

Collection, Preservation and Transport of Suitable Material for Laboratory Diagnosis

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Prompt and perfect disease diagnosis is amongst the key factors for control and containment of infectious and other diseases of animals. Usually this depends upon the kind of specimen collected either on clinical examination or at the time of post mortem. It is found that due to improper submission of samples along with the incorrect history may lead to inconclusive diagnosis. Isolation of etiological agent is always confirmatory method for establishing the diagnosis of infectious diseases. Serological and histo-pathological means are also the other methods of confirmation of disease. Therefore, emphasis should always be given on following points before submission of material for confirmation of disease.

- 1) Where ever required blood sample should be collected in the suitable anticoagulant.
- 2) If serum samples are required to be submitted for retrospective diagnosis, always submit the paired samples along with the history of vaccination, if any. One sample at the time of onset of disease and another after recovery or 3-4 weeks after onset of disease.
- 3) All clinical /morbid materials should be accompanied with full history of disease outbreak viz. Morbidity and mortality rates, Clinical signs, duration of disease, species affected, population of the village, treatment given, disease suspected, prevalence of similar disease condition in adjoining area etc. The material of 5 to 6 ailing animals should be collected at the height of body temperature. Samples are collected with all due precautions in which antibiotics may be added where virological examination is desired. No antibiotics are to be added in cases where bacterial isolation is attempted.
- 4) All biological samples should be sent on ice at the earliest after conducting post mortem. Putrified materials are unfit for laboratory examination.

- 5) Materials collected for bacteriological examination should not be kept at subzero temperature (- 20°C) while for virus isolation these are to be stored at -20 to -80 °C. For most of the diseases material can be kept in refrigerator at 4 °C.
- 6) Detailed post mortem report should be attached along with morbid material.

Following specimens should be collected for proper diagnosis.

No.	Name of Disease	Sick/live animals	Dead Animals
1	2	3	4
1	Bacterial Diseases		
A	Haemorrhagic Septicemia	Fixed Smears from blood and throat swelling	Smears from heart blood and Liver. Heart blood in sterile bottle, lymph node and spleen on ice
B	Anthrax	Flame fixed blood smears of cattle and Sheep. From subcutaneous swelling in horse, swine and dogs	Swab of blood from ear vein for cultural examination or piece of ear in saline or without any preservative in sterile bottle on ice duly sealed.
C	Black Quarter	Impression smears from affected muscle tissues and exudates from lesions on ice	Pieces of affected muscles on ice
D	Enterotoxaemia or lamb Dysentery	--	Contents of small intestine with or without chloroform separately on ice, Kidney and urine
E	Brucellosis	Paired serum samples, Vaginal swabs in PBS in separate bottle on ice, Neat semen in sterile vial or semen straws on ice.	Heart blood or abomasal contents of aborted foetus, placenta with two /three cotyledons. Whole foetus on ice if

			small
F	Compylo-bacteriosis	In males – Prepuccial mucus/smegma/washing in specific media In females- Vaginal mucus/lavage.	In females- aborted foetus/placenta/ stomach contents
G	Johne’s Disease	Rectal pinch smears, bowl washing (at least 10 gm preserved in 10% neutral formal saline solution)	Terminal portion of ilium with ilio-caecal valve, mesenteric lymph gland in 10% neutral formal saline solution.
H	Glanders	Exudate from skin and lung lesions in vials on ice. Impression smears from exudates duly fixed	--
I	Tuberculosis	Cough material in sterile container from live animals , sample of milk in sterile container	Smears from lesions fixed by heat, lymph glands, lung lesions in sterile containers for bacterial isolation in 50% buffaered glycerin
J	Leptospirosis	Blood serum, Milk and urine about 20 ml in sterile vials by adding 1 drop of formalin	Pieces of liver, kidney in 10% neutral formal saline solution,
K	Salmonellosis	--	Intestinal swab, heart blood, bile in sterile container on ice
L	Actinomycosis and actinobacillosis	Smears from pus lesions, pus in vial on ice	Formalin preserved materials from affected muscle.
M	Listeriosis	--	Aborted foetus, brain, placenta& all internal organs in sterile vials on ice and 10% neutral formal saline solution
N	Mycoplasmosis /CCPP/CBPP/Coryza	Paired serum samples, swabs from lesions, nasal and vagina in PBS on ice.	Piece of lung preserved in 10%formaline for histopathological examination and on ice
O	Contagious Equine Metritis	Urogenital swabs from mare and stallion, paired serum samples	--
P	Mycotic Infections	Deep Skin Scrappings in sterlie vials for fugal	--

