DIAGNOSTIC POULTRY POST- MORTEM EXAMINATION IN AVIAN MEDICINE

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by

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General in situ lay of organs of a chicken (Source: Corry Brown et al 2012)
1. **Themes in Post-mortem Diagnostics**

1. Clinical presentation diversity and flock history
2. Disease pathogenesis, pathogen virulence, and inflammation
3. Multiple infections, diversity of disease manifestations - atypical presentations
4. Differentials
5. Epidemiology and flock health management
6. Forensic implications
2. SAFETY: WHY IS IT CRITICAL?

- Zoonoses
- To minimise exposure ensure that personal protective equipment (PPE) are worn in routine work. These include:
  - Coveralls; plastic apron; gum boots/Laboratory shoes; Disposable gloves; Respiratory masks; Protective goggles; head gear.
- During autopsy wear a protective visor for maximum facial protection
Full Personal Protective Equipment

- Hair cover (Cap)
- Eye wear (goggles)
- Mask
- Gown
- Apron
- Gloves
- Shoe covers
3. EQUIPMENT FOR POSTMORTEM

- Disinfectants
- Scissors; knifes; forceps; scalpel blades
- Tray (40x 30 cm)
- Personal protective equipment (PPE):
  + Surgical gloves of proper size
  + Rubber gloves- heavy duty
  + Mask(N95)
  + Gown-full body cover
  + Rubber boots
  + Plastic apron
  + Head dress and goggles
EQUIPMENT

- **Sampling containers:**
  - with 10% formalin for histological samples

- Sterile **polyester swabs** with plastic shaft

- Viral transport medium (VTM) for virology samples in cryovials.

- Swabs and carrying medium for bacteriology

- Cool box and ice packs

- Sample packaging for diagnostic specimen transport
EXAMINATION PROCEDURE: STEPS

1. Obtain flock history
2. Examine the bird externally
3. Open the body; observe organs in situ
4. Remove the organs for detailed examination and sampling
5. Examine and sample the organs
6. Consider observations for d/diagnosis and write the report
3. GENERAL EXAMINATION PROCEDURE

- **General body examination**: Plumage; body symmetry; palpate for crepitation, ascites; abdominal tumors.
- **General body condition** - weight: normal, low or high
- **Skin, legs, feet etc.** - Indicate effect of management and environment; fracture and trauma (self or externally inflicted)
- **Inflammation of feet** may indicate: poor litter, wet bedding, chronic Pasteurella multocida infection of the head, fowl pox, ectoparasites
- **Cloaca examination** may give a clue to: enteritis, salpingitis, uraemia, criminal abuse;
GENERAL EXAMINATION

- Examine for ectoparasites: around the head, on the body; under the wings and thighs

- Examine mucous membranes: conjunctiva, oral cavity and cloaca (DD: anaemia; inflammation, hemorrhages, purulent discharges; avitaminosis A);
Carcass is placed on its right side with head pointed towards the examiner and dampened with disinfectant

Infra-orbital sinus is opened and examined for inflammation: Mycoplasma infections, Haemophilus paragallinarum, Newcastle disease, Avian influenza, Infectious laringotracheitis, Infectious bursal disease.
The beak is opened and the oral cavity is opened (the blunts part of the scissors within the cavity)

Incision is extended down the neck to open the oesophagus and the crop

Oral cavity, pharynx, oesophagus and crop are examined for inflammation – fowl pox, ILT, degenerative changes e.g. avitaminosis A
dissection procedure 3

- Larynx and trachea are subsequently opened and examined for inflammation or other pathology: NDV, ILT, syngamus trachea, wet form of fowl pox, etc.

- Thymus is inspected and incised (well developed in young birds (broilers) atrophied in old birds (laying flocks))
The carcass is placed on its back with the legs towards the examiner

The skin is incised transversely behind the xyphoid process and the incision is extended towards both knees. The skin is removed over the pectoral muscles by pulling in a cranial direction.

Parasternal bursa is inspected for: inflammation associated with recumbency (immobile birds); inflammation due to mycoplasma and reovirus infections.
- Pectoral muscles are incised and examined for lesions e.g. Tumours due to marek’s disease, Haemorrhages in septicaemic diseases, deep pectoral necrosis, and IBD variant manifestations

- Legs and hips are bent outwards until each femoral head is dislocated from the acetabulum
Transverse incision is made behind the xyphoid process to open into the thoracic cavity.

