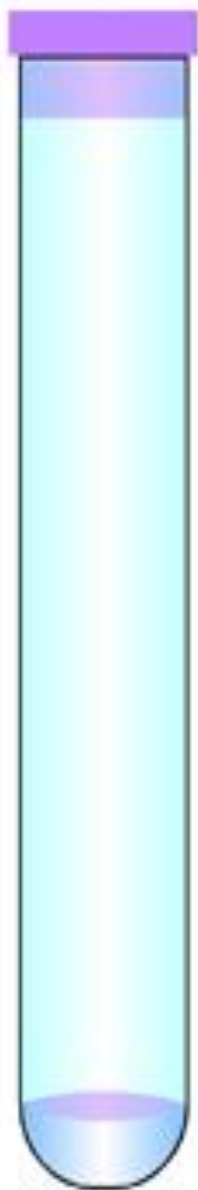
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Lavender-top tube containing EDTA

Tube containing whole blood



Draw blood →



Centrifuge →

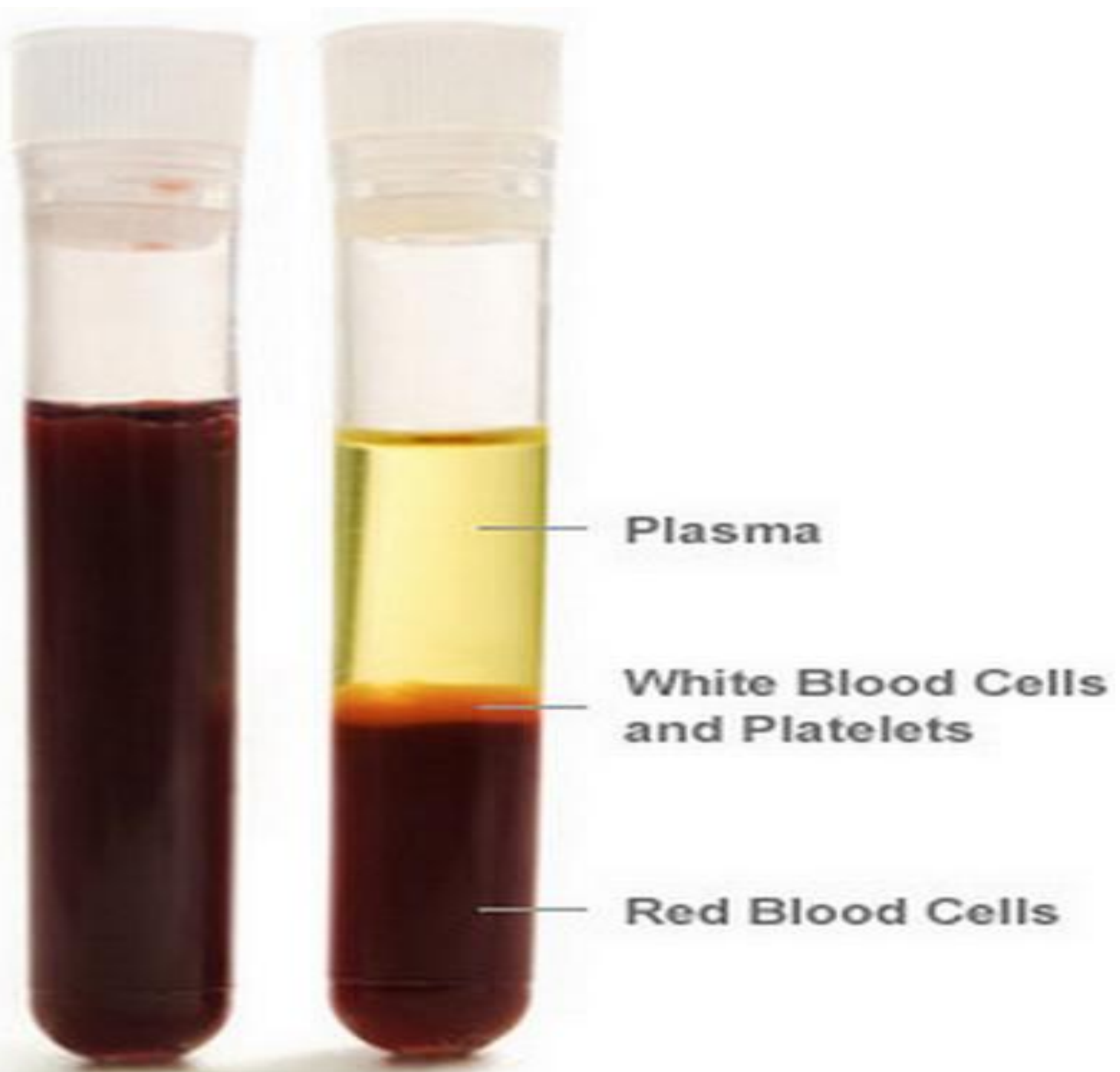


~55%  
Plasma  
volume

< 1%  
Buffy coat  
(white cells and  
platelets)