		isolation.	
2	Viral Diseases		
A	PPR	Eye, mouth, nasal, rectal swabs in PBS on Ice, Blood at the height of temperature in anticoagulant, Pre-scapular lymph node biopsy.	Lymphnodes ,spleen,pieces of intestine on ice, all other vital tissues on ice and 10% formalin
B	Foot and Mouth Disease	Vescicular Fluid from unruptured oral vesicles and epithelium from fresh lesions in 50% GPB	Pieces of pancreas, heart and other organs on ice
C	Rabies	--	Half portion of brain in 50% GPB and half in 10% formalin.
D	Pock Diseases	Scabs in sterile container on ice / 50% GPB,	Skin lesions in 10% neutral formal saline separately
E	Infectious Bovine Rhinotracheitis	Paired serum on ice, Swabs from vaginal and nasal lesions from suspected animals. From bulls neat semen sample on ice.	--
F	Classical Swine Fever	Paired serum	Spleen, lymph node and pancreas (10 to 15 G each) in 50% GPB. Pieces of brain, lungs, intestines especially ileo- caecal region and kidney in 10% neutral formal saline. For isolation do not put glycerine and send material on ice.
G	Blue Tongue	Blood at the height of temperature in heparin, paired sera samples	Spleen and lymphnode on ice for virus isolation. All vital organs to be collected in 10% neutral formal saline.

H	Caprine Arthritis Encephalitis	Paired serum	Joint capsule, brain and lungs in 10% formalin and on ice.
I	Equine Influenza	Nasal swabs in VTM on ice and paired serum	Vital organs on ice and in 10% formal saline.
J	Equine Infectious Anaemia	Paired serum	All internal organs in 10% neutral formal saline solution
K	African Horse Sickness	Paired serum sample	Spleen, brain and lungs in 50% buffered Glycerine and 10% Formal Saline separately
L	Ranikhet Disease	Serum samples	Freshly dead/morbid bird on ice, pieces of liver, spleen, trachea, bronchi, lungs in 50% buffered glycerine saline on ice and proventriculus in 10% neutral formal saline solution.
M	Marek's Disease	Paired serum, feather follicles from chest and neck region in transport medium	Portion of peripheral nerve, trachea, ovary, liver, kidney, spleen and skin in 10% neutral formal saline solution.
N	Avian Influenza	Cloacal, nasal Swabs Faecal content in VTM on ice	Dead Bird, All important vital organs in VTM on ice and in 10% neutral formal saline
O	Infectious Bursal Disease	Affected bird, Paired serum	Bursa of fabritious in transport medium, Kidney, spleen and bursa of fabritious in 10% neutral formal saline solution.
P	Lichee Heart Disease	--	Liver, spleen, bursa, kidney, heart on ice and in 10% formalin
<p>Note : It is advisable to send sick birds/ moribund/dead birds for post-mortem examination and collection of suitable materials for diagnosis.</p>			

3	Parasitic Diseases	
A	Theileriosis	Biopsy smears from swollen lymph nodes fixed with methanol, blood smears from each case from ear vein fixed with methanol
B	Babesiosis	Thin blood smears from early stage of disease taken from ear vein fixed with methanol
C	Anaplasmosis	Thin blood smears from early stage of disease taken from ear vein fixed with methanol
D	Trichomoniasis	In females-vaginal and uterine discharge just before and after abortion In males- preputial washing in transport media
E	Trypanosomiasis/surra	Blood in anticoagulant on ice and fixed blood smears.
F	Gastrointestinal parasites	Faecal sample in 10% neutral Formal Saline solution and from dead animals parasites for identification.
G	Lungworm infestations	Faecal sample in 10% neutral Formal Saline solution. Nasal swabs in PBS or normal saline and infected lung tissues in formal saline
H	Ectoparasitic Infestations	Deep skin scrapings in sterile vials.

For diagnosis of Poisoning /toxicity in animals and poultry it is necessary to send brief account of –

- 1) Historical or Circumstantial Evidence.
- 2) Symptomatic evidence.
- 3) Pathological evidence.

For toxicological examination / investigations, stomach / intestinal / ruminal content, liver, kidney, lung, spleen, etc. (Minimum of 300 to 500 Gm each) should be sent on common salt. Water sample (About one litre) should also be sent in leak proof clean bottle, suspected poisonous material, feed, fodder, green fodder requires no preservative.

Tissues for histopathological examination shall also be sent in 10% formal saline.