

General rules for collection of samples

1. The sample must be as fresh as possible and obtained and preserved in the correct manner.
2. The sample must be representative.
3. Select an animal that correctly represent the diseased condition.
4. An animal in advanced stage of the disease is most desirable.
5. In herd problem, collect specimen from more than one diseased animal.
 - a) Collect samples from one or two recently died animals.
 - b) Collect samples from animals in various stages of the disease.
6. Avoid as much as possible contamination of the specimen with intestinal content, hair or dirt's.
7. Sufficient quantity of material must be provided to permit through examination.
8. The sample must be examined directly after collection.
9. Container must be thoroughly cleaned.
10. Each sample must be labeled and easily identifiable.
11. The clinician should include the following information with the sample:
 - a) Owners name, address.
 - b) Description of animal species, age and sex.
 - c) Duration of the condition, mortality rate, number of animal affected and clinical signs observed.
 - d) Clinical diagnosis and tentative diagnosis submitted.

Types Of Samples

1. Blood Sample

Collection and handling of blood

Venous blood is preferred for most hematological examinations; peripheral samples can be almost as satisfactory for some purposes.

Site of collection

Differ according to the type of the animal species and the amount of blood to be collected. Small amount of blood is generally collected from the ear vein in different animal species.

Cattle	Jugular vein, milk vein, coccygeal vein.
Horse	Jugular vein.
Donkey	Jugular vein.
Buffaloes	Jugular vein.
Sheep & Goat	Jugular vein.
Dog	Cephalic vein, lateral saphenous vein.
Cat	Ear vein, femoral vein.
Rabbit	Marginal ear vein.
Rat and mouse	Tail vein.

Note

1- The blood sample is taken, as cleanly as possible. The skin at the site of vein puncture should be shaved and swabbed with alcohol 70 % and allowed to dry, then the blood taken either by syringe and needle, or by needle and vacutainer tube

2- For samples that are collected with anticoagulant, thorough mixing is necessary as soon as the sample has been taken, using gentle agitation only .

3- It may also be necessary to make a smear of fresh blood on a microscope slide; both thick and thin smears may be prepared.

Serum and Plasma :

Serum or plasma may be collected from whole blood as follows :

Separation of serum :

- 1- Blood left to stand at ambient temperature (but protected from excessive heat or cold) for 1-2 hours until the clot begins to contract .
- 2- The clot can then be ringed round with a sterile rod then place the tube in a refrigerator at 4 ° C for several hours or overnight.
- 3- The sample can be centrifuged at about 1000 rpm for 10-15 minutes .
- 4- Aspirate the serum with a pipette .
- 5- Avoid the tip of the pipette being poked into the clot, or too much suction applied near the surface because fragments may be detached .

Separation of plasma :

- 1- Blood with anticoagulant centrifuged at about 1000 rpm for 10-15 minutes .
- 2- Aspirate the plasma with a pipette .
- 3- Do not disturb the cell layer; otherwise blood cells will be withdrawn .

Notes

Serum and plasma may be stored at 4 ° c in a refrigerator for up to 4 days, or should be placed into deep-freeze at (-15 ° c) to (-20 ° c) until used .

Collection of blood in a syringe:

To prevent hemolysis, certain precautions must be taken:

1. The needle and syringe must be dry for hypotonic moisture will rupture some blood cells.
2. The blood should flow freely into the syringe with minimum use of suction or pumping action on the plunger, because excessive negative pressure will rupture cells and collapse the vein.

3. When transferring blood from a syringe to a container, remove the needle from the syringe, then slowly withdraw the plunger from the barrel and allow the blood to flow gently from the syringe down the side of the container.