~45%  
Hematocrit  
(packed red  
cell volume)

100%



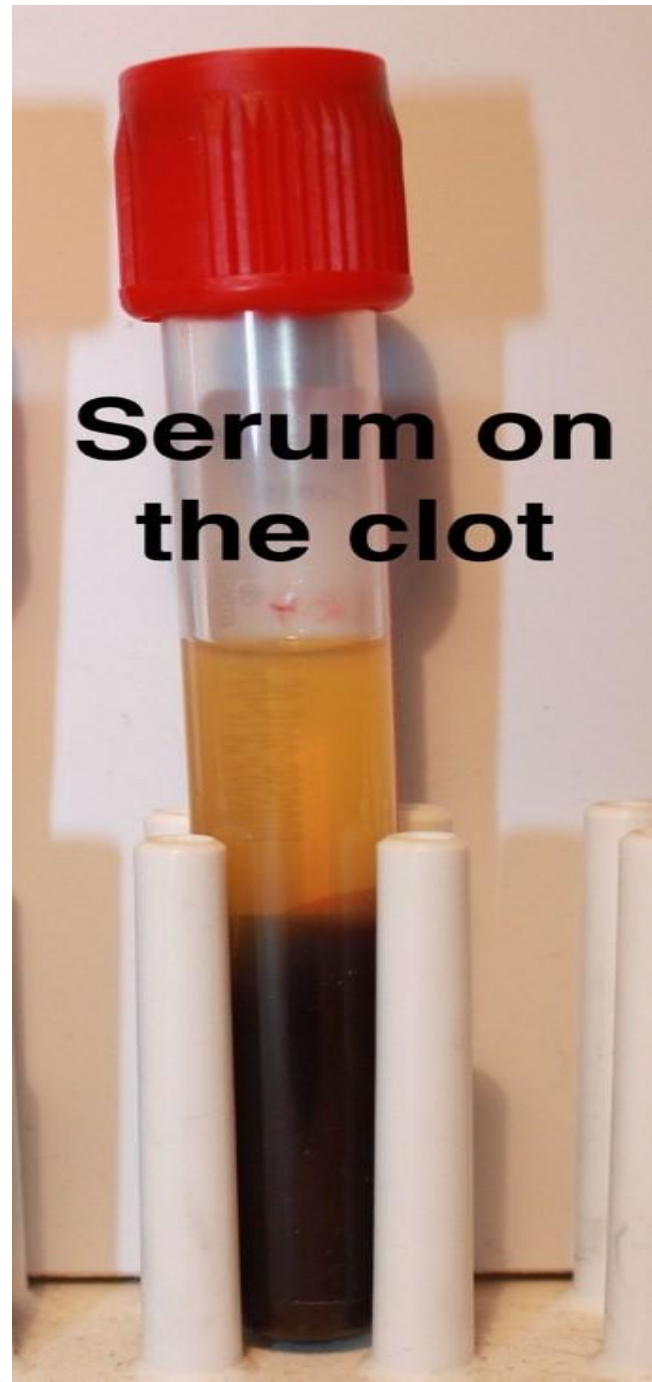
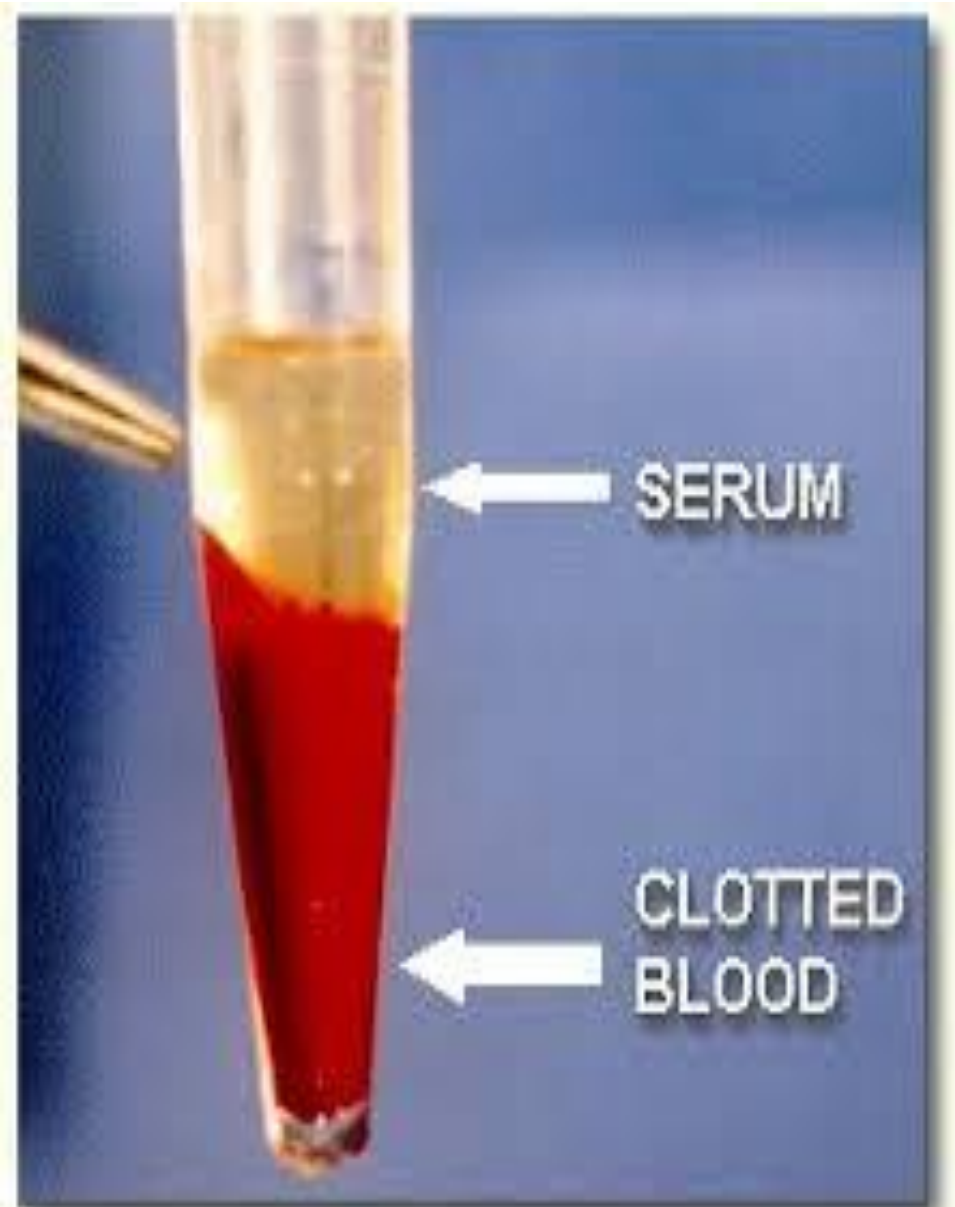
Plasma

White Blood Cells  
and Platelets

Red Blood Cells

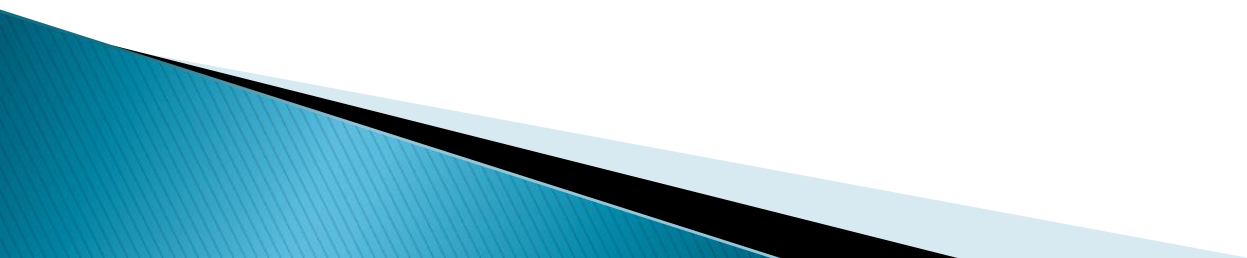
# Blood for serum samples

- Serum samples are used for serological analysis.
- A **positive** serological reaction does **not** necessary means that the animal is **clinically infected**: vaccination, post-infection immunity, persistence of maternal antibodies in young animals, and lack of specificity of the test, are common cause of "**false**" positive.



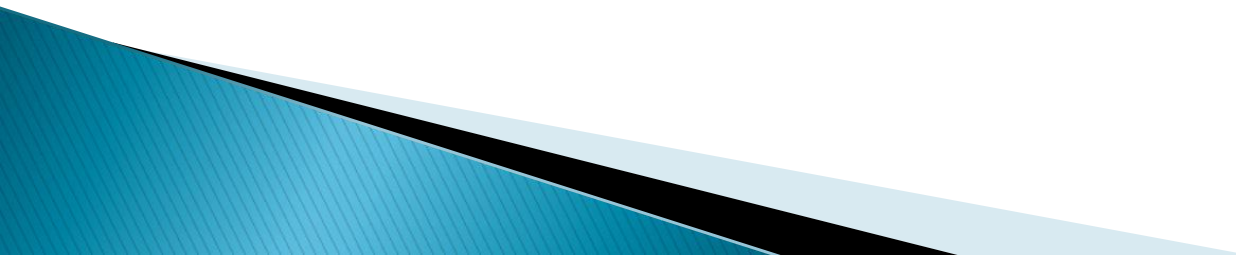
- If a **bacterial** disease is suspected, it is often useful to keep and submit the **clot** to the laboratory for **bacterial culture**.

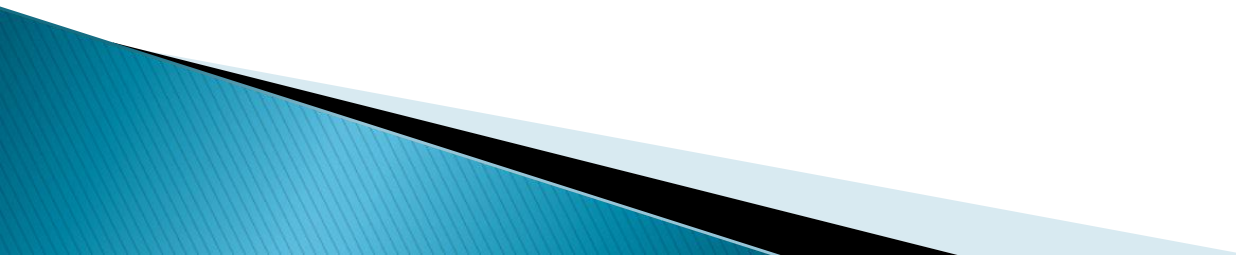
- Hemolysis can interfere with many serological diagnosis tests



- **Plasma** samples can also be used for **serological diagnosis** (blood should be cooled in an ice bath and centrifuged as soon as possible; then the plasma should be separated immediately after centrifuging).

# Collection of Tissue Samples

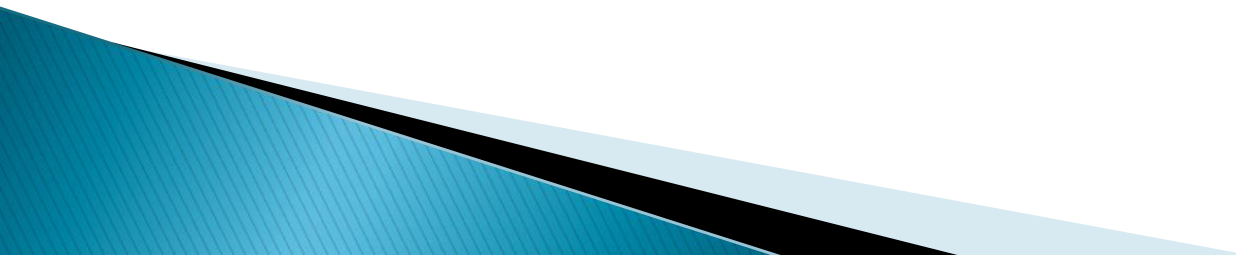
- **Tissues** are collected for histopathology, Parasitology, biochemistry, immunohistochemistry, culture and PCR.
  - For histopathology samples can be preserved by fixation, for all other techniques, fresh samples are required.
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- Organs can be wrapped in absorbent paper to prevent leaking. Care should be taken not to contaminate tissues with other tissue, body fluids or intestinal contents.
  - Sampling boxes should be labeled and the submission forms fill at the time of collection.
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- Commonly used transport medium include:
- 0.04M phosphate buffer (pH 7.6).
- Glycerol-phosphate buffer.
- Samples should be refrigerated at **4C** and sent on **ice**.

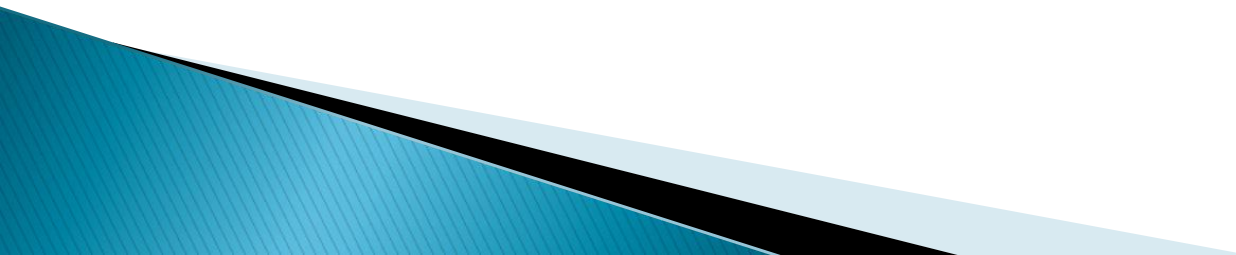
**Freezing** samples is **not** recommended unless immunohistochemistry is used.

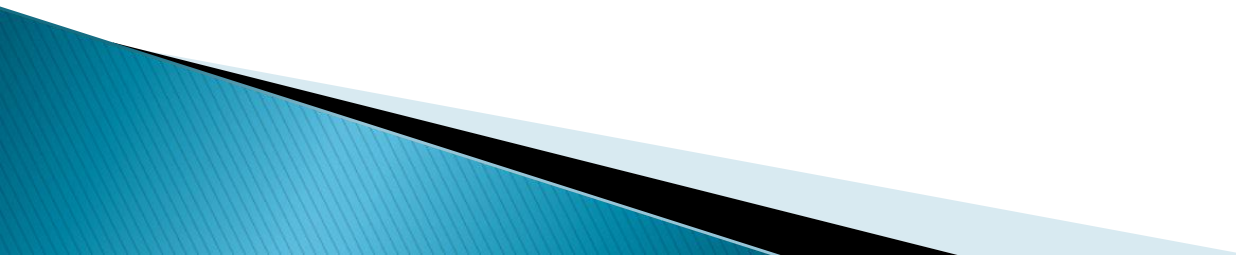


## •Secretions, Excretions and Other Specimens

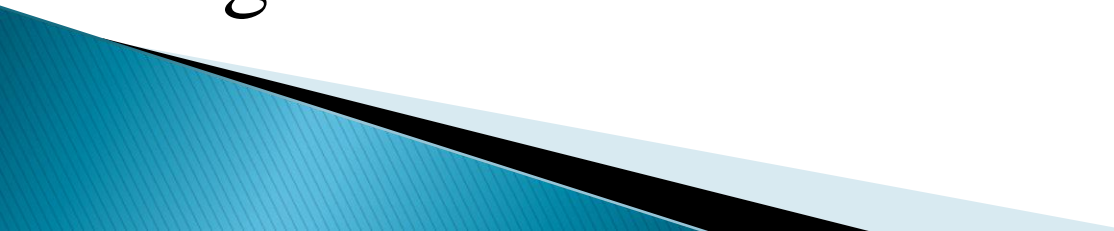
•Part from tissue and blood which are the most commonly used samples; a number of other specimens can be submitted for laboratory diagnosis.

# Feces

- Feces can be sent for parasitological examination, bacteriology or virology.
  - 5 to 10g of feces should be collected from the rectum (or cloacae) or collected as voided. Feces are placed in labeled a sterile container, and submission form is filled.
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- Transport medium may be required for some examination and the laboratory should be contacted first.
  - For **Parasitology**, fecal material should **fill the recipient** or be **completed with sterile water** as air accelerates hatching of the eggs.
  - Feces are best stored and transported at 4° C.
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# Urine

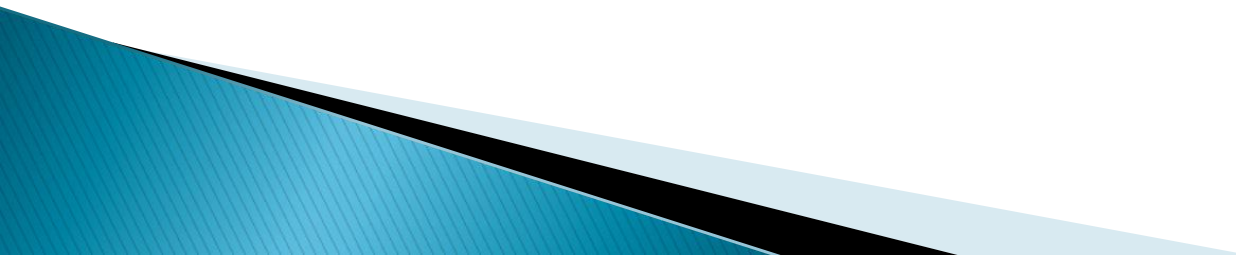
- Although there are not widely used, urine samples can be used for the identification of pathogen, specially using PCR techniques.
  - Urine samples should be collected in screw cap tubes, in amounts of 5-10 ml and kept refrigerated.
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# Skin

- Skin specimen can be sent for bacteriology and virology. In diseases producing vesicular rashes or where lesions are exclusively in the skin, samples are taken from the lesions themselves.

# Pus

- Pus from abscess can be collected with a sterile swab and kept in a sterile universal. Material is preferably **not** collected from the **center** of the lesion. Approximately 3ml is required for bacteriology Material from recently formed abscesses is preferred.

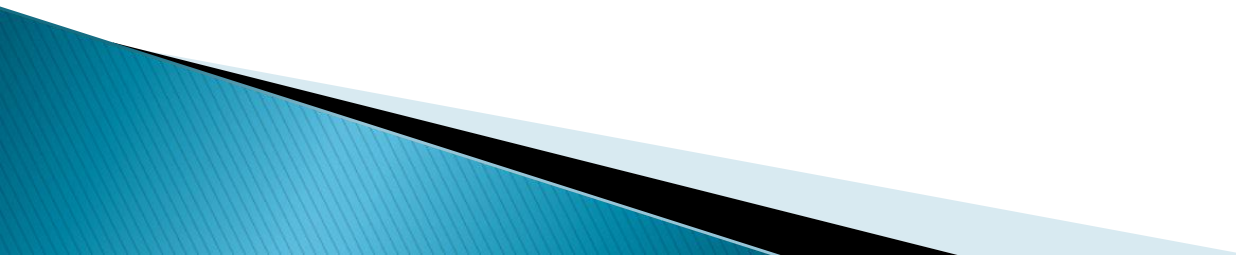


# Eye

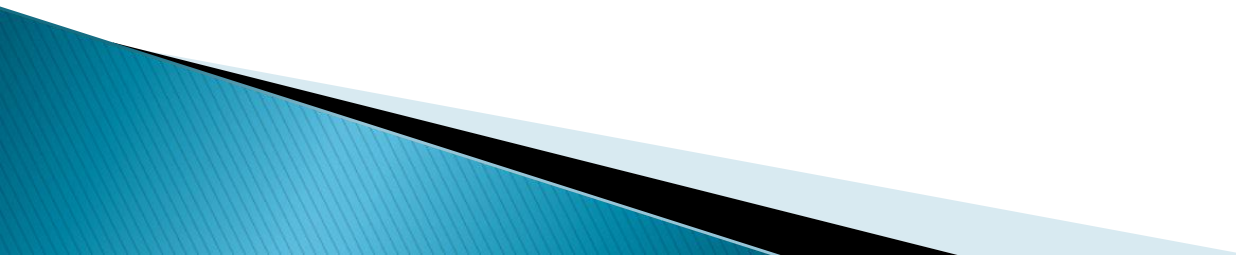
- Eye swabs can be submitted for bacteriology and virology.
- The swab is then broken off into transport medium.
- The handles of metal-handled swabs are useful for this, to ensure that sufficient cells are removed for microscopic examination.