Incisions are made on both sides of the thorax up to the brachial region or shoulders to open the thoracic inlet.
Sternum is slightly lifted to examine the abdominal and thoracic cavities in situ for evidence of mycoplasmal, bacteriological or virological infections e.g. swollen organs, haemorrhages, exudates etc. If lesions are present take swabs or tissue samples aseptically for microbiological examination.

Sternum with pectoral muscles is cut and rotated upwards and cranially to expose the abdominal and thoracic cavities.
Examine all air sacs in situ for evidence of inflammation (cloudiness; thickening; hemorrhagic reaction, oedema)

Remove the heart with pericardial sac and incise and examine it for lesions (incisions of the heart are made along the blood circulation...
DISSECTION PROCEDURE 9

- The liver and gall bladder are removed and examined
- The spleen is removed and examined
- Presence of an enlarged liver (with rounded edges) and an enlarged spleen are typical for septicaemia
**DISSECTION PROCEDURE 10**

- make transverse incision cranial to the proventriculus and remove the whole intestinal tract in a caudal direction. Make a transverse incision 1-2 cm cranial to the cloaca and remove the entire intestines

- Examine the serosal surface of the intestines and then open the intestines in a caudal direction starting from the proventriculus. Simultaneously examine the pancreas.
Examine the entire intestines for:

- Haemorrhages
- Oedema
- Contents (ingesta, parasites, foreign bodies)
- Ulcers, neoplasms
- Thickening or thinning of the intestinal wall
Examine the intestinal mucosa for inflammation:

- Necrotic enteritis
- Salmonellosis (typhilitis)
- Newcastle disease; avian Influenza; coccidiosis
- Haemorrhages
- Ulcerations
- Thickening; Parasite infestations
In females: cut the reproductive tract free of its ligaments in a caudal direction and subsequently open the infundibulum, magnum, isthmus, uterus and vagina (these are inspected both from the serosal and mucosal surfaces) – *E coli* often causes purulent inflammation at these sites.
In young birds the Bursa of Fabricius is opened through its cloacal opening and examined for swelling, oedema, haemorrhages.

Gumboro disease often causes swelling, oedema or haemorrhages in the Bursa of Fabricius similar to HPAI.

Tumours such as Leucosis (not Mareks) often occur in the Bursa of Fabricius.
Examine the kidneys for lesions such as atrophy, nephropathies, inflammation, tumours.

Kidneys are often pale in Clostridia infections as a result of toxaemia.

Examine for congestion and haemorrhages in HPAI.

Swelling – visceral gout.
DISSECTION PROCEDURE 17

- Examine the thoracic air sacs for cloudiness, thickening
- Remove the lungs and examine for size, colour, consistency, oedema tumors
- Examine the heart sac for adhesions, lesions, fluid contents
- Examine the heart for lesions (hemorrhages, myocardial dystrophy, neoplasms)
DISSECTION PROCEDURE 18

- Examine the brachial plexus for Mareks disease
- Examine the sciatic nerve bilaterally for Mareks disease by exposing it through a blunt dissection to separate the gracilis muscle
- Check for swelling and loss of striations
Palpate all the joints and open them in case of swellings and asymmetry

Open the knee and hock joints and inspect the tendons and tendon sheaths for haemorrhage, ulcers, exudates.

Joint disease can be caused by:
Staphylococcus aureus, Reoviruses, Mycoplasma synoviae, E. coli, Pasteurella multocida and Salmonella gallinarum / pullorum
Examine young birds for ossification of the ribs and their spinal and costo-chondral junctions. Swellings may occur in avitaminosis D; neoplasia.

Make parallel incisions on the tibial/tarsal bones to examine for TD (Thiamine deficiency) lesions.
DISSECTION PROCEDURE 21

- Remove the brain by gently lifting the cranial bones and by detaching the dura mater
- Inflammatory foci in the brain occur in avian epidemic tremor,
- Encephalomalacia occurs in Vitamin E deficiency (crazy chick disease)
- Tumours can spread to the brain
Lesions observed are summarised and their pathogenesis reviewed.

A differential diagnosis is made based on clinical history, symptoms, lesions and other epidemiological factors narrated by the farmer = what are at least three most probable diseases.

A tentative diagnosis is made and samples taken.

Confirmatory diagnosis is made from results of sample analysis in the laboratory.