2. Milk Sample

The udder must be cleaned by brushing off any dirt's, loose straw and mud that is clinging to the skin

1. Wash your hands and put on new disposable gloves.
2. Using a permanent marker, label a new sample tube with the date, cow ID, and the quarter that the milk will be collected from (RF for right front, LF for left front, RR for right rear, LR for left rear). Keep the sample tube closed until the sample will be collected.
3. Make sure that the udder and teats are clean and dry. Pre-dip the teats with an effective germicidal teat dip and leave the dip on for 30 seconds.
4. Wipe each teat dry with a single-use paper or cloth towel, making sure there is no teat dip left behind on the teat, as it will kill the bacteria in your milk sample.
5. Discard 3 to 4 streams of milk to minimize risk of contamination of the sample with bacteria in the teat canal.
6. Open the sample tube immediately before the sample is taken. Do not let your hands or the teat end come into contact with the inside of the tube, including the lid. Collect milk until the sample tube is $\frac{1}{3}$ to $\frac{1}{2}$ full, holding the tube at an angle to prevent loose dirt or hair from falling into it. Immediately close the tube once filled.
7. Immediately put the sample tube in the refrigerator or on ice. Samples that will not be plated within 24 hours should be frozen. It is best to freeze samples before shipping to the lab.

3- Fecal Samples

Fecal samples must be collected directly from the rectum by back racking in large animals. In small animals, finger covered with sterile cot or by using sterile fecal spoon or by enema (worm water).

1. Should be collected 10 g of freshly voided feces.
2. Feces for parasitology should fill the container "Screw top containers or sterile plastic bags" and be sent to arrive at the laboratory within 24 hours.
3. If transport times are likely to be longer than 24 hours, the sample should be sent on ice or refrigerated to prevent the hatching of parasite eggs.
4. Feces are best stored and transported at 4 ° C.
5. Sterile swabs can be taken from the rectum for bacterial examination.

4- Urine Sample

Methods of collection:

Urine in the urinary bladder is normally sterile, the urethra has microflora. Urine collected from animals via several methods .

a) Manual compression of urinary bladder :

A continuous pressure should be done for several minutes until the sphincters of the urethra relax and urine is expelled. The bladder in small animals is obtained by abdominal palpation, while a gentle continuous pressure in large animals can be applied through rectal palpation

b) Catheterization :

Catheterization used in female large animal .Care must be taken to avoid contamination and traumatic injury. The sterile catheter should be use, utilized sterilized forceps, and wearing sterilized rubber gloves .The first portion of aspirated urine should be discarded. Samples collected by catheterization often contain a few RBCs.

c) Cystocentesis :

Cystocentesis indicated for bacterial culture and used in small animals only when the bladder contains a sufficient volume of urine so that it is readily palpable.

1. The skin should be cleansed and prepared aseptically .
2. The bladder should be held firmly .
3. Use a syringe and needle (16 gauges).

5. Abortion cases :

1. If possible, whole fetus should be submitted ,else fetal abomasal contents, lung, liver and a sample of any gross lesions in or on the fetus should be sent .
2. A piece of affected placenta and two or more cotyledon from aborted animals.
3. Uterine discharge, especially if no placenta is available.
4. If there is Leptospirosis abortion, 20 ml of mid-stream urine from the dam preserved with 1.5 ml of 10% formalin.

6- Nasal discharge, Saliva and Tears :

1. Samples may be taken with cotton or gauze swabs, (wire handles).
2. It may be helpful if the swab is first moistened with transport medium.
3. The swab should be allowed to remain in contact with the secretions for up to 1 minute, then placed in transport medium, such as nutrient broth, and sent to the laboratory without delay at 4 ° C .

7- Eye :

1. A sample from the conjunctiva can be taken by holding the palpebral apart and gently swabbing the surface .
2. The swab is then put into transport medium .
3. Scrapings may also be taken on to a microscope slide .

8- Skin scraping and hair

Skin scraping for diagnosis of mange:

- 1- Scraping of the periphery of the lesion till oozes of blood using dry or moist scalpel (mineral oil or glycerin).
- 2- Skin scraping collected in screw capped bottle, test tubes or Petri dishes.
- 3- Large amounts of hair or skin can digested in 10 % KOH for 12-24 hours, and the sediment can be examined for mites directly after centrifugation

Hair sample for diagnosis of Ring worm:

- 1- Pull a tuft of hair but not cut to obtain the root of the hair.
- 2- Wrap in a paper or put in envelop or in clean test tube.