## **Nasal discharge (Saliva, tears)**

- Samples may be taken with dacron, cotton or gauze swabs.
  - They should be allowed to remain in contact with the secretions for up to 1 minute, then placed in transport medium and sent to the laboratory without delay at 4° C.
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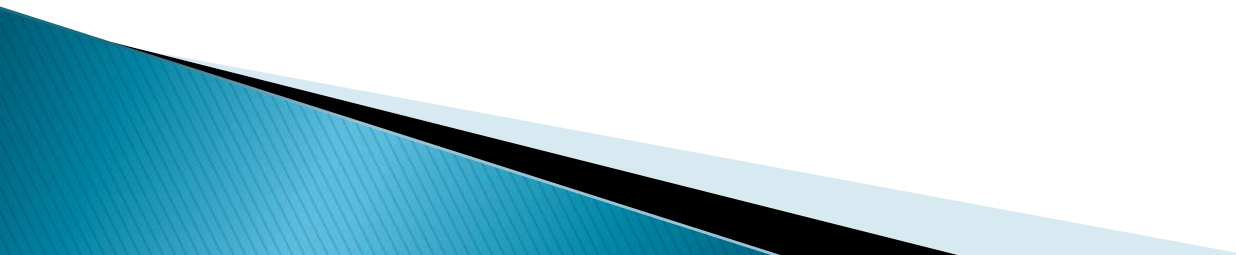
# Milk

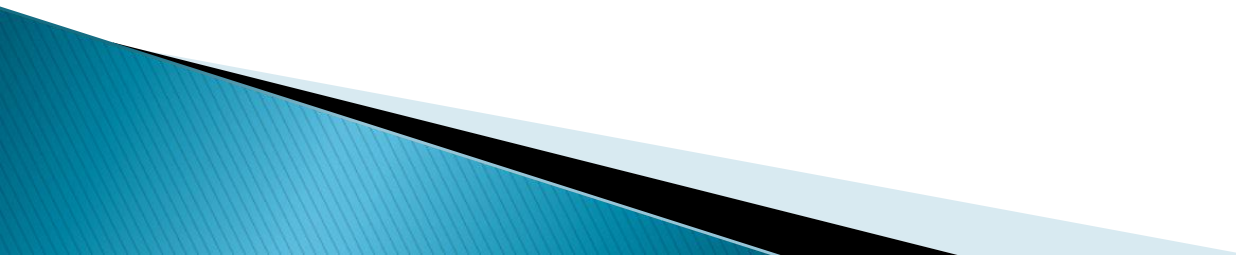
- Samples of milk can be used for serological tests, bacteriology and virology. They should be collected after cleansing and drying the tip of the teat. The **initial stream** of milk should be **discarded**.
  - Milk samples should be submitted in screw top tubes frozen or placed on ice packs.
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- ▶ Honey bees also may be collected
- ▶ Other arthropods collected in sampling boxes and conserved by freezing or in 10% formalin

# Information to be sent with specimens

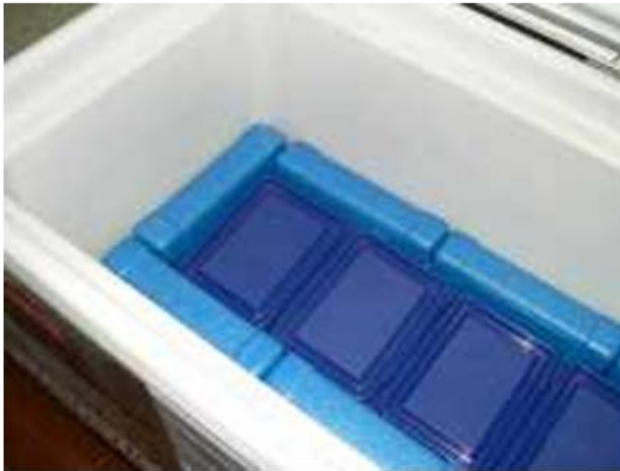
## (Sample labeling)

- Information on the case history should always be sent with the specimens to the laboratory
  - The following information should be recorded:
  - Sender's name and location, telephone/fax, date of submission.
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- Owner's name and location of the animals with telephone/fax if available.
  - Suspected disease(s).
  - List and description of specimens collected, tests required (transport medium used), date of sampling.
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Specific packaging and labeling instruction apply to specimen sent with dry ice (see [WHO/EMC/97.3](#)).



**Samples transfer on Ice**