Several birds may be examined for an effective flock diagnosis.
SAMPLES

- **Swabs**: oropharyngeal / tracheal, cloacal, ocular
- **Organs**: Trachea, lungs, duodenum, caecal tonsil, brain, liver, spleen and any organ with lesions from fresh carcasses
- **Blood** for serum: sick and recovered birds
- For each sample, n = at least 20 per affected flock
- At least 5 whole fresh carcases should be submitted for autopsy wrapped in double layer polythene bags and accompanied by full case history as seen by the referring clinician
A) Oropharyngeal swabs:
- Swab the oral pharynx and choanae rotating the swab several times
- Transfer the swab aseptically into a cryovial containing 1ml viral transport medium
- Label the sample appropriately.
- Cryovials are wrapped in absorbent material and placed in 50ml plastic tubes which are decontaminated on the outer surface with 70% alcohol
- All samples are placed in a third sealable biohazard container and placed in a cool box packed with ice for shipment.
- Appropriate labelling and markings are made on the packaging for the specimen shipping to the laboratory.
SPECIMEN COLLECTION AND SHIPMENT
B) Cloacal swab:
- Insert the swab into the cloaca through the vent and twist gently round the surface of the cloaca to pick some fecal material and withdraw gently.

C) Conjunctival swab
- Gently rotate the swab in the conjunctiva sac twice
- Ship the swab as described for oral pharyngeal swab

D) Tears
- Collected for viral nucleic acid and mucosal antibody testing
E) Blood sample for serum

- Use gauge 23-25 needle, and 2-5 ml disposable syringe
- Pluck the feathers from the wing web and swab the wing web
- Collect 2ml blood from the wing web and transfer to a 5 ml sterilin tube or a sterile bijoux bottle without an anticoagulant (Patience and experience are vital)
- Decontaminate syringe and needle in a disinfectant and dispose the needle in a sharps container
- Decontaminate the surface of the sample bottle and ship in the same manner as for the swabs with full history from the farmer
F) Organ sampling

- As soon as the carcass is opened and before any opening of the intestinal, aseptically obtain 1-2 cm of organ samples and transfer each organ into a separate sterile tube containing transport medium for viral isolation, bacterial culture and special buffer for nucleic acid detection by PCR technique.

- Obtain a sample and put into neutral buffered formalin solution for histopathology

- Label the samples appropriately
The labeling for contents should include the words:
“UN 3373 Diagnostic Specimens”
EMERGENCY SAMPLE SHIPMENT

Improvise the tripple parking principle:

- The samples, in their collection vials are packed in double layer plastic bags with absorbent tissue wrappers. The bags with samples are assembled in a carton which is then sealed and wrapped in a polythene bag

- Put the sealed carton in a clean outer carton and Label this third container carton appropriately with the name of the sender; consignee and what materials are contained in the carton.

- Transfer the package for shipment to civilian section of the lab and attach a filled sample shipment form with all details entered in a sample submission form.
SAMPLE TRANSPORT

- All samples must be properly labelled and accompanied with a sample submission record containing full historical description including the suspected disease for which laboratory confirmation is being sought.
- Pack the samples in ice to ensure that the shipment temperature are between 2-8°C.
- Faeces and organs: in sterile leak proof boxes.
- Cloacal swabs: transport in sterile cryovials with viral transport medium, separate from the fecal samples.
DISPOSAL OF INFECTED MATERIAL

- Disinfect the carcass and organs while still on the pm tray
- Secure the carcass in two water tight biosafety bags
- All carcasses are wrapped and transported for incineration in a tightly closed litter bucket or tank internally lined with a watertight polythene disposal bag
- Carcasses and all disposable items are incinerated to ashes
- Decontaminate all buckets, tanks or other containers and carriers and any other items used to move the carcasses at the incineration facility
DECONTAMINATION OF EQUIPMENTS AND SURFACES

- Disinfect all equipments and surfaces used, wash thoroughly in running tap water and disinfect a second time
- Send the postmortem kit for autoclaving
- Fumigate the postmortem room appropriately
CONCLUSIONS

- Themes help clarify observations made
- Careful observations and recording help make accurate differential diagnosis
- Samples: well selected and preserved help confirm diagnosis
- Personal safety is essential for personal safety
- PPE be available
- Diagnostic post-mortem key to clinical disease management in avian medicine
THANK YOU
